5.5.2. *PINCTADA MARGARITIFERA.*

5.5.2.1. Valves.

In Figure 5.82, a is anterior,p, posterior, d is dorsal and v ventral.

IViewed medially, the Inner Nacreous Layer (inl in Figure 5.82), has a bright silvery surface and the Outer Nacreous Layer (onl in Figure 5.82), is a darkened area peripheral on the anterior ventral and posterior edges of the Nacre.

The Hinge (h in Figure 5.82. b) has a ventrally enlarged part which occupies about the 3rd and 4th sevenths of the Hinge going from anterior to posterior..

The Byssal Notch is an almost rectangular indentation on the Antero-dorsal periphery (bn in Figure 5.82.).

The Prismatic Layer occupies the entire lateral surface of the Valve except where lost, (pl in Figure 5.82. c), and the outer periphery of the medial surface (pl in Figure 5.82. a and b). Where the Prismatic Layer has been lost from the lateral surface of the Valve, the lateral surface of the Outer Nacreous Layer can be seen, (onl in Figure 5.82, c). The Growth Processes (gp in Figure 5.82. a and c), are spurs of Prismatic Layer which occur at intervals around the Anterior Ventral and Posterior borders of each Growth Scale. The medial surface of the last laid down Growth Scale is almost entirely exposed medially (pl in Figures 5.82. a and b) and usually, as in this case, it forms the actual Anterior, Ventral and Posterior borders of the Valve.

The Growth Processes from successive Growth Scales come to lie on curved lines (l in Figure 5.82. c) radiating from the umbonal region (u in Figure 5.82. a and c), on the lateral surface of the Valve. In this species the central part of each Growth Process lacks the dark colouration of the rest of the Growth Scale (gp in Figure 5.82. a).

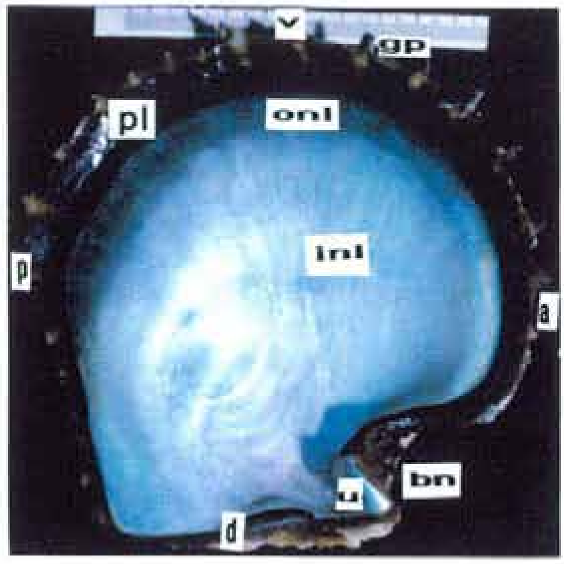
FIGURE 5.82. Valves of *Pinctada margaritifera.* Databars in millimetres.

a. L.M. Medial surface Right Valve.

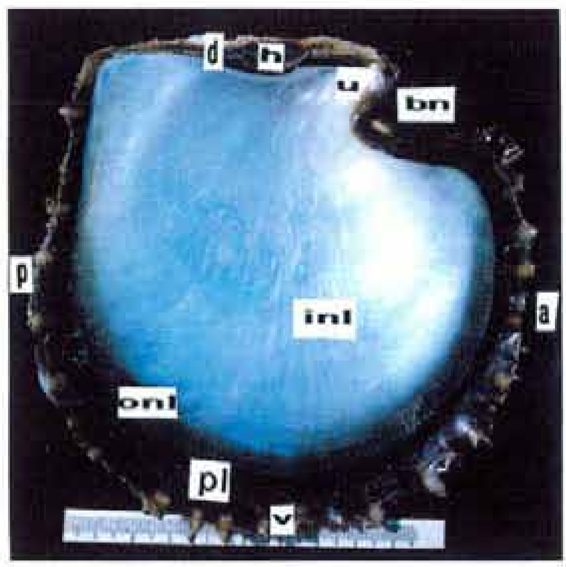
b. L.M. Medial surface Left Valve.

c. L.M. Lateral surface Right Valve.

a



b



c

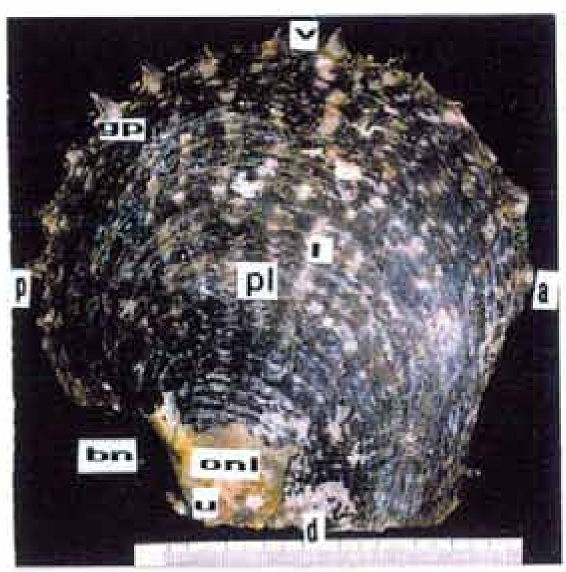


Figure 5.82

5.5.2.2. Hinge.

At the enlarged central part of the Hinge it is composed of parallel fibres about 0.14 μm in diameter. (Figure 5.83. a and b). At high magnification the protein rubber fibres appear to show periodic banding. (Figure 5.83. b).

FIGURE 5.83. *Pinctada margaritifera* – Fibres of Hinge Enlargement.

a. S.E.M. x 4800 Parallel fibrils of hinge protein rubber.

b. S.E.M. x 19000 As for a above at higher magnification.

a

x4800

b

x19000

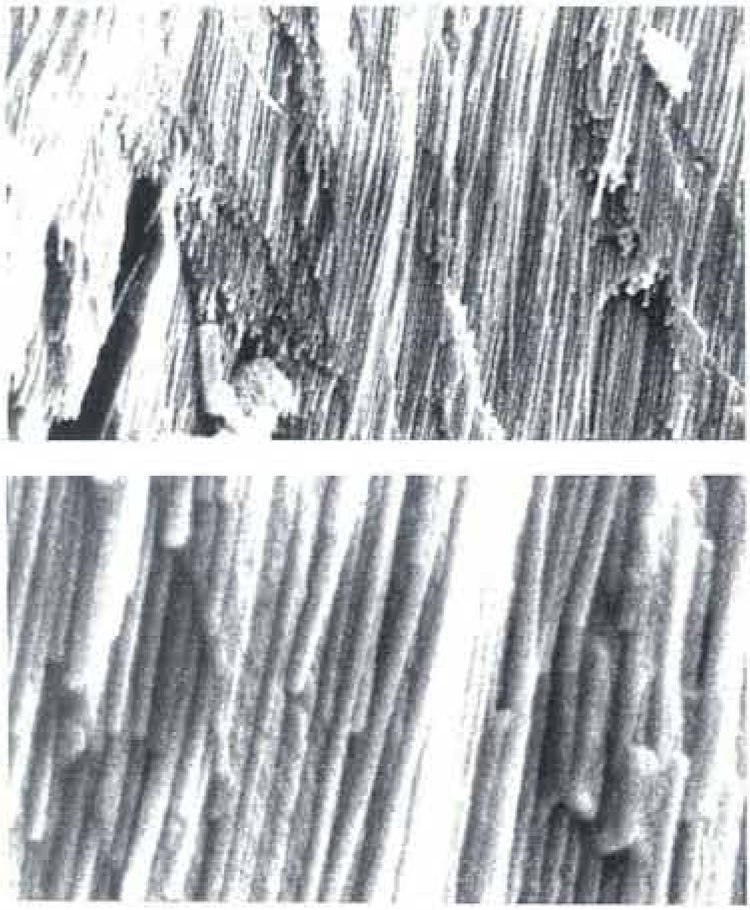


Figure 5.8 3

5.5.2.3. Nacreous Layers.

5.5.2.3.1. Positioning for specimens for L.M. and S.E.M. of medial surface of Inner Nacreous Layer and Outer Nacreous Layer.

FIGURE 5.84.Medial View of Right Valve of *Pinctada margaritifera* showing the Seventeen Positions on the Medial Surface selected for Light Photography and S.E.M. of the Nacre.

These positions are:

1. Shoulder Nacre near Hinge.
2. Shoulder Nacre near Adductor Muscle.
3. Ventral Pallial Gland Nacre.
4. Posterior Pallial Gland Nacre.
5. Proximal Ventral Pallial Nacre.
6. Middle Ventral Pallial Nacre.
7. Distal Ventral Pallial Nacre.
8. Ventral Nacreo-prismatic Junction.
9. Proximal Posterior Pallial Nacre - Posterior Pallial Gland Nacre.
10. Middle Posterior Pallial Nacre.
11. Distal Posterior Pallial Nacre - Posterior Nacreo-Prismatic Junction.
12. Ventro-Posterior Nacreo-prismatic Junction.
13. Anterior Pallial Gland Nacre.
14. Proximal Anterior Pallial Nacre.
15. Middle - Distal Anterior Pallial Nacre.
16. Anterior External Nacreous Layer and Nacreo-prismatic Junction.
17. Antero-ventral External Nacreous Layer and Nacreo-prismatic Junction.

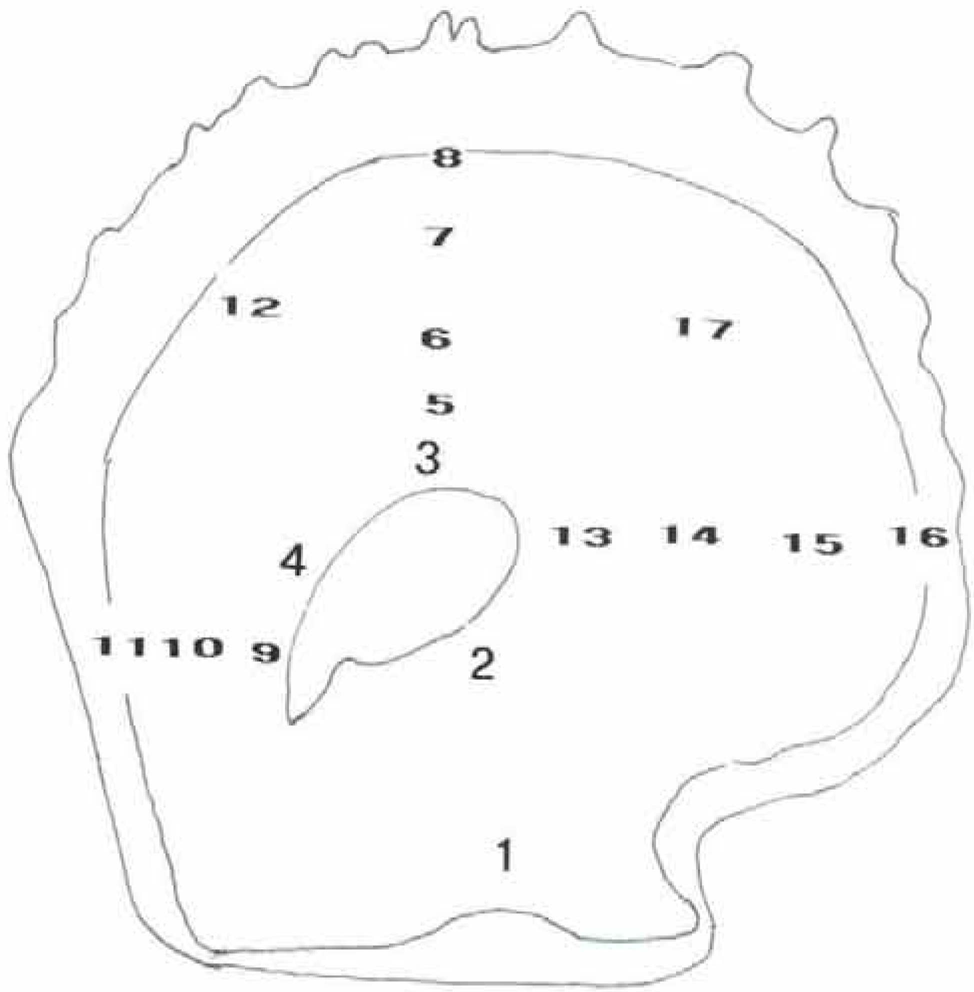


Figure5.84

5.5.2.3.2. Medial Surface Inner Nacreous Layer Shoulder Region Near Hinge - Figure 5.84, Position 1. This Region displays concentric circular curvilinear and parallel patterns (Figure 5.85 a).

The partly-bound and free Nacre Tiles cover about half the distance between successive Nacre Sheets and this distance, in any one location, is greater the more tightly curved are the edges of the Nacre Sheets. The average distance between Nacre Sheets from b to c in Figure 5.85 a is about 90 µm and that from d to e is about 40 µm.

Figure 5.85 b is to the left of c in 5.85 a, and c is just left of b in 5.85 a. e is a higher magnification of 5.85 b, and f is higher magnification of 5.85 c. d is from an area outside 5.85 a of parallel linear patterns of edges of Nacre Sheets.

While the geometry of the partly-bound and free Nacre Tiles from similar patterns of edges of Nacre Sheets in the one Region are almost invariably similar and their orientation to the related edge of a Nacre Sheet the same, (Figure 5.85 b, d and e), the geometry of the Nacre Tiles from a different pattern in the same region may be very different (Figure 5.85 c and f c.f. Figure 5.85, b, d and e). However, in any one pattern, the orientations of the equivalent geometric axes of the medial surfaces of the Nacre Tiles to the edges of the related Nacre Sheet remain the same, Figure 5.85. (d, e and f).

FIGURE 5.85 *Pinctada margaritifera* - Position 1 Figure 5.84.

Medial Surface of Inner Nacreous Layer Near Hinge.

a. S.E.M. x 80 Concentric, curvilinear and parallel linear patterns.

b. S.E.M. x 770 Higher magnification of curvilinear patterns, a above.

c. S.E.M. x 770 Higher magnification of concentric pattern in a above.

d. S.E.M. x 1500 Higher magnification of parallel linear patterns near a above.

e. S.E.M. x 1500 Higher magnification of b above.

f. S.E.M. x 1500 Higher magnification of c above.

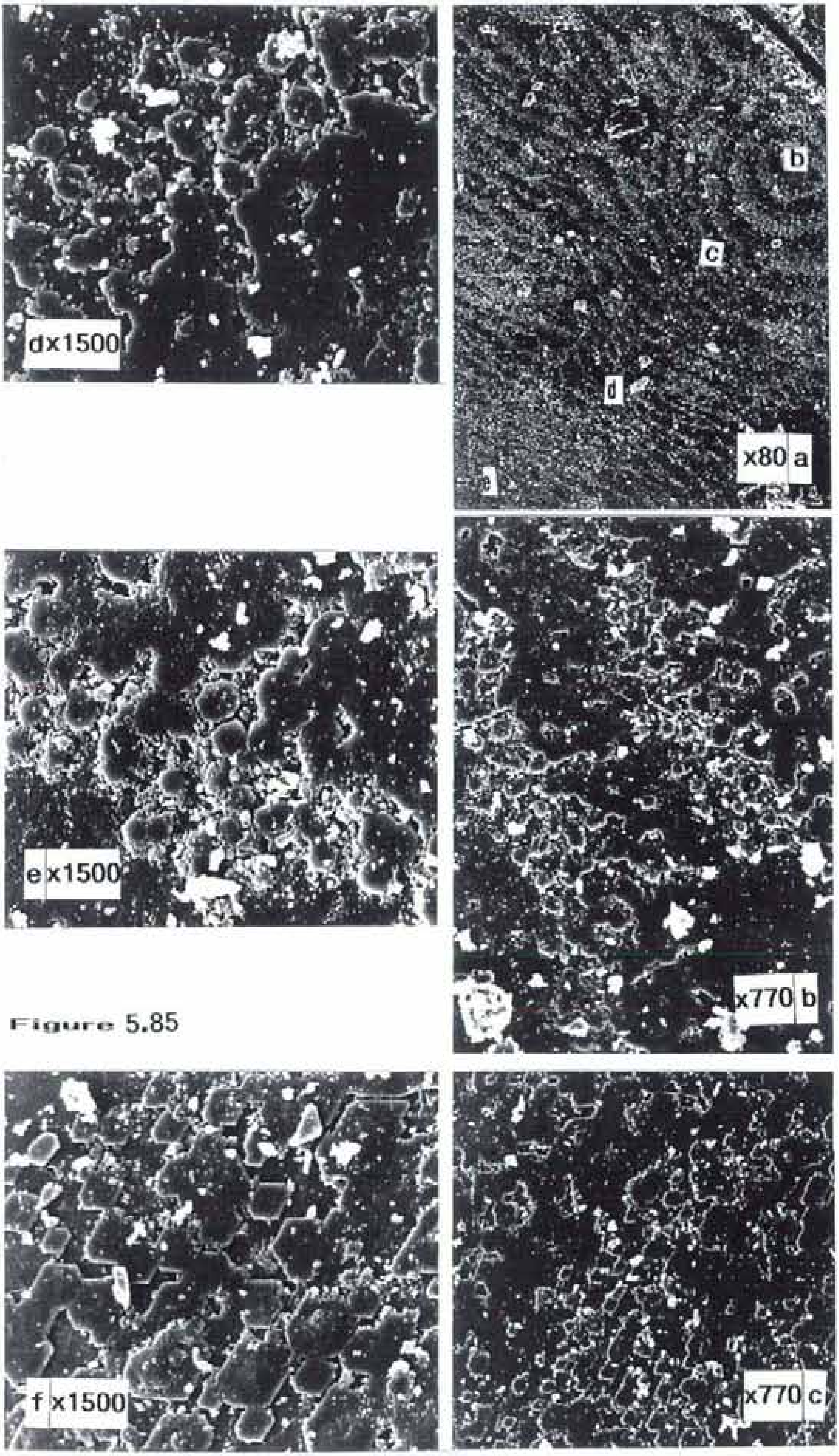


Figure 5.85

5.5.2.3.3. Medial Surface, Inner Nacreous Layer Shoulder Region Near Adductor Muscle Scar - Figure 5.84, Position 2.

Here the patterns of edges of Nacre Sheets are sometimes concentric circles (Figure 5.86. a), but there are areas of short spiral, anastomosing, tightly curved, and crooked edges to Nacre Sheets (Figure 5.86. b). In the latter, the geometric shape of the partly-bound and free Nacre Tiles is very variable ranging from nearly regular hexagons to truncated diamond shapes and diamond shpaes (Figure 5.86. c and d).

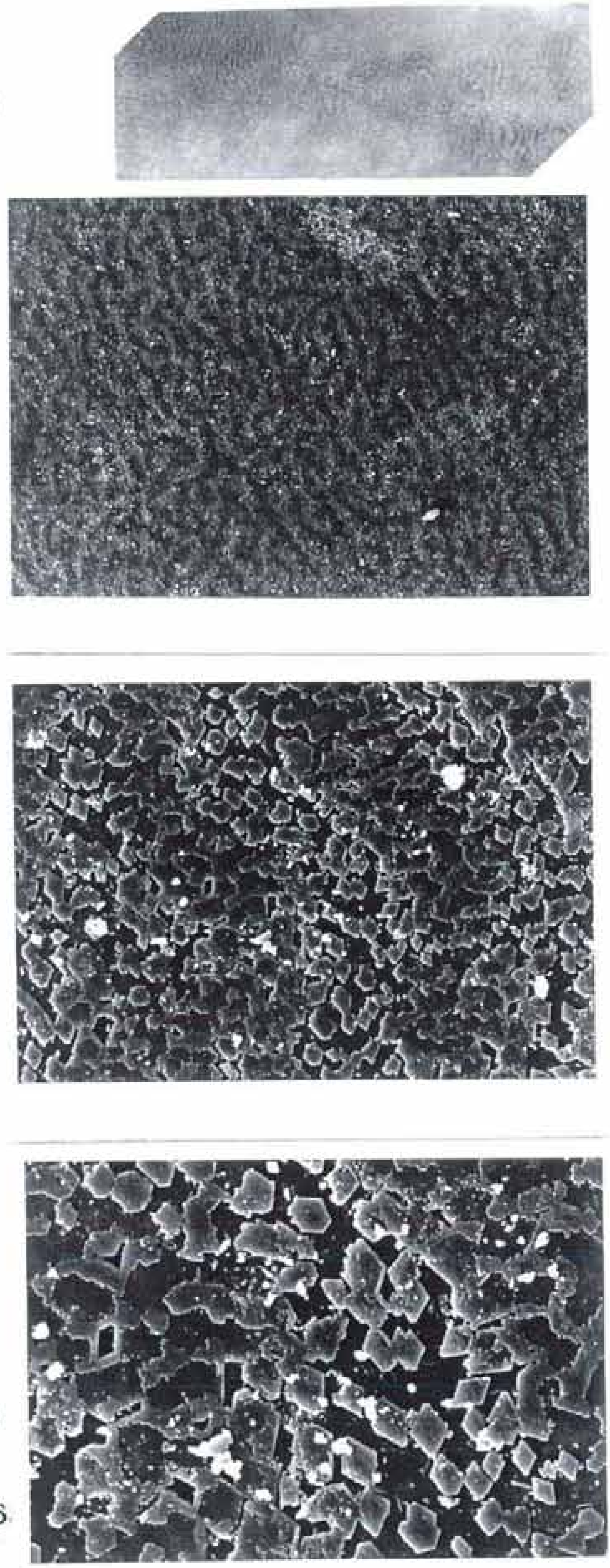
FIGURE 5.86. *Pinctada margaritifera* - Medial Surface Inner Nacreous Layer, Shoulder Region Near Adductor Muscle Scar, Position 2 Figure 5.84.

a. L.M. x 28 Concentric, curvilinear and nearly parallel linear patterns.

b. T.E.M. x 80 Short spiral, anastomosing and crooked patterns.

c. T.E.M. x 770 Higher magnification near centre of b above.

d. T.E.M. x 1500 Higher magnification near centre of a above.



a x28

b

x80

c

x770

d

x1500

Figure5.86

5.5.2.3.4. Medial Surface Inner and Outer Nacreous Layers from Ventral Pallial Gland to Ventral Nacreo-prismatic Junction - Figure 5.84. Positions 3, 5, 6, 7 and 8. (Figure 5.84. 4 is very like Figure5.84. Position 3.

Between the Ventral Pallial Gland and the Ventral Nacreo-prismatic Junction there is, generally, a gradation from a complex array of small concentric and spiral patterns with many anastomoses between edges of Nacre Sheets and discontinuous edges in the Pallial Gland Region (Figure 5.87. a), to the narrowly spaced parallel linear pattern with gentle curving of the Outer Nacreous Layer near the Nacreo-prismatic Junction (Figure 5.87. e).

Intervening, on the medial surface of the Proximal, Middle and Distal, Ventral Pallial Regions of the valve (Figure 5.87. b, c and d respectively) are progressively more regular curvilinear and parallel line patterns, although embayments of tightly curved patterns with the curvature directed towards the valve periphery (c in Figure 5.87. c) persist in the Middle Pallial Region between parallel linear patterns (p in Figure 5.87. c) and, more rarely even into the Distal Pallial Region (c and p respectively in Figure 5.87. d).

At the Nacreo-prismatic Junction, going distally, lines of progressively smaller Nacre Tiles lie in the grooves between the medial surfaces of the Major Prisms of the Prismatic Layer (Figure 5.87. f).

FIGURE 5.87. *Pinctada margaritifera* - Medial surface Inner and Outer Nacreous Layers from ventral Pallial Gland to ventral Nacreo-prismatic Junction.

a. L.M. x 28 Figure 5.84. Position 3.

b. L.M. x 28 Figure 5.84. Position 5.

c. L.M. x 28 Figure 5.84. Position 6.

d. L.M. x 28 Figure 5.84. Position 7.

e. L.M. x 28 Figure 5.84. Position 8.

f. T.E.M. x 80 Figure 5.84. Position 8.

a
x28

b
x28

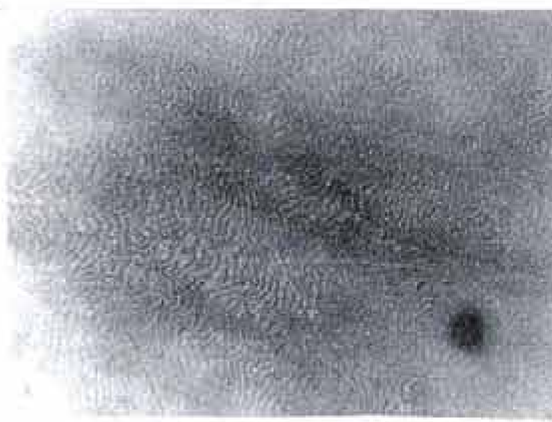
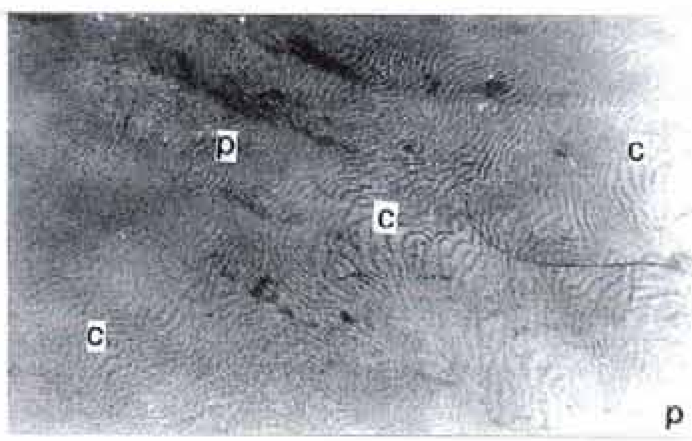
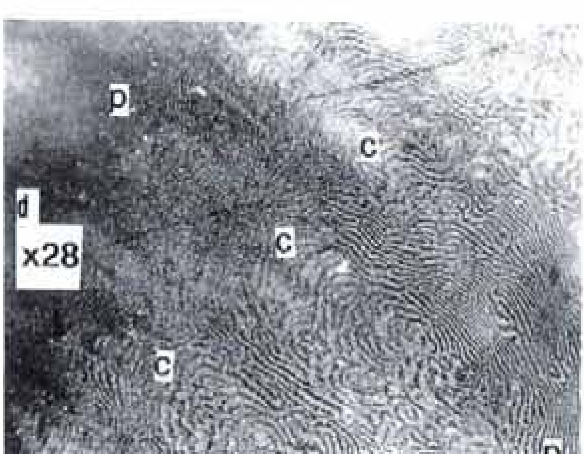


Figure5.87

c
x28





5.5.2.3.5. The Medial Surface of the Posterior Inner and Outer Nacreous Layers and Nacreo-prismatic Junction - Figure 5.84. Positions 9, 10 and 11.

The Posterior Pallial Gland Region of the medial surface of the Inner Nacreous Layer displays tightly curving anastomosing and discontinuous edges to Nacre Sheets with intervening curvilinear patterns and some narrowly spaced parallel linear patterns (Figure 5.88. a).

Going distally, the Middle Posterior Pallial Region (Figure 5.88. b) shows fewer tightly curved patterns and more curvilinear and narrowly spaced parallel linear patterns with fewer anastomoses.

This trend continues distally so that the Distal Posterior Pallial Region mostly displays fairly gently curved curvilinear patterns (Figure 5.88. c). In this region the partly-bound and free Nacre Tiles mostly have a diamond or truncated diamond geometric shape medially, with the long axes of the diamonds usually normal to the direction of the edges of the related Nacre Sheets (Figure 5.88. d).

The Medial surface of the Outer Nacreous Layer shows slightly wavy parallel edges to Nacre Sheets with a few anastomoses with the direction of the edges roughly parallel to the Nacreo-prismatic Junction.

FIGURE 5.88. *Pinctada margaritifera* -Medial Surface of Posterior Inner and Outer Nacreous Layers and Nacreo-Prismatic Junction.

a. L.M. x 28 Figure 5.84. Position 9.

b. L.M. x 28 Figure 5.84. Position 10.

c. T.E.M. x 80 Figure 5.84. Position 10.

d. T.E.M. x 1500 Figure 5.84. Position 10.

e. L.M. x 28 Figure 5.84. Position 11.

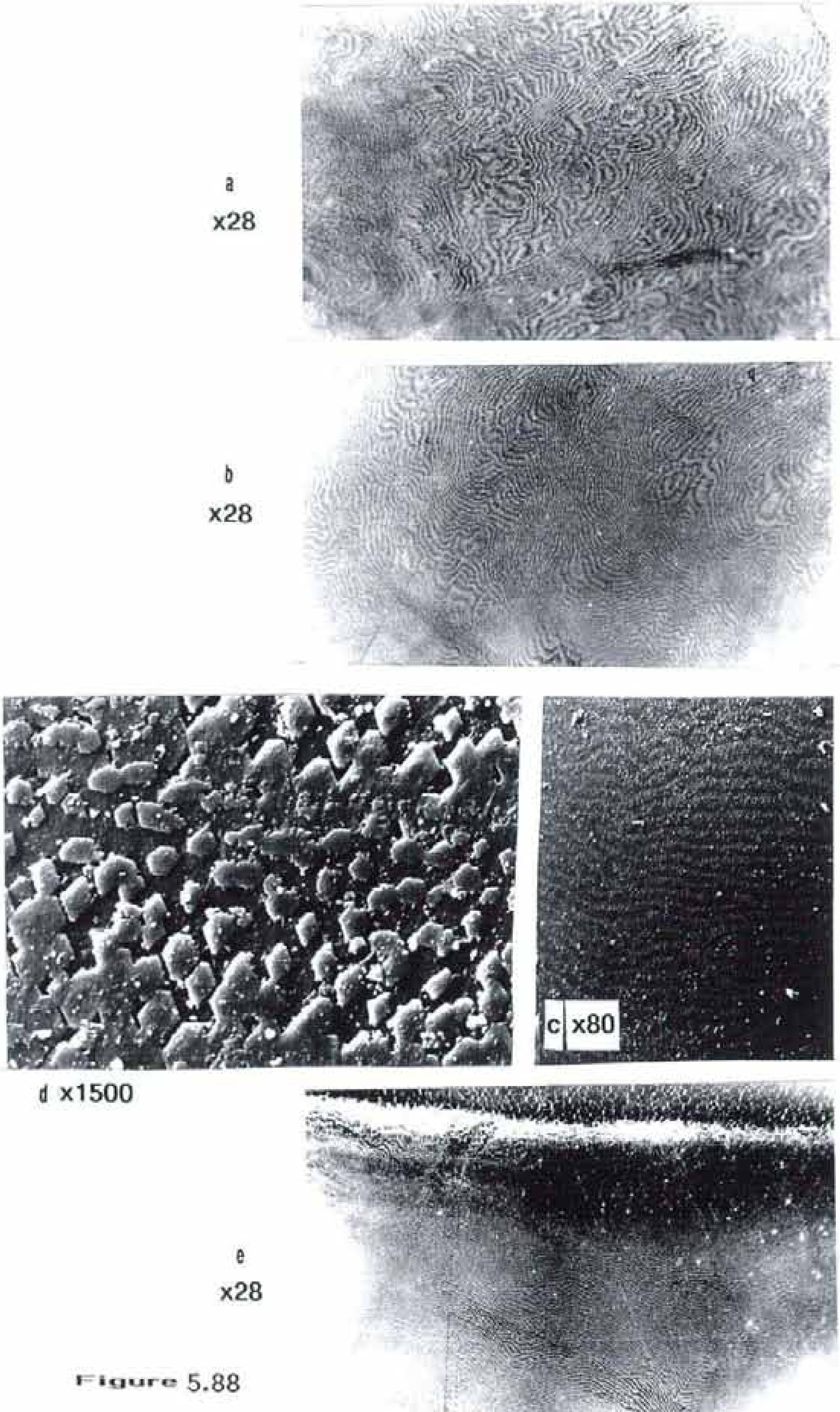


Figure 5.88

5.5.2.3.6. Medial Surface, Anterior Inner and Outer Nacreous Layers.

The medial surface of the Anterior Pallial Gland Nacre shows tightly curved and widely spaced edges to successive Nacre Sheets (Figure 5.89. a). The partly-bound and free Nacre Tiles tend to regular hexagons (Figure 5.89. b).

More distally, the Anterior Proximal Pallial Inner Nacreous Layer displays relatively large embayments of widely spaced tightly curved patterns of edges of Nacre Sheets surrounded by concentrically curved patterns whose curvature projects distally (Figure 5.89. c).

These patterns of anastomosing, tightly curved, and widely spaced edges to Nacre Sheets are still found in the Distal Anterior Pallial Nacre (Figure 5.89. d) and up to the area of junction of the Inner and Outer Nacreous Layers (Figure 5.89. e) just proximal to the Nacreo-prismatic Junction.

The Anterior Outer Nacreous Layer shows partly-bound and free Nacre Tiles which are truncated diamond shapes tending towards regular hexagons, with the long axis of symmetry parallel to the edges of the Nacre Sheets, (Figure 5.89. f).

FIGURE 5.89. *Pinctada margaritifera* - Medial Surface, Anterior Inner and Outer Nacreous Layers.

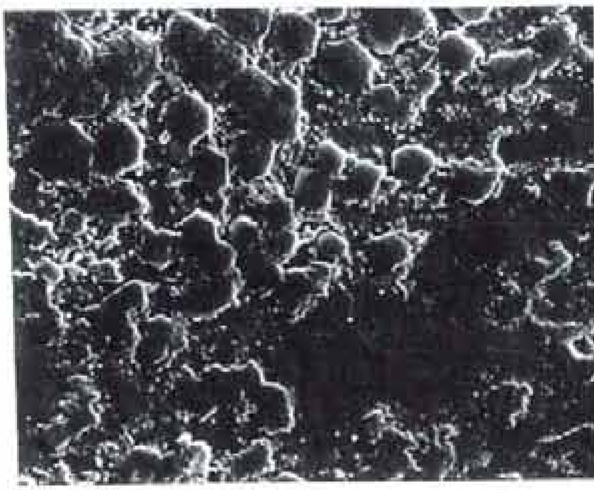
a. T.E.M. x 80 Inner Nacreous Layer, Figure 5.84, Position 13.

b. T.E.M. x 1500 As for a above.

c. T.E.M. x 80 Inner Nacreous Layer, Figure 5.84, Position 14.

d. T.E.M. x 80 Inner and Outer Nacreous Layers, Figure 5.84, Position 15.

e. T.E.M. x 1500 Outer Nacreous Layer, Position 16.



b x1500

a x80

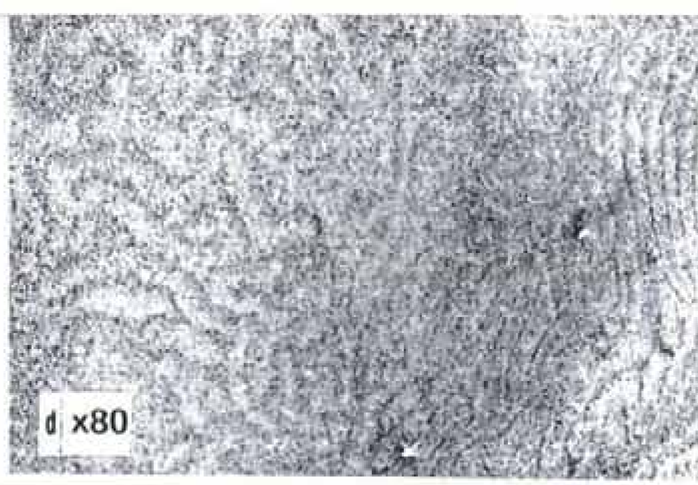


Figure 5.89

5.5.2.3.7. Medial Surface Inner and Outer Nacreous Layer and Nacreo-Prismatic Junction - Antero-ventral Periphery.

In Figure 5.90. P is proximal and D. distal.

Areas with patterns of edges of Nacre Sheets featuring short spiral, circular and anastomosing structures occur here until near the junction of the Inner Nacreous Layer and the Outer Nacreous Layer (inl and onl respectively in Figure 5.90. a, b, c and d). The tightly curved structures generally have their convexities directed distally. (Figure 5.90. b and d). Where the individual Nacre Tiles appear to be mostly Nacreous Organic Matrix or only forming Nacreous Organic Matrix (Figure 5.90. d, e and f) the patterns of edges of Nacre Sheets (Figure 5.90. d) and the disposition relative to each other and to the edges of the Nacre Sheets of the Nacre Tiles, (Figure 5.90. e and f), appear to be the same as in calcified Nacre Sheets.

The Outer Nacreous Layer (onl in Figure 5.90. a and b), has narrowly spaced linear edges to Nacre Sheets about parallel to the Nacreo-prismatic Junction.

At the Nacreo-prismatic Junction partly-bound Nacre Tiles lie in the Grooves between the medial ends of the Major Prisms of the Prismatic Layer (Figure 5.90. g).

FIGURE 5.90. *Pinctada margaritifera* - Medial Surface Inner and Outer Nacreous Layers and Nacreo-prismatic Junction - Antero-ventral Periphery - Figure 5.84, Position 17.

a.	L.M. x 28	Antero-ventral Nacreo-prismatic Junction.
b.	L.M. x 28	Junction of Inner (i) and Outer (o) Nacreous Layers.
c.	L.M. x 28	Inner Nacreous Layer proximal to b above.
d.	T.E.M. x 80	Partly calcified Inner Nacreous Layer.
e.	T.E.M. x 770	As for d above, higher magnification.
f.	T.E.M. x 1500	As for d and e above, higher magnification.
g.	T.E.M. x 1500	Antero-ventral Nacreoprismatic Junction.

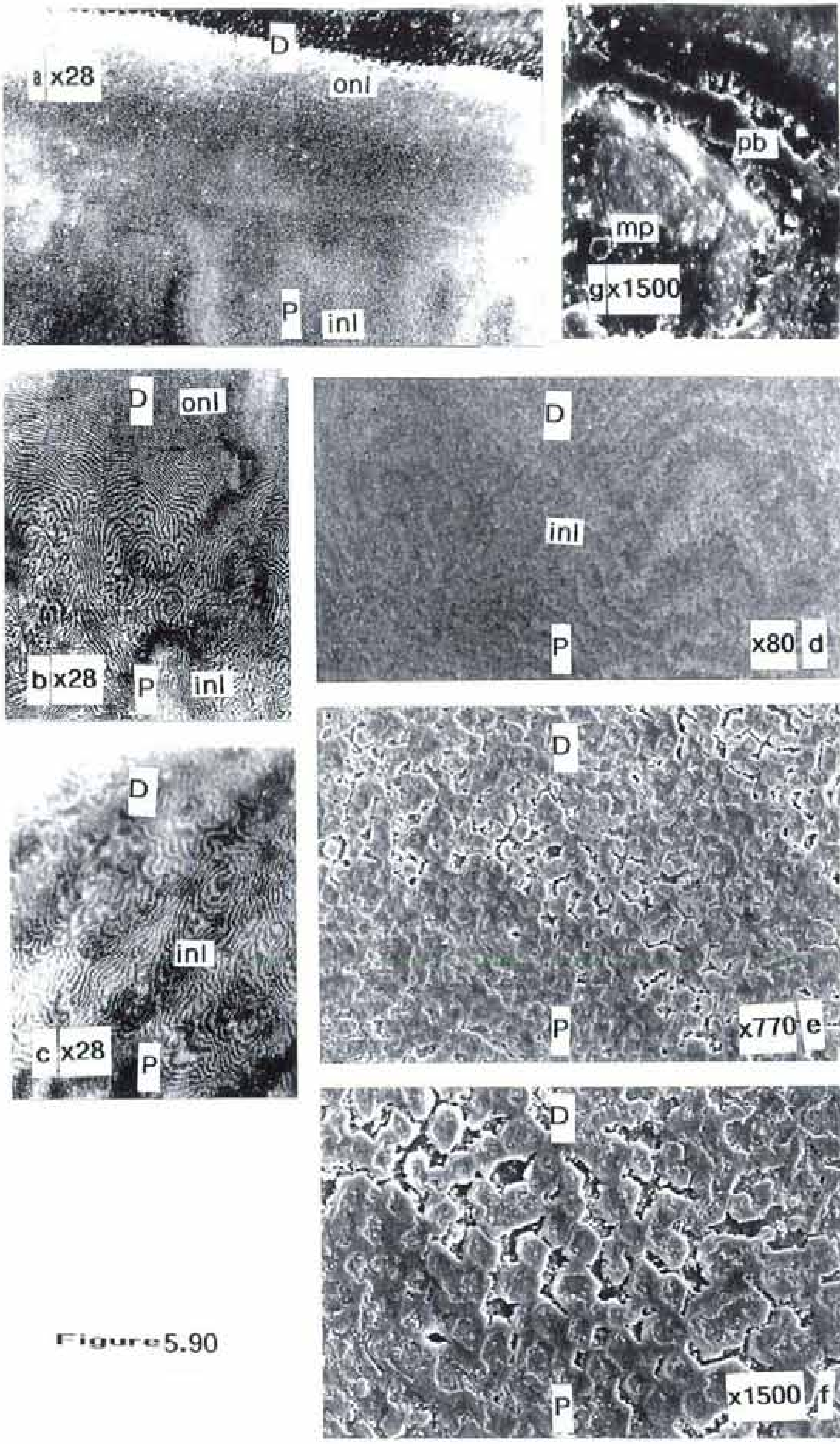

D
a x28
onl
P
inl
pb
mp
g x1500
D
onl
b x28
P
inl
D
inl
P
x80
d
D
inl
c x28
P
D
P
x770
e
D
P
x1500
f

Figure 5.90

5.5.2.3.8. Inner and Outer Nacreous Layers - Edge-on Views.

In Figure 5.91. D is distal, L. lateral and M. medial.

The Inner Nacreous Layer Nacre Tiles which abut the Hinge appear to do so in an edge-on plane surface (Figure 5.91. a).

The Nacre Tiles in the Inner Nacreous Layer near the Hinge show the curved surfaces on the medial and lateral surfaces normally found in Nacre Tiles in this region (Figure 5.91. b).

The Outer Nacreous Layer shows extensive patterns of Nacre Stairs (Figure 5.91. c).

The widths of Nacre Sheets throughout the Inner and Outer Nacreous Layers of this species are quite constant and average about 0.6um (Figure 5.91. a, b, c, and d).

Between the Outer Nacreous Layer and the Major Prisms of the Prismatic Layer (onl and mp respectively in Figure 5.91. d) there is sometimes what appears to be a layer of aragonite prisms which are about 10um thick medio laterally, (ap in Figure 5.91. d).

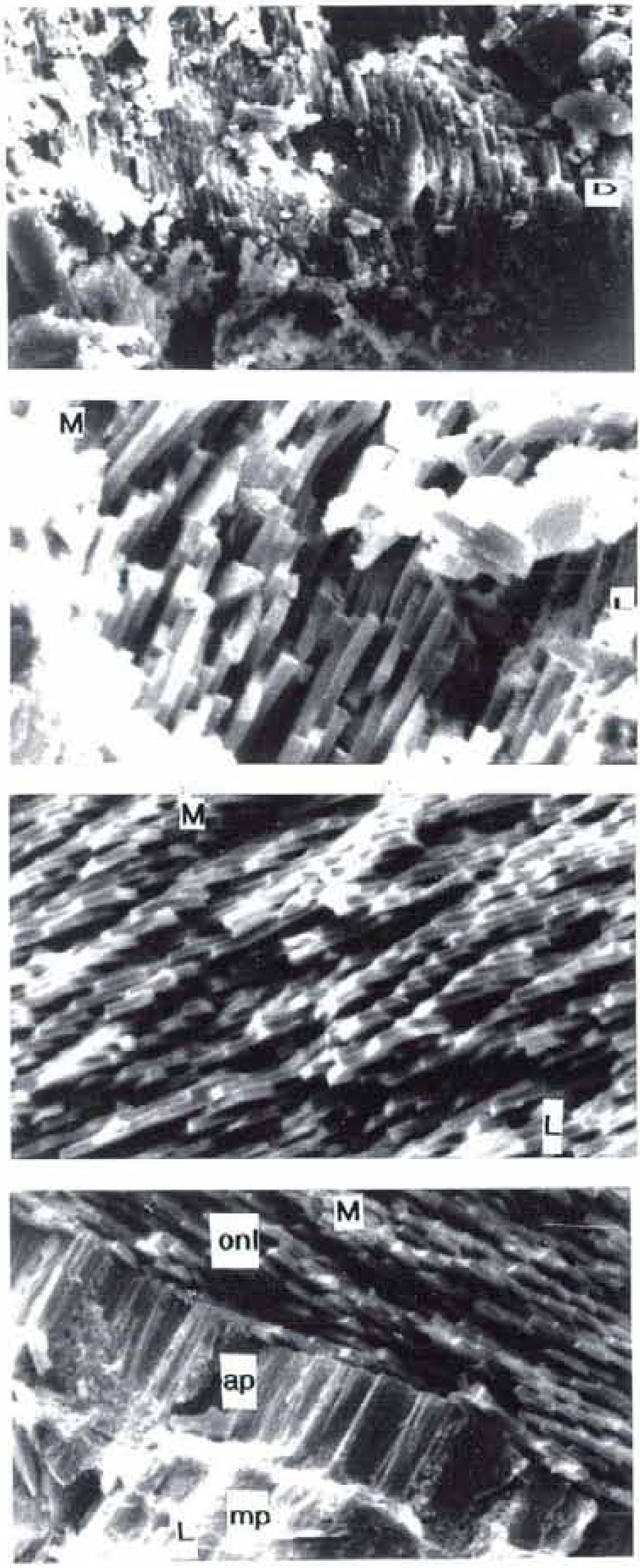
FIGURE 5.91. *Pinctada margaritifera* Inner and Outer Nacreous Layers - Edge on View.

a. S.E.M. x 1900 Inner Nacreous Layer Hinge abutment.

b. S.E.M. x 3700 Radial Broken Surface Outer Nacre Layer.

c. S.E.M. x 1900 Radial Broken Surface - Inner Nacreous Layer.

d. S.E.M. x 1900 Radial Broken Surface - Ventral Nacreo-Prismatic Junction.



a
x1900

b
x3700

c
x1900

d
x1900

Figure5.91

5.5.2.4. S.E.M. Prismatic Layer.

5.5.2.4.1. Medial surface of Growth Scale.

In Figure 5.92. a P is proximal and D distal.

The medial surface of the Growth Scale is not covered by an Inner Fibrous Sheath near the Nacreo-prismatic Junction (Figures 5.87. f, 5.88. e and 5.90. g) but is so covered more distally (ifs in Figure 5.92. b, c and d). The outlines of the underlying Inner Prismatic End Plates are discernable through the Inner Fibrous Sheath (Figure 5.92. b and d).

The fibres of the Inner Fibrous Sheath are about 0.3 µm apart (Figure 5.92. c and d). The Growth Process is broken off just proximal to D in Figure 5.92. a. Medial to the Inner Fibrous Sheath on the Distal Growth Scale, (proximal part of the Growth Process) is a coarsely fibred material which appears to lie as a detrital material on the medial surface (d in Figure 5.92. b).

FIGURE 5.92. *Pinctada margaritifera* -Medial Surface of Distal Growth Scale - Inner Fibrous Sheath and Detrital Material.

a. S.E.M. x 15 Medial surface distal Growth Scale and Growth Process.

b. S.E.M. x 80 Higher magnification of a above.

c. S.E.M. x 13000 Higher magnification of Inner Fibrous Sheath from b above.

d. S.E.M. x 6100 Lower magnification, same centre as c above.

a
x15

b
x80

c
x18000

d
x6100

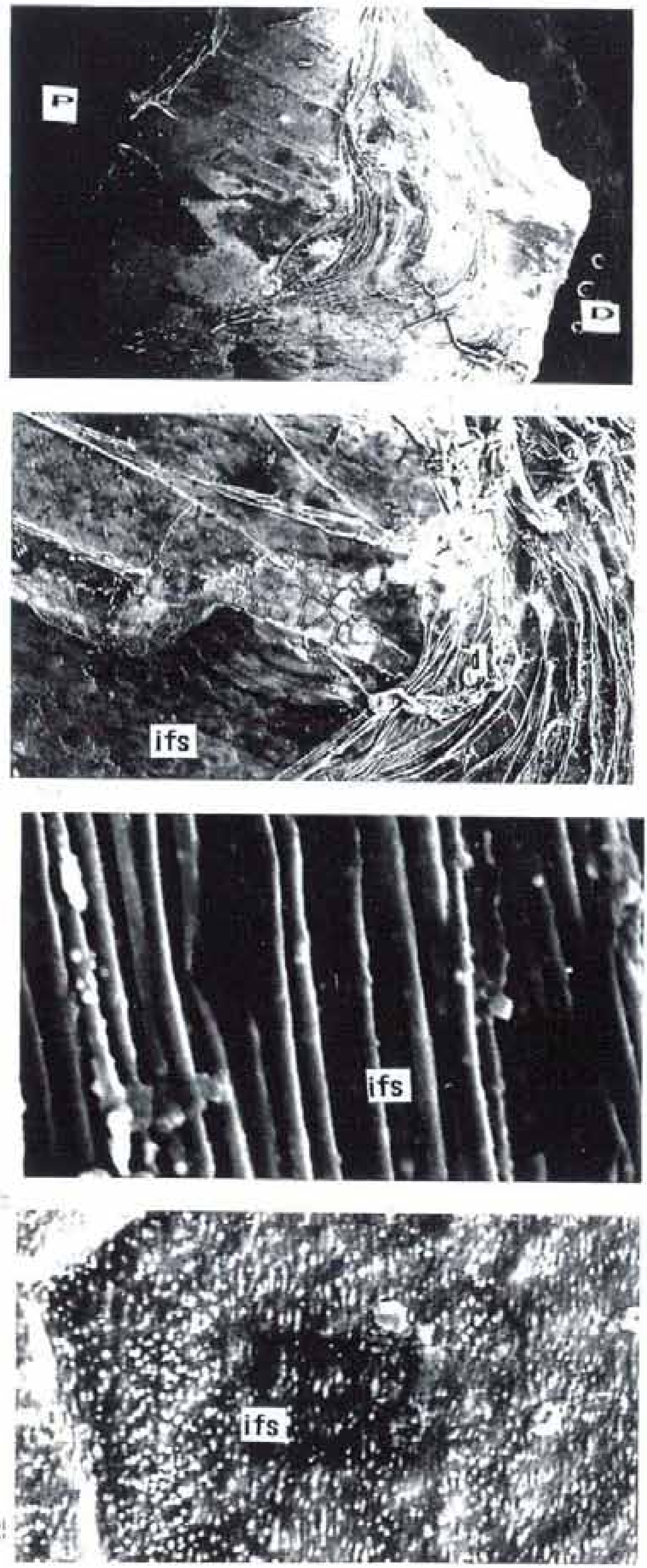


Figure 5.92

5.5.2.4.2. Outer Fibrous Sheath of Prismatic Layer.

The free outer surface of all Growth Scales and some of the overlain surface of Growth Scales are covered by an Outer Fibrous Sheath. (Figure 5.93. a, b, c and d).

The transverse periodicity of the surface fibres are about 1.4 µm apart (Figure 5.93. c and d). The outline of the Outer Prismatic End Plates of the underlying prisms is discernable through the Outer Fibrous Sheath and there is a indentation in the Outer Fibrous Sheath aproximately centrally placed over the underlying prisms. (Figure 5.93. a).

Where there is an Outer Fibrous Sheath on one Growth Scale (GS1 in Figure 5.93), and an Inner Fibrous Sheath on the Growth Scale lateral to it, (GS2 in Figure 5.93), the torn edges of both can sometimes be seen on a radial broken surface (Figure 5.93. b).

FIGURE 5.93. *Pinctada margaritifera* - Outer Fibrous Sheath of Prismatic Layer.

a. S.E.M. x 250 Outer surface Growth Scale.

b. S.E.M. x 230 Broken radial surface Prismatic Layer, showing torn edges of Outer and Inner Fibrous Sheaths.

c. S.E.M. x 1900 Higher magnification Outer fibrous Sheath from a.

d. S.E.M. x 1900 As for c above.

a

x250

b

x230

c

x1900

d

x1900

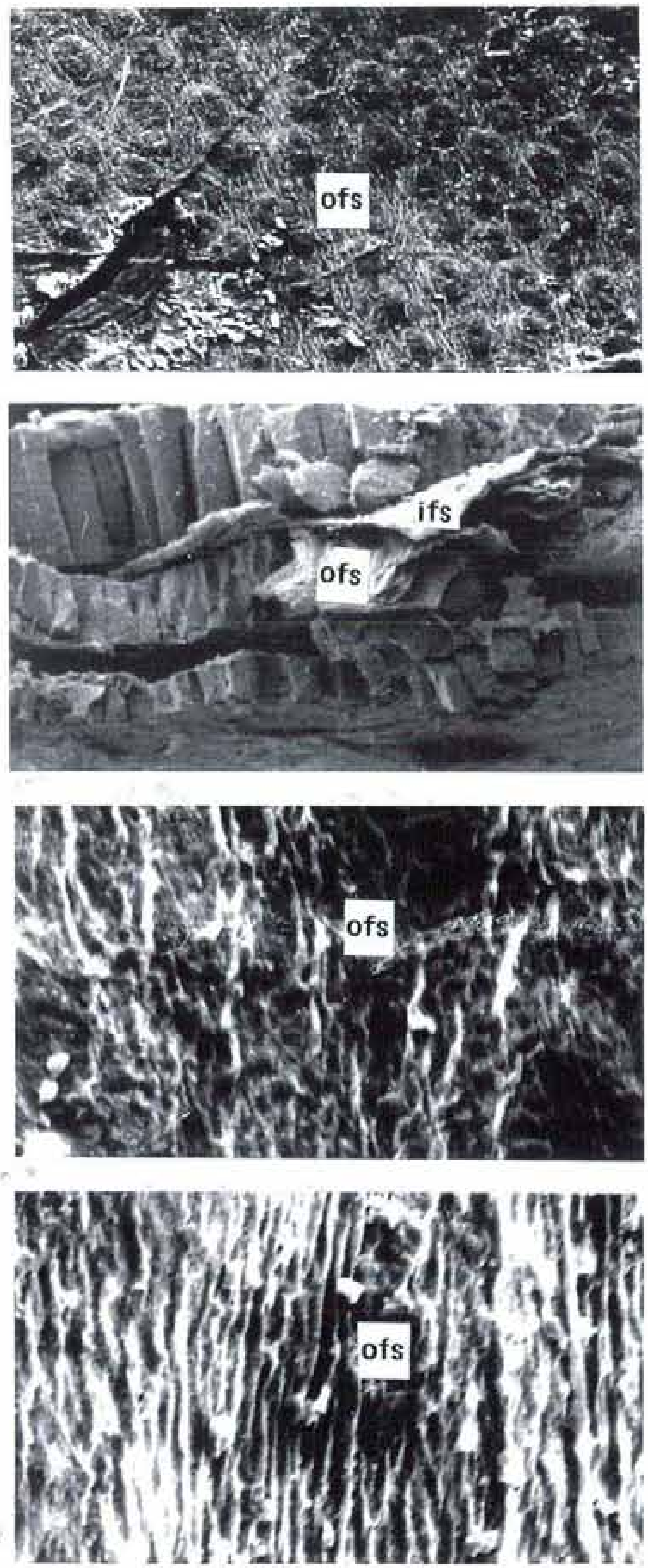


Figure 5.93

5.5.2.4.3. Growth Process. Medial Surface.

In Figure 5.94 P is proximal and D. distal.

This is covered with an Inner Fibrous Sheath (Figure 5.94. b, c and d). The transverse periodicity of the fibres is about 0.3 μm. (Figure 5.94. c and d). The outlines of the Inner Prismatic End Plates are visible through the Inner Fibrous Sheath (Figure 5.94. c). In places the Inner Fibrous Sheath appears to be aberrantly folded on itself indicating that there is no strict spatial relationship between the Inner Fibrous Sheath and the Inner Prismatic End Plates which it covers (Figure 5.94. c and d).

FIGURE 5.94. *Pinctada margaritifera* - Medial surface . Growth Process.

a. S.E.M. x 15 Medial srface of Growth Process which is broken distally

b. S.E.M. x 230 Higher magnification near ifs in a.

c. S.E.M. x 1500 Folds in torn Inner Fibrous Sheath over Inner Prismatic End Plates.

d. S.E.M. x 1500 As for c above.

a
x15

b
x230

c
x1500

d
x1500

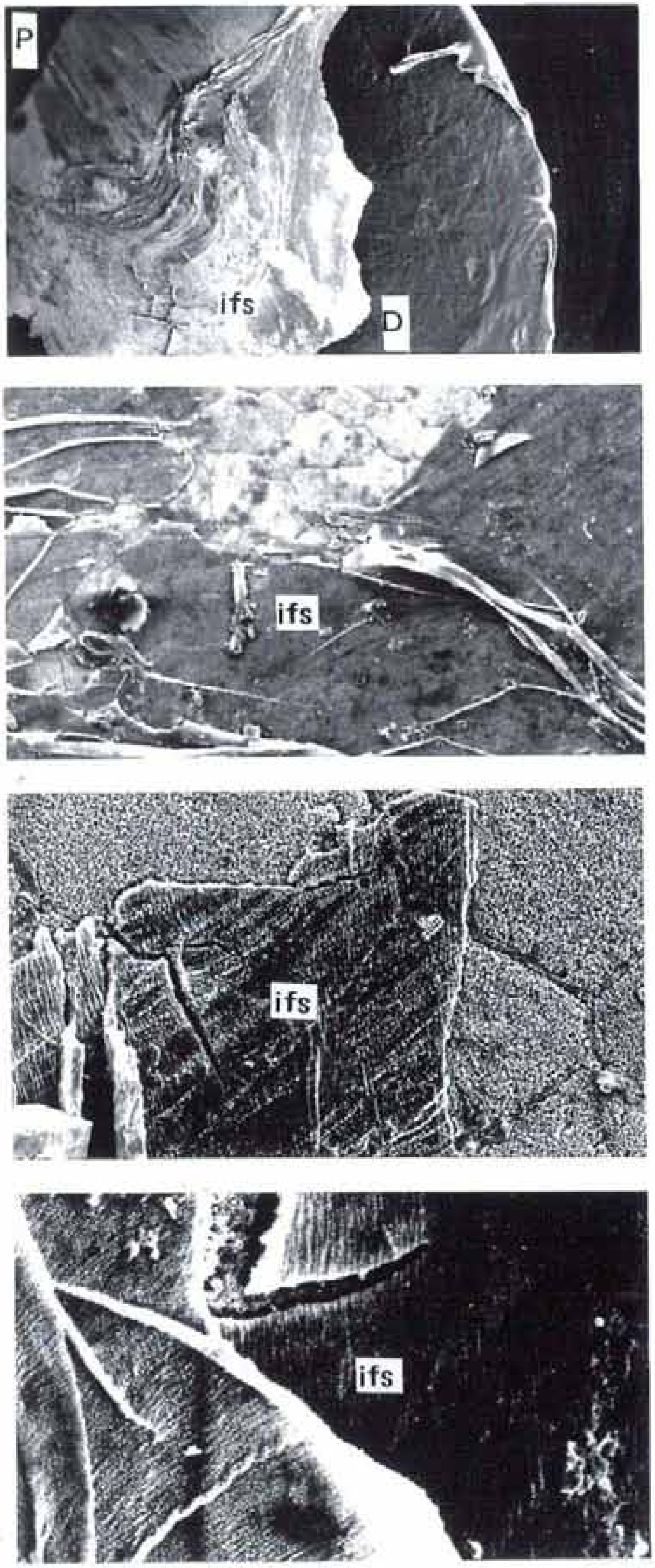


Figure5.94

5.5.2.4.4. Growth Process. Inner Prismatic End Plates, Outer Prismatic End Plates, Outer Fibrous Sheath.

The Growth Processes invariably display Inner Prismatic End Plates under L.M. of decalcified Shell but this is not obvious under S.E.M. (Figure 5.95. a and b).

With light acid etching a herringbone pattern of Inner Prismatic End Plate is shown (Figure 5.95. c).

The Outer Fibrous Sheath covers the lateral surface of the Growth Process unless abraded or destroyed (Figure 5.95. d, e, f and g).

The transverse periodicity of the fibres of the Outer Fibrous Sheath is about 1.4 µm and the surface bears small granules (Figure 5.95. f and g).

The underlying pattern of the Outer Prismatic End Plates is visible through the Outer Fibrous Sheath and a depression in the latter is common about the centre of the lateral end of the Growth Process Prisms (d in Figure 5.95. d and e).

Beneath the Outer Fibrous Sheath the Outer Prismatic End Plates of the Growth Process have a spider web pattern of Organic Matrix (OPEP in Figure 5.95. d and e).

FIGURE 5.95. *Pinctada margaritifera* - Growth Process - Inner Prismatic End Plates, Outer Prismatic End Plates, Outer Fibrous Sheath.

a. S.E.M. x 480 Medial surface of Growth Process Prisms.

b. S.E.M. x 2000 As for a above at higher magnification.

c. S.E.M. x 6000 Acid etched medial surface of Growth Process Prisms.

d. S.E.M. x 770 Lateral surface of Growth Process.

e. S.E.M. x 1500 Higher magnification of d.

f. S.E.M. x 950 As for e above.

g. S.E.M. x 8100 As for e and f above.

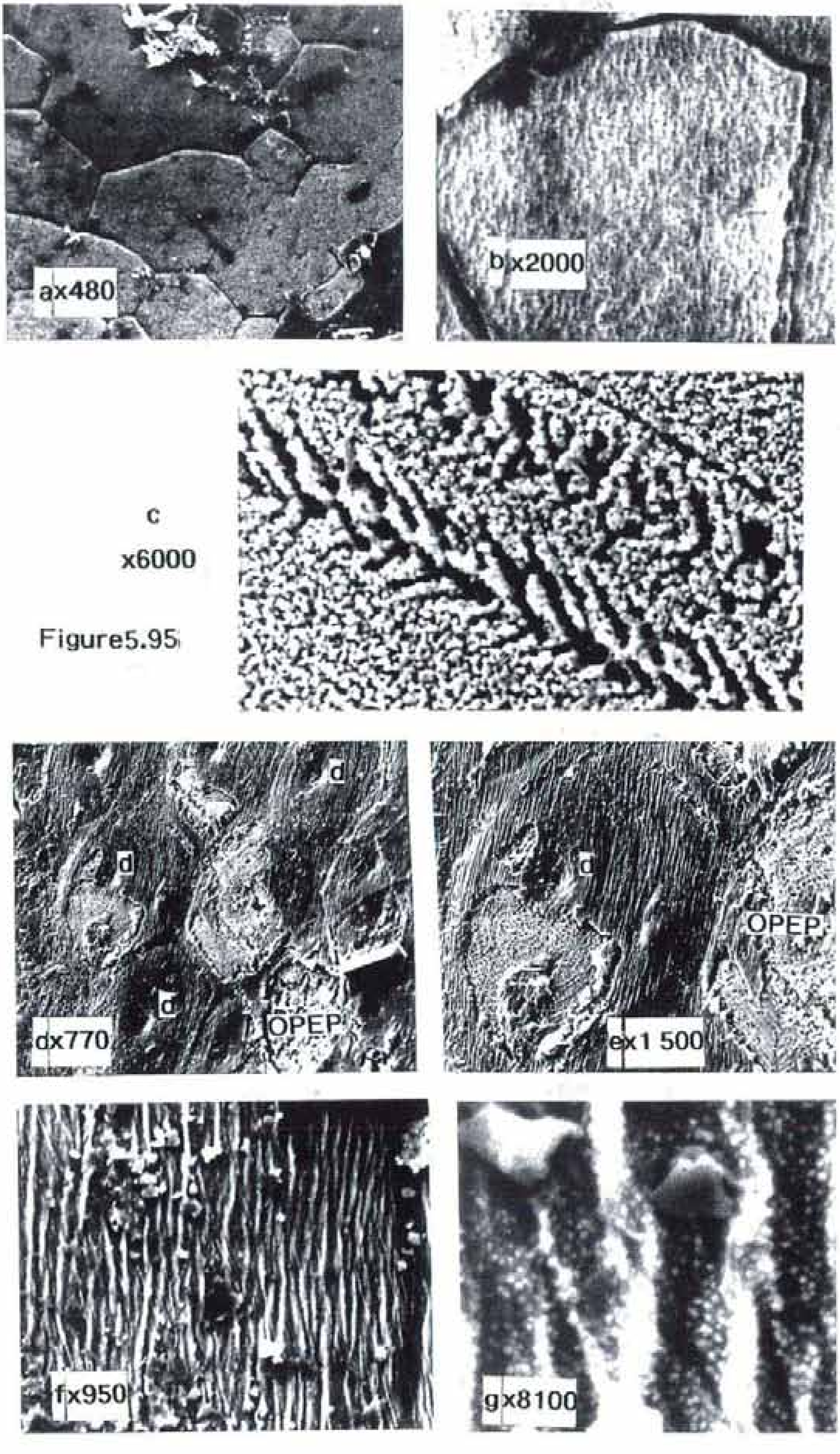

a x480
b x2000
c
x6000
d
d
d
d x770
OPEP
d
OPEP
e x1 500
f x950
g x8100

Figure5.95

5.5.2.4.5. Radial Broken Surface of Prismatic Layer.

Between the prisms of the Prismatic Layer are the Transverse Parallel Side Walls (tp in Figure 5.96. b). The side wall comprised of this material is common to adjoining prisms. Internally lining this material appears to be the Linear Structure Interprismatic Organic Matrix (ls in Figure 5.96. d). This, in turn, appears to be lined internally by Reteform Interprismatic Organic Matrix (riom in Figure 5.96. a, d and e). In Figure 5.96. d, it appears the Reteform Interprismatic Organic Matrix (riom) is structurally related to the Linear Structure Inter-prismatic Organic Matrix (ls) and in Figure 5.96. e it appears that the twin layers of the Intraprismatic Organic Matrix (iom) are structurally related to the Reteform Intraprismatic Organic Matrix (riom). On a radial broken surface where the Intraprismatic Organic Matrix has pulled out leaving the broken Calcite Tables (ct in Figure 5.96. g) there are usually series of lens shaped holes from which the Intraprismatic Organic Matrix has been pulled (l in Figure 5.96. g). In this species where a more lateral Growth Scale overlies the next more medial Growth Scale there sometimes appears to be no structural barrier proximally (j in Figure 5.96. f); sometimes there are Outer and Inner Prismatic End Plates (opep and ipep respectively in Figure 5.96. c); and in other places both Outer Fibrous Sheath and Inner Fibrous Sheath (ofs and ifs respectively in Figure 5.96. b).

FIGURE 5.96. *Pinctada margaritifera* Rad. Sufarce of Prism.Layer.

a. S.E.M. x 930 Reteform Interprismatic Organic Matrix.

b. S.E.M. x 1900 Transvers Parallel Side Walls.

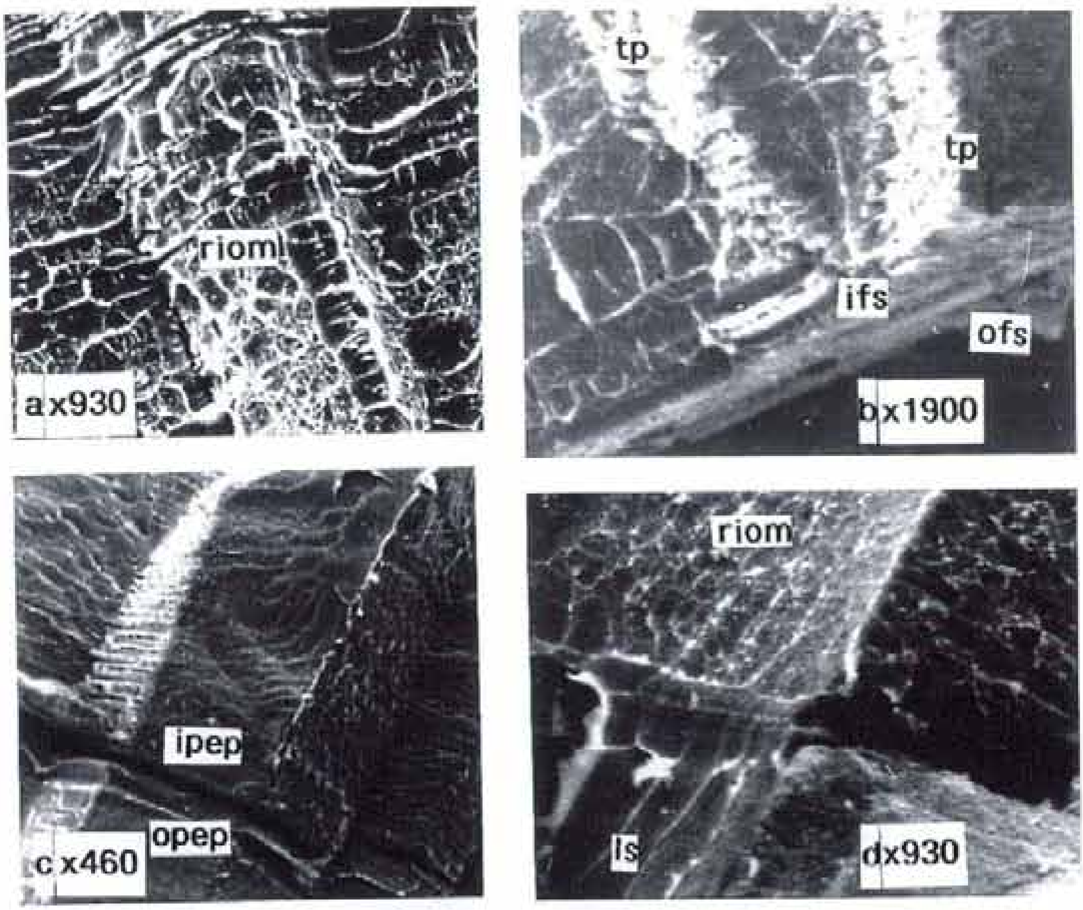
c. S.E.M. x 480 Inner and Outer Prismatic End Plates.

d. S.E.M. x 930 Linear Structure and Reteform Interprismatic Organic Matrix.

e. S.E.M. x 1900 Intraprismatic Organic Matrix.

f. S.E.M. x 460 Junction of Growth Scales.

g. S.E.M. x 1900 Calcite Tablets.


riom
ax930
tp
tp
ifs
ofs
bx1900
ipep
opep
c x460
riom
ls
dx930

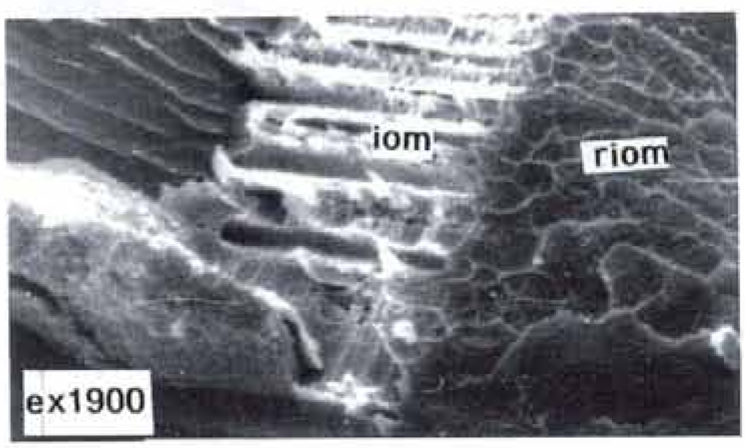
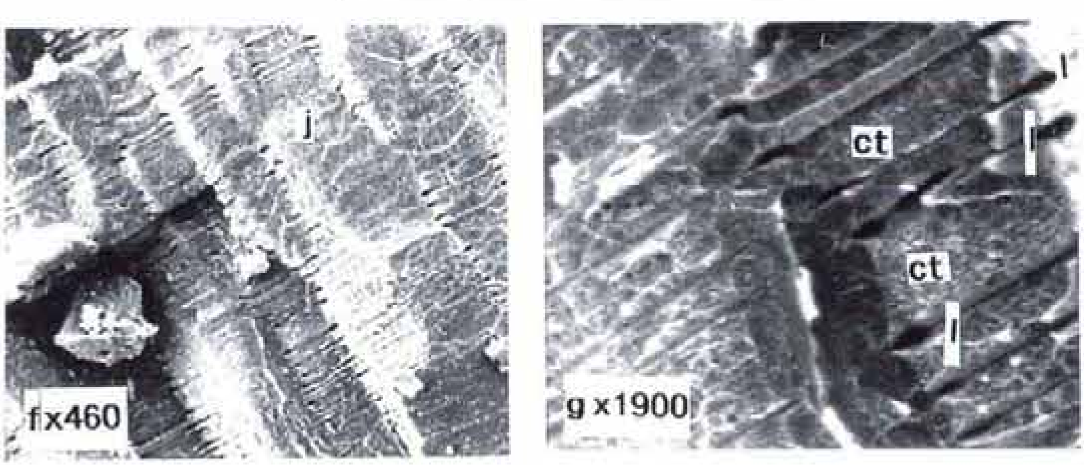

iom
riom
ex1900

Figure 5.96


j
fx460
l
l
ct
ct
l
g x1900

5.5.2.5. L.M. Decalcified Shell.

5.5.2.5.2. L.M. Decalcified Prismatic Layer.

In the Prismatic Layer of this species the successive Growth Scales may infrequently be physically defined for all of their length by an Outer Fibrous Sheath but more usually the separation is partial for only part of the radial length of each Growth Process. In such cases the definition is usually provided by an Outer Fibrous Sheath over Outer Prismatic End Plates and, usually Inner Lateral Structure.

In Figure 5.97. a, b and c tp is Transverse Parallel Side Walls, opep is Outer Prismatic End Plates ipep (staining pink) is Inner Prismatic End Plates il is Inner Lateral Structure.

The Transverse Parallel Side Wall Organic Matrices usually stain golden with A.B./M.S.B. but bands of aberrant purple staining lying parallel to the lateral surface of the Growth Scale in which they occur are common (Figure 5.97. a, b c and d). The aberrantly stained Transverse Parallel Side Wall Organic Matrices are invariably partly destroyed by acid hydrolysis (Figure 5.97. b, c and d). The turquoise material (ip in Figure 5.97. b and d), is remnants of Intraprismatic Organic Matrix.

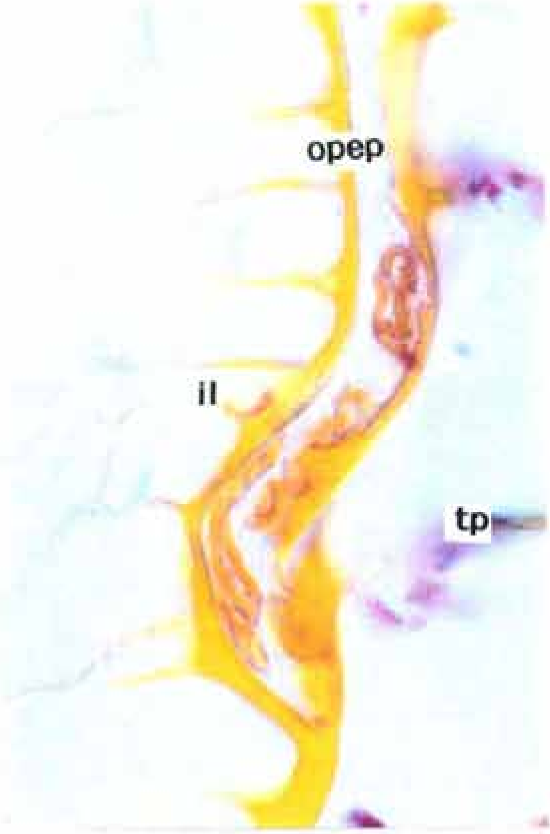
FIGURE 5.97. *Pinctada margaritifera* - Stained Radial Section of Prismatic Layer.

a. L.M. x 46 M.S.B. Multiple Growth Scales.

b. L.M. x 230 M.S.B. Three Growth Scales.

c. L.M. x 1200 M.S.B. Outer Prismatic End Plate and Inner Lateral Structure.

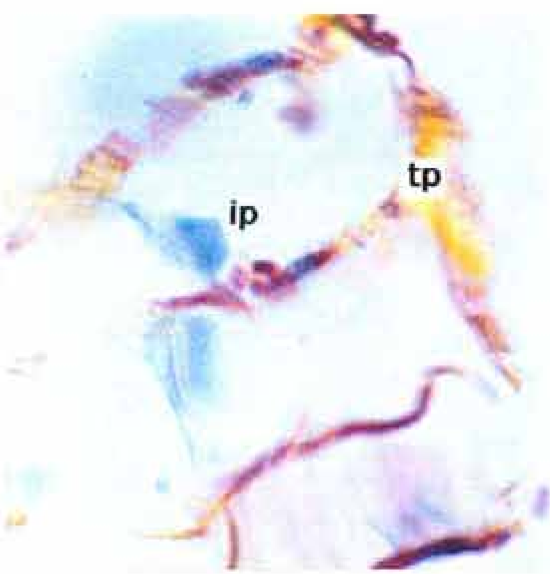
d. L.M. x 1200 M.S.B. Intraprismatic Organic Matrix.



c x1200

a x46

d x1200



b x230

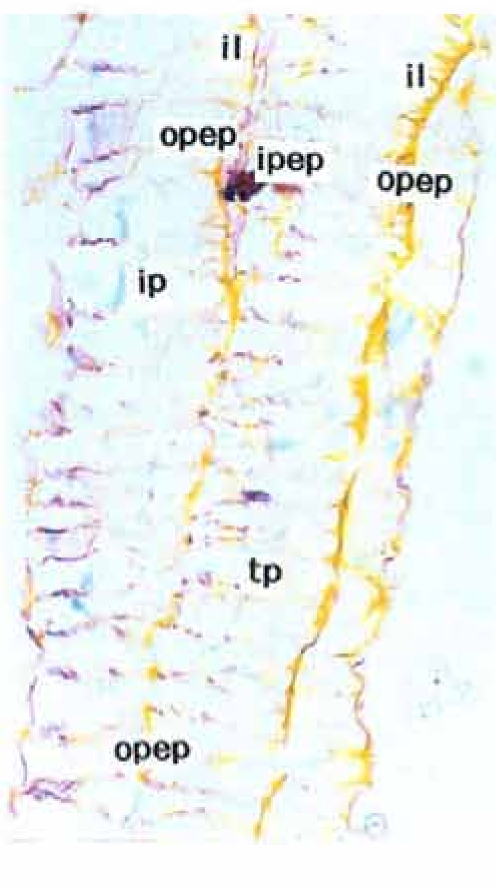


Figure5. 97

5.5.2.5.2. Decalcified Nacreous Layer and Prismatic Layer.

The decalcified Nacreous Layer usually stains a uniform blue with Azan (Figure 5.98. a). The denser material on the bottom left of Figure 5.98. a, may be the aragonite Prismatic Layer seen in Figure 5.91. d.

The Inner Fibrous Sheath of the Prismatic Layer stains pink with Azan and appears as a sheet of material stretched over the medial surface of a Growth Scale (ifs in Figure 5.98. b). The thickening where the Transverse Parallel Side Walls meet the Inner Prismatic End Plates is visable through the Inner Fibrous Sheath (tp in Figure 5.98. b).

The Transverse Parallel Side Walls (tp in Figure 5.98.), parallel straps of scleroprotein which are very acid resistant, are thickened at the corners of the prisms where they join straps of the same material from adjacent side walls. They appears to demonstrate periodicity, and commonly surrounds remnants of blue staining Inner Prismatic Organic Matrices (ip in Figure 5.98. c and d).

FIGURE 5.98. *Pinctada margaritifera* - Decalcified Nacreous Layer and Prismatic Layer.

a. L.M. x 1200 Azan. Nacreous Layer.

b. L.M. x 1200 Azan. Inner Fibrous Sheath.

c. L.M. x 1200 Azan. Transverse Parallel Side Walls and Intra-Prismatic Organic Matrix.

d. L.M. x 1200 As for c above.

a
x1200

b
x1200

c
x1200

d
x1200

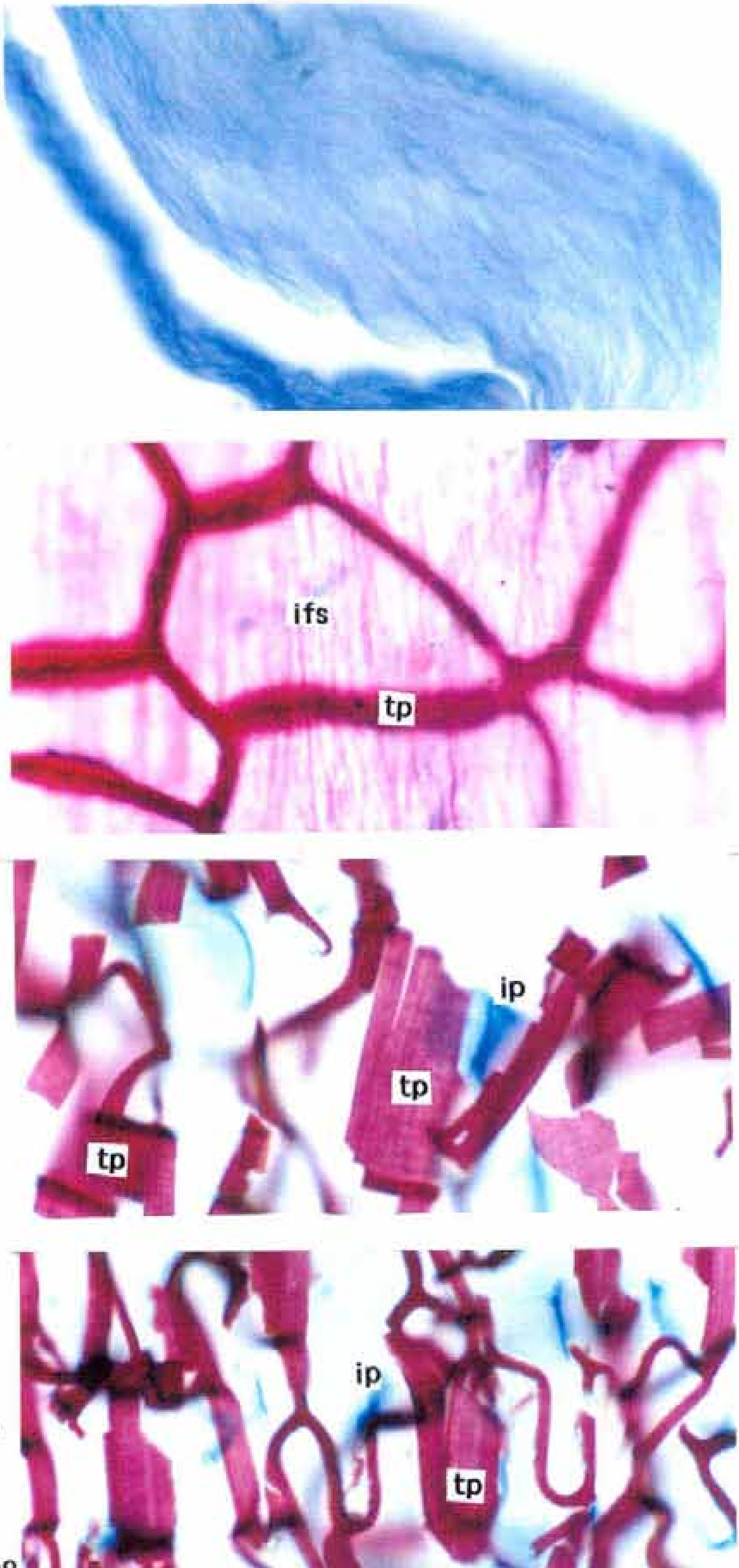


Figure5.98

5.5.2.5.3. Prismatic Layer.

Where there is no intervening Inner Fibrous Sheet of the more lateral, nor Outer fibrous Sheet of the more medial of two successive Growth Scales, the Inner Prismatic End Plates of the Major Prisms of the more Lateral may abut the Outer Prismatic End Plates of the Major Prisms of the more medial. This arrangement is illustrated in Figure 5.99.

The Outer Prismatic End Plates of Major Prisms (light pink sheets of material, opep in Figure 5.99. a, b, and c), are joined internally by the Organic Matrices of the Innter Lateral Structure (ils in Figure 5.99. a, b and c).

The thickening of the End Plates where joined by the Transverse Parallel Side Walls or the Inner Lateral Structure is quite marked (t in Figure 5.99. a, b c and d).

Figure 5.99. c is focussed on the junction of the Inner Lateral Structure (ils) with the Outer Prismatic End Plate (opep) of a Major Prism of the more medial Growth scale and the junction of the Transverse Parallel Side Wall Organic Matrix (tp) with the underlying Inner Presmatic End Plate (ipep) of a Major Prism of the more lateral Growth Scale is out of focus. The reverse is true in Figure 5.99. d.

Figure 5.99. e is a sagittal section through a Growth Scale showing normal (red) and aberrantly staining (blue) Transverse Parallel Side Wall Organic Matrices.

FIGURE 5.99. *Pinctada margaritifera* - Decalcified Prismatic Layer.

a. L.M. x 1200 Azan Outer Prismatic End Plates and Inner Lateral Structures.

b. L.M. x 1200 Azan as for a.

c. L.M. x 1200 Azan Outer and Inner Prismatic End Plates and Inner Lateral Structures.

d. L.M. x 1200 Azan as for c.

e. L.M. x 1200 Transverse Parallel Side Walls.

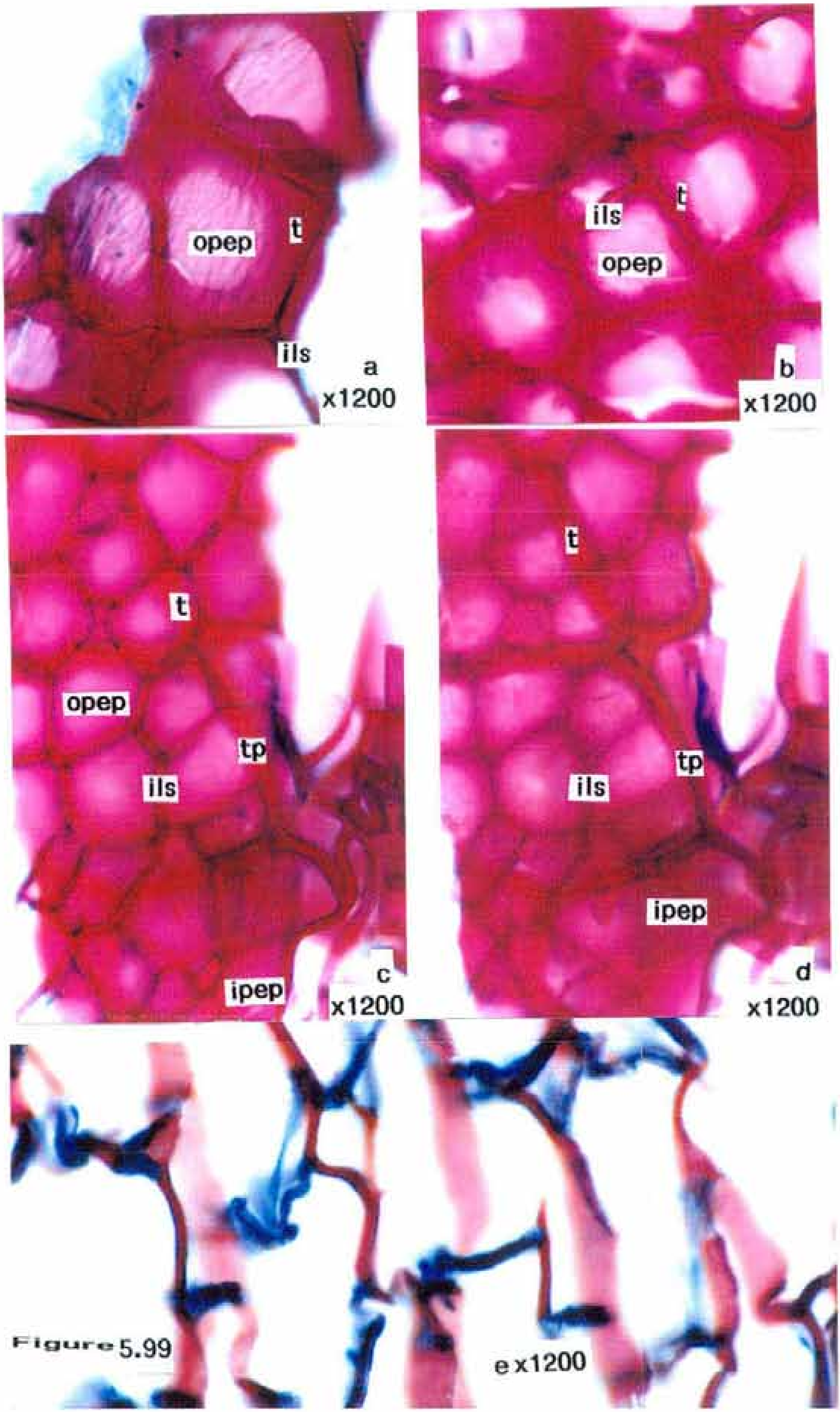

opep
t
ils
a
x1200
t
ils
opep
b
x1200
t
opep
tp
ils
ipep
c
x1200
t
tp
ils
ipep
d
x1200
e x1200

Figure 5.99

5.5.2.6. L.M. and T.E.M. of the External Mantle and Mantle Margins.

5.5.2.6.1. The Isthmusistic Epithelium.

The Isthmusistic Epithelium on the dorsal surface of the Isthmus is a very elongate columnar epithelium coextensive with the overlying Hinge. There is a deep medial sulcus (ms in Figure 5.100. e), and the tissue is bounded laterally by two lateral sulci.

The elongate cells have elongate ovoid nuclei slightly apical to the middle of the cell (n in Figure 5.100. a). The specimen illustrated in Figure 5.100. had areas of the medial surface of its Nacreous Layers covered with what appeared to be newly formed Nacreous Organic Matrix and this may explain the heavy concentration of small ovoid mitochondria in the apical cytoplasm of this tissue (mi in Figure 5.100. c, d and e), and the active secretion of vesicles from the apical microvilli (v and am respectively in Figure 5.100. b and c). Infrequently embedded in the Isthmustic Epithelium are relatively small Spherular Cytoplasm Turquoise Glands (sct in Figure 5.100. d).

FIGURE 5.100. *Pinctada margaritifera* –Isthmusistic Epithelium.

a. L.M. x 1200 Mallorys.

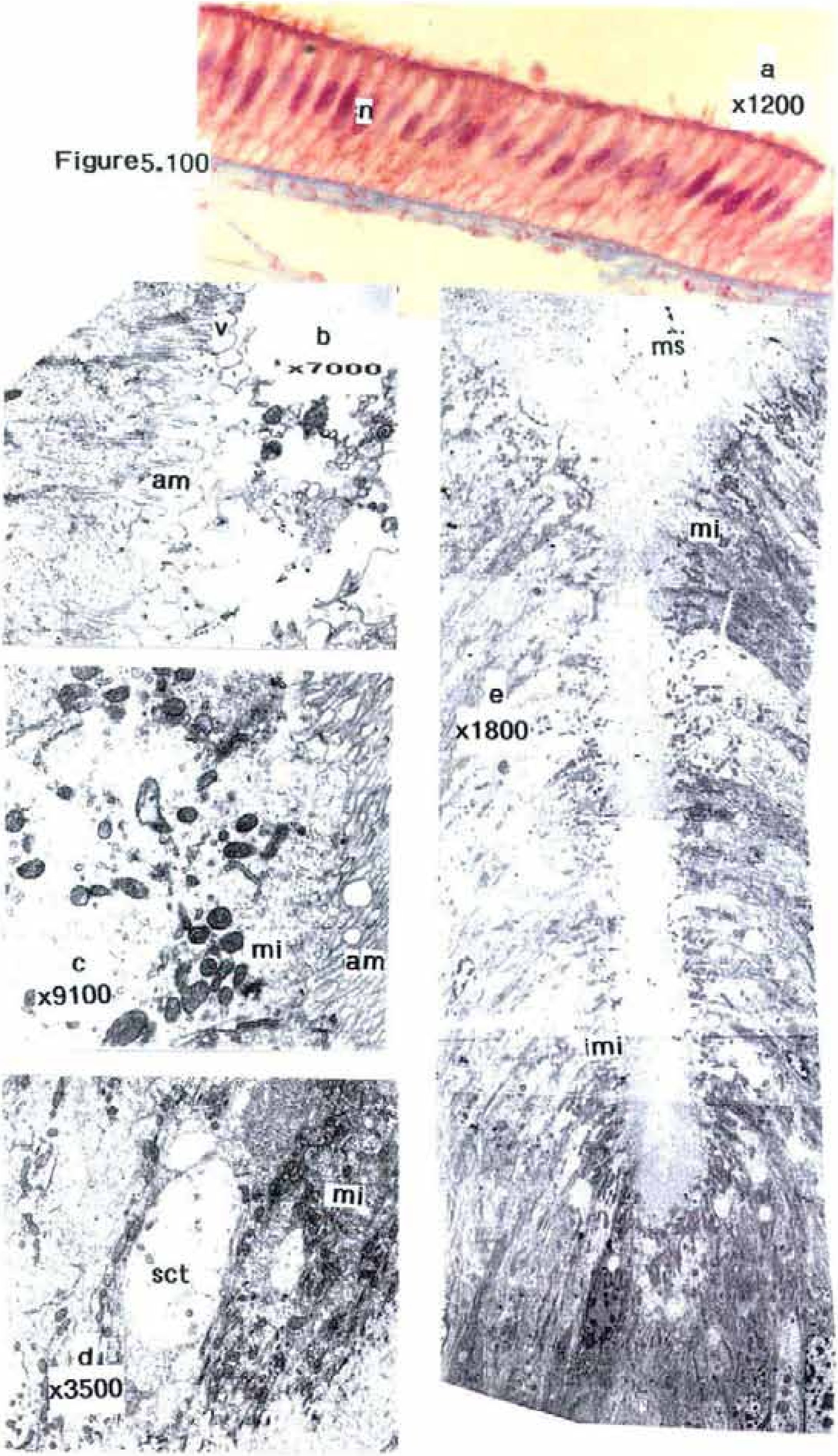
b. T.E.M. x 7000 Vesicular Secretion from Apical Microvilli.

c. T.E.M. x 9100 Mitochondria beneath Apical Microvilli.

d. T.E.M. x 3500 Spherular Cytoplasm Turquoise Gland.

e. T.E.M. x 1800 Isthmusistic Cells Lining the Median Sulcus.

Figure5.100


a
x1200
n
b
x7000
v
am
c
x9100
mi
am
d
x3500
mi
sct
e
x1800
ms
mi
imi

5.5.2.6.2. Shoulder Region.

This Region has a columnar epithelium with secretory apical microvilli, (am in Figure 5.101. c). The fibrous connective tissue layer which underlies the surface eptihelium (fct in Figure 5.101. a and b), grows much stronger going from near the Isthmus (Figure 5.101. a) to the centre of the Region (Figure 5.101. b).

Near the Isthmus there are a large number of Granular Cytoplasm Secretory Glands which stain amber wtih Mallorys whose bodies are situated in the middle depth of the Gland near the Trabecular Turquoise Glands (a and tt in Figure 5.101. a respectively).

The deepest layer of glands have granules which stain purple with Mallorys (p in Figure 5.101. a).

Midway between the Hinge and the Adductor Muscle, the Trabecular Turquoise Glands form a tissue beneath the subepithelial fibrous Connective Tissue and the two predominant types of Granular Cytoplasm Secretory Glands have granules which stain magenta and purple (m and p in Figure 5.101. b).

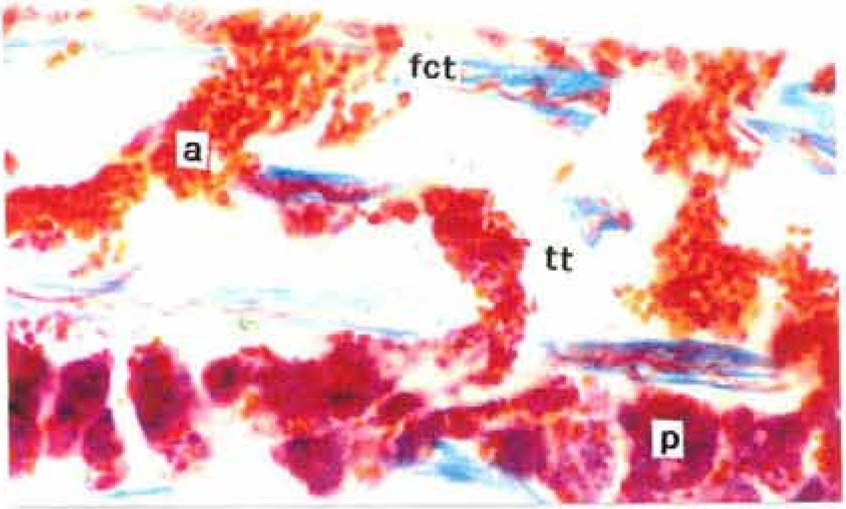
T.E.M. shows three types of Granular Cytoplasm Secretory Glands (g1, g2 and g3) beneath the tissue of Trabecular Turquoise Glands (tt in Figure 5.101. c).

FIGURE 5.101. *Pinctada margaritifera* - Shoulder Region.

a. L.M. x 770 Mallorys Shoulder Region near Isthmus.

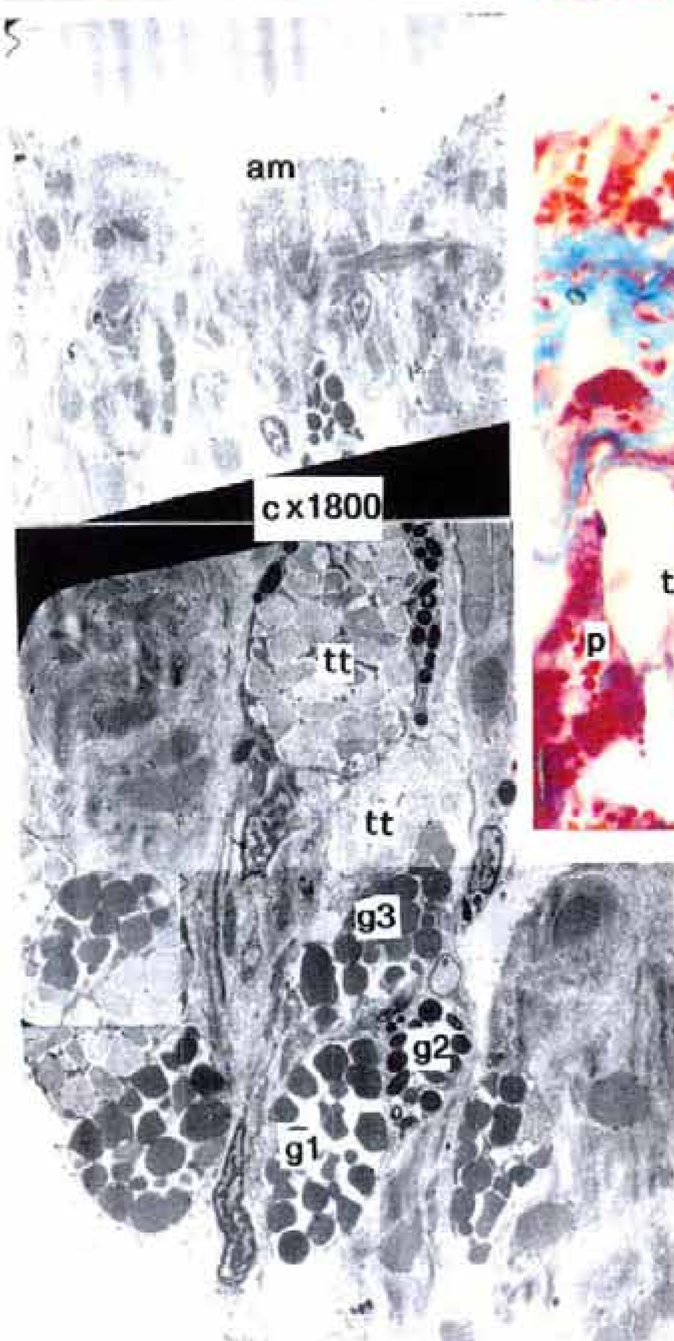
b. L.M. x 770 Mallorys Mid Shoulder Region.

c. T.E.M. x 1800 Mid Shoulder Region.



a x770

b x770



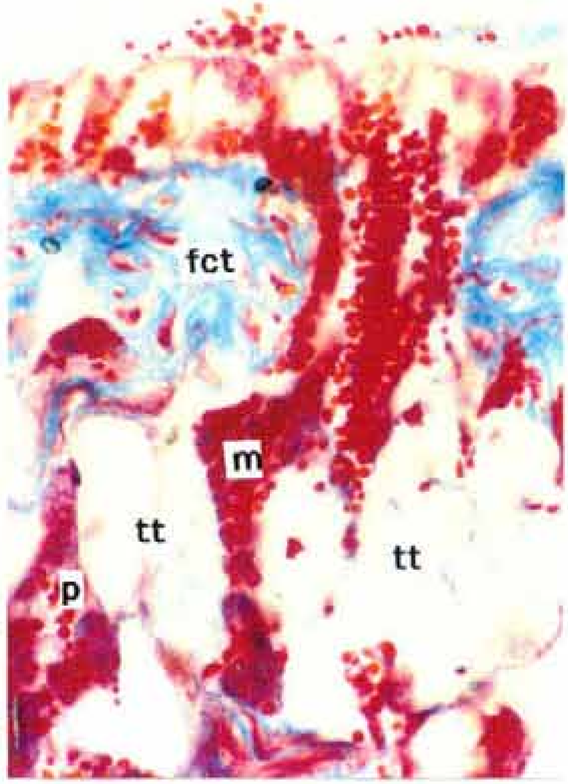


Figure 5.101

5.5.2.6.3. Anterior Pallial Gland and Pallial Mantle.

The Pallial Gland is similar to the Shoulder Gland in that the bodies of the Trabecular Turquoise Glands lie immediately beneath the subepithelial fibrous connective tissue and those of the Granular cytoplasm Secretory Glands are generally deeper. (tt and g respectively in Figure 5.102. a). The subepithelial connective tissue of the Pallial Gland is denser than that of the Shoulder Gland as is the intraglandular connective tissue. The Pallial Gland is about 2mm thick latero-medially. (Figure 5.102. a).

The Proximal Anterior Pallial Region is distinguished from the Middle Anterior Pallial Region by the greater depth of subcutaneous tissue bearing the unicellular secretory glands (Figure 5.101. b and c). The columnar cells of the Distal Anterior Pallial Region are more elongate with more strongly staining cytoplasm than those of the Middle Anterior Pallial Region and the bodies of the Trabecular Turquoise Glands are larger and deeper set in a more muscular subepithelium (Figure 5.102. c and d).

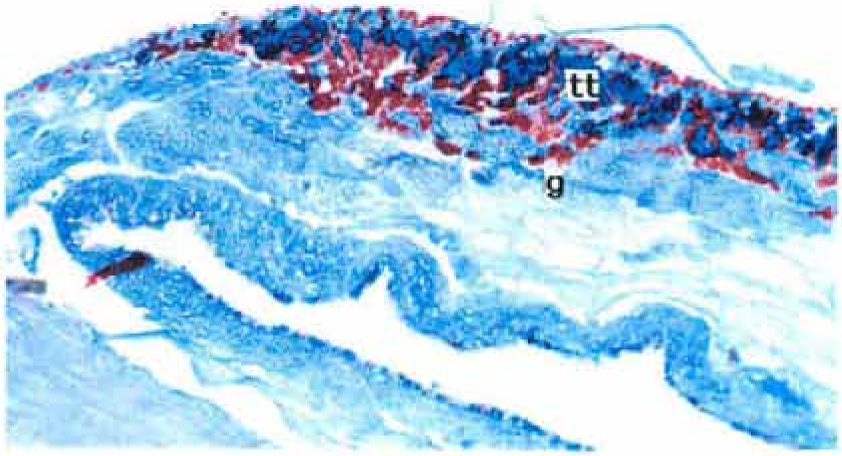
FIGURE 5.102. *Pinctada margaritifera* - Anterior Pallial Gland and Proximal, Middle and Distal Anterior Pallial Regions.

a. L.M. x 120 A.B./M.S.B. Anterior Pallial Gland.

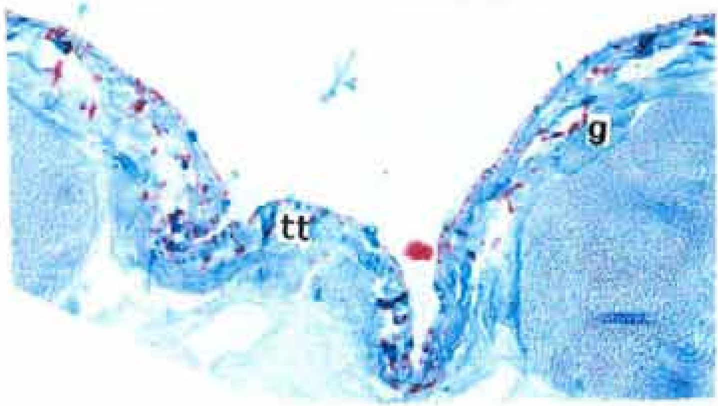
b. L.M. x 120 A.B./M.S.B. Proximal Anterior Pallial Region.

c. L.M. x 120 A.B./M.S.B. Middle Anterior Pallial Region.

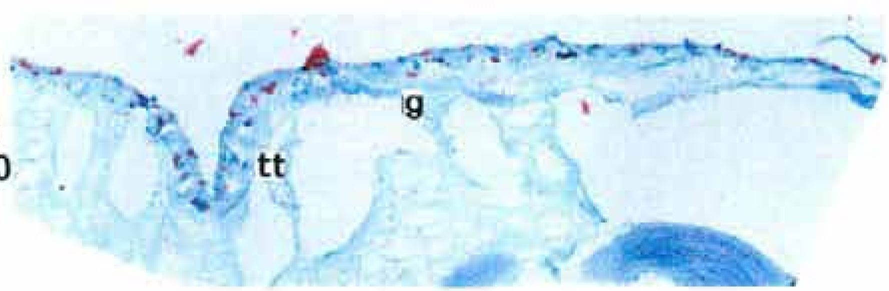
d. L.M. x 120 A.B./M.S.B. Distal Anterior Pallial Region.



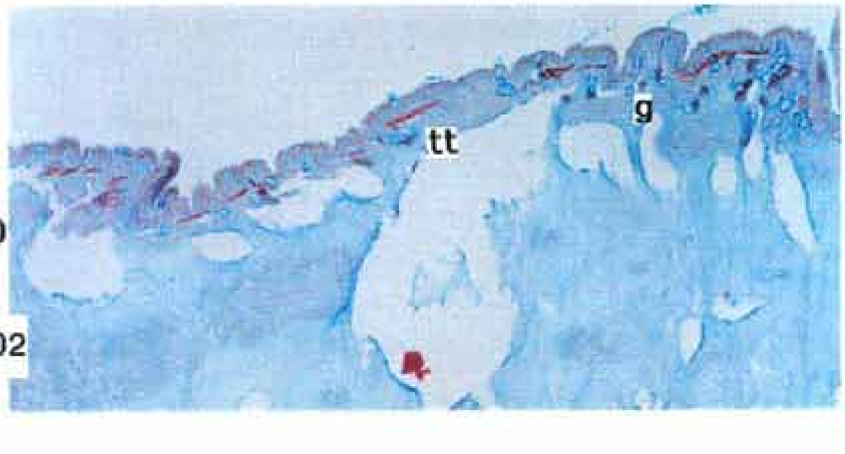
a
x120



b
x120



c
x120



d
x120

Figure 5.102

5.5.2.6.4. The Anterior Distal Folded Region and Tissue Regions of Folds F1. and F2..

The Distal Folded Region, (df in Figure 5.103. a), is here shown displaced distally. It is lateral to the Circum-pallial Nerve and Circum-pallial Sinus (cpn and cps in Figure 5.103. b).

In Figure 5.103. b, LF1, MF1, LF2 and MF2 signifiy LatF1., MedF1., LatF2., and MedF2. respectively.

The three tissue Regions of LatF1. going proximo-distally are Proximal LatF1., the Mantle Edge Gland and Terminal LatF1. (pl1, meg and t respectively in Figure 5.103. b).

The three tissue Regions of MedF2. going disto-proximally are Distal MedF1., Middle MedF1. and Proximal MedF1., (dm1, mm1, and pm1 respectively in Figure 5.103. b).

Groove F1F2. is the Marginal Mantle Groove between F1. and F2. (gF1F2. in Figure 5.103. b) and the Omega Gland occupies the lateral aspect of the Apex of Groove F1F2. (o and a12 respectively in Figure 5.103. b).

The three tissue Regions of LatF2. going proximo-distally are Proximal LatF2., Middle LatF2. and Distal LatF2., (pl2, ml2 and dl2 respectively in Figure 5.103. b).

The two tissue Regions of MedF2. are distal MedF2. and Proximal MedF2., (dm2 and pm2 respectively in Figure 5.103. b).

The Trabecular Turquoise glands of the type found from the Hinge to the Apical Region of Groove F1F2. (a12 in Figure 5.103. b) are designated tt1 and those of the type found between Apical Groove F1F2. and Distal LatF3. are tt2 in Figure 5.103. b.

FIGURE 5.103. *Pinctada margaritifera* - Anterior Distal Folded Region and Tissue Regions of Anterior Folds F1. and F2..

a. L.M. x 120 A.B./M.S.B. Distal Folded Region.

b. L.M. x 120 A.B./M.S.B. FoldF1. and FoldF2..

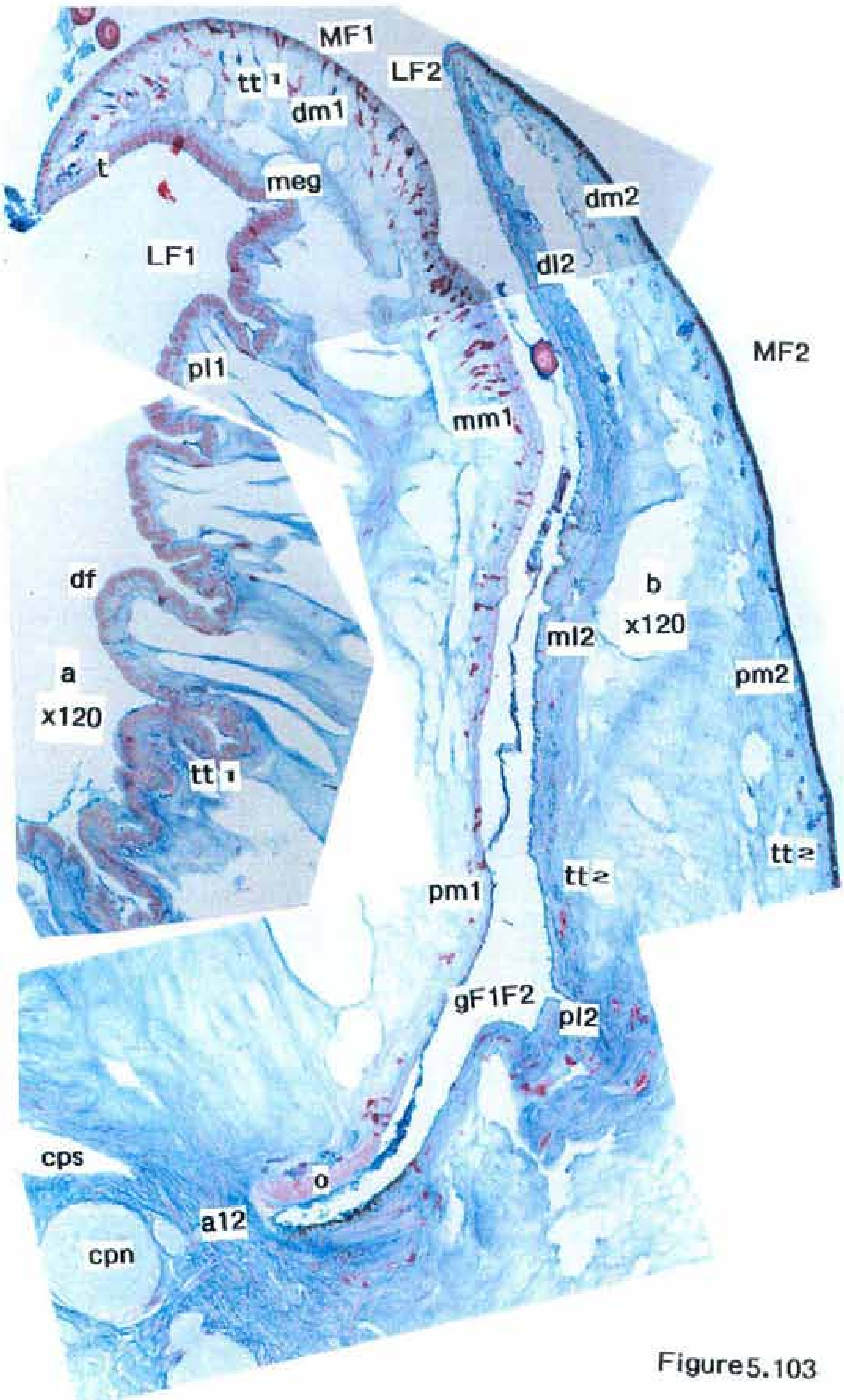

MF1
tt1
LF2
dm1
t
meg
dm2
LF1
dl2
MF2
pl1
mm1
df
b
x120
ml2
a
x120
pm2
tt1
tt2
tt2
pm1
gF1F2
pl2
cps
o
a12
cpn

Figure 5.103

5.5.2.6.5. Anterior Pallial Gland.

That Pallial Gland has a dense fibrous connective tissue (fc in Figure 5.104. a and b) both subepithelially, and providing a structure for the glandular cells.

The bodies of the Trabecular Turquoise Glands are clustered under the subepithelial conncective tissue (tt in Figure 5.105. a and b).

Deep to the Turquoise Glands are several types of Granular Cytoplasm Secretory Glands (g1 and g2 in Figure 5.104. b).

FIGURE 5.104. *P. margaritifera* Anterior Pallial Gland.

a. L.M. x 770 A.B./M.S.B. Epithelium, connective tissue and unicellular

glands.

b. T.E.M. x 1800 As for a above.

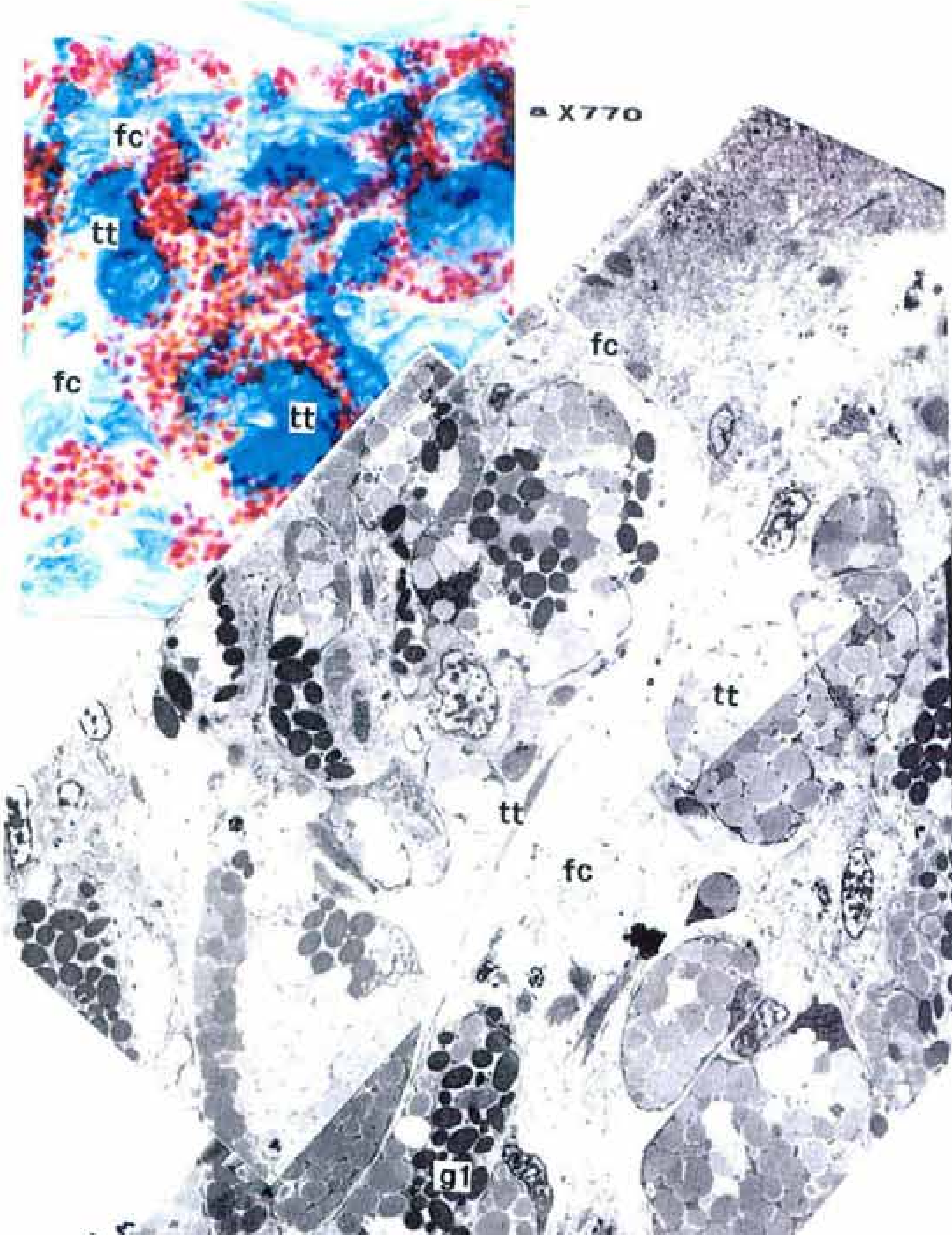


Figure 5.104

5.5.2.6.6. Anterior Proximal Pallial Region.

The surface epithelium has columnar cells with apical microvilli (am in Figure 5.105. a and c).

Some Trabecular Turquoise Glands appear to be intraepithelial while others have their cell bodies in the subepithelium (itt and stt respectively Figure 5.105. a, b and e).

At least two different kinds of Granular Cytoplasm Secretory Gland are illustrated (g1 and g2 in Figure 5.105. a, b d and e).

The cytoplasm of the Trabecular Turquoise Glands is clearly striate (Figure 5.105. b and d).

The surface epithelium bears cilia in the sulcus (c in Figure 5.105. e) and mitochondria in the apical cytoplasm (mi in Figure 5.105. e). Nerves with prominent elliptical neurosecretory granules lie in the connective tissue which carries muscle bundles in the subepithelium superficial to the deeper bodies of unicellular glands (n in Figure 5.105.a).

FIGURE 5.105. *Pinctada margaritifera* – Anterior Proximal Pallial Region.

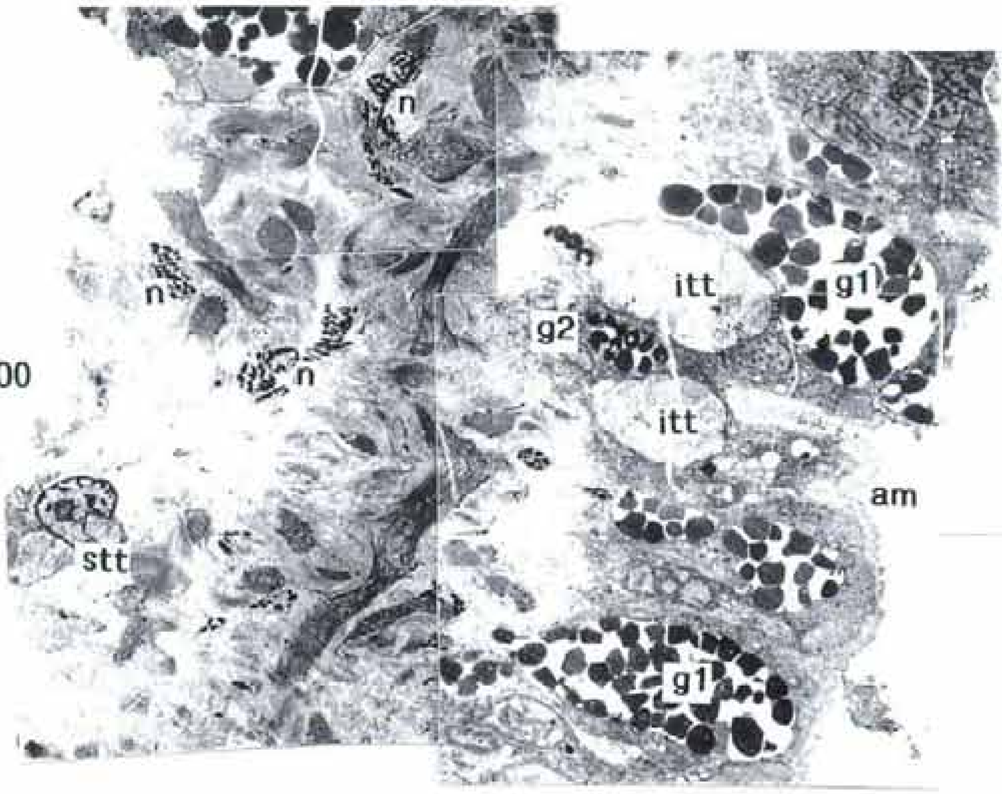
a. T.E.M. x 1800 Surface epithelium and intraepithelial and subepithelial secretory structures.

b. T.E.M. x 3500 Intraepithelial Secretory Glands.

c. T.E.M. x 12000 Ultrastructure of apical region of surface epithelium.

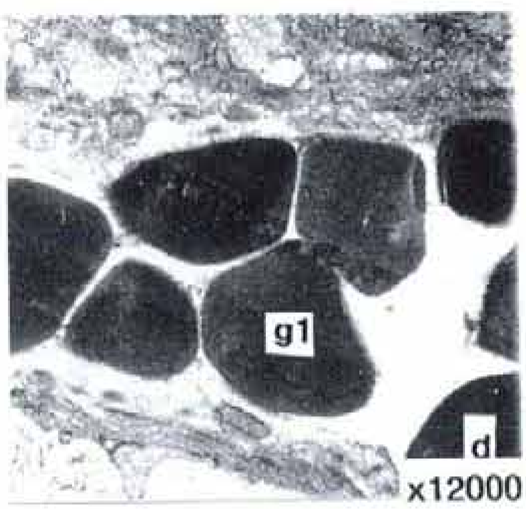
d. T.E.M. x 12000 Higher magnification of a gland in b above.

e. T.E.M. x 12000 Higher magnification of a different gland in b above.



a

x1800



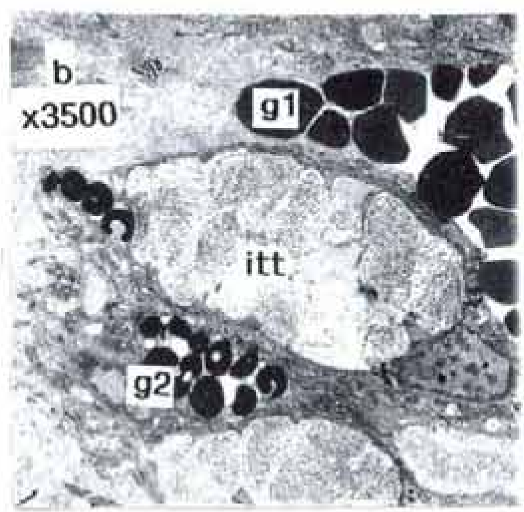
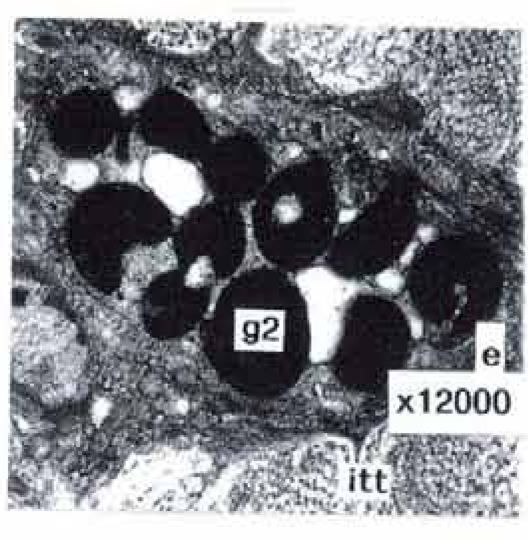
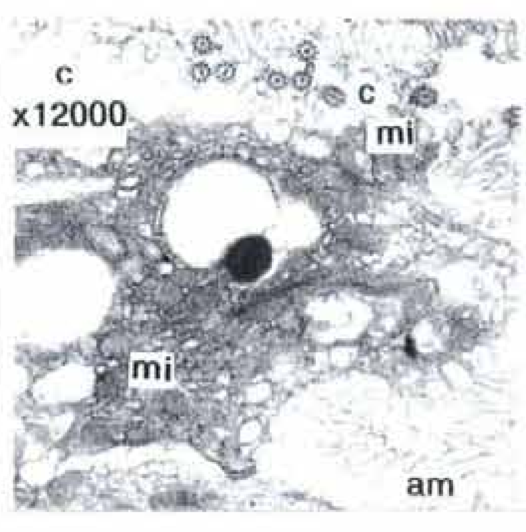


Figure 5.105





5.5.2.6.7. Anterior Distal Folded Region.

The chief characteristics of this Region are the relatively large outfoldings of the epithelium and subepithelium and the pattern of elongate subepithelial sinus lying normal to the epithelial surface. (s in Figure 5.106. a, b and c).

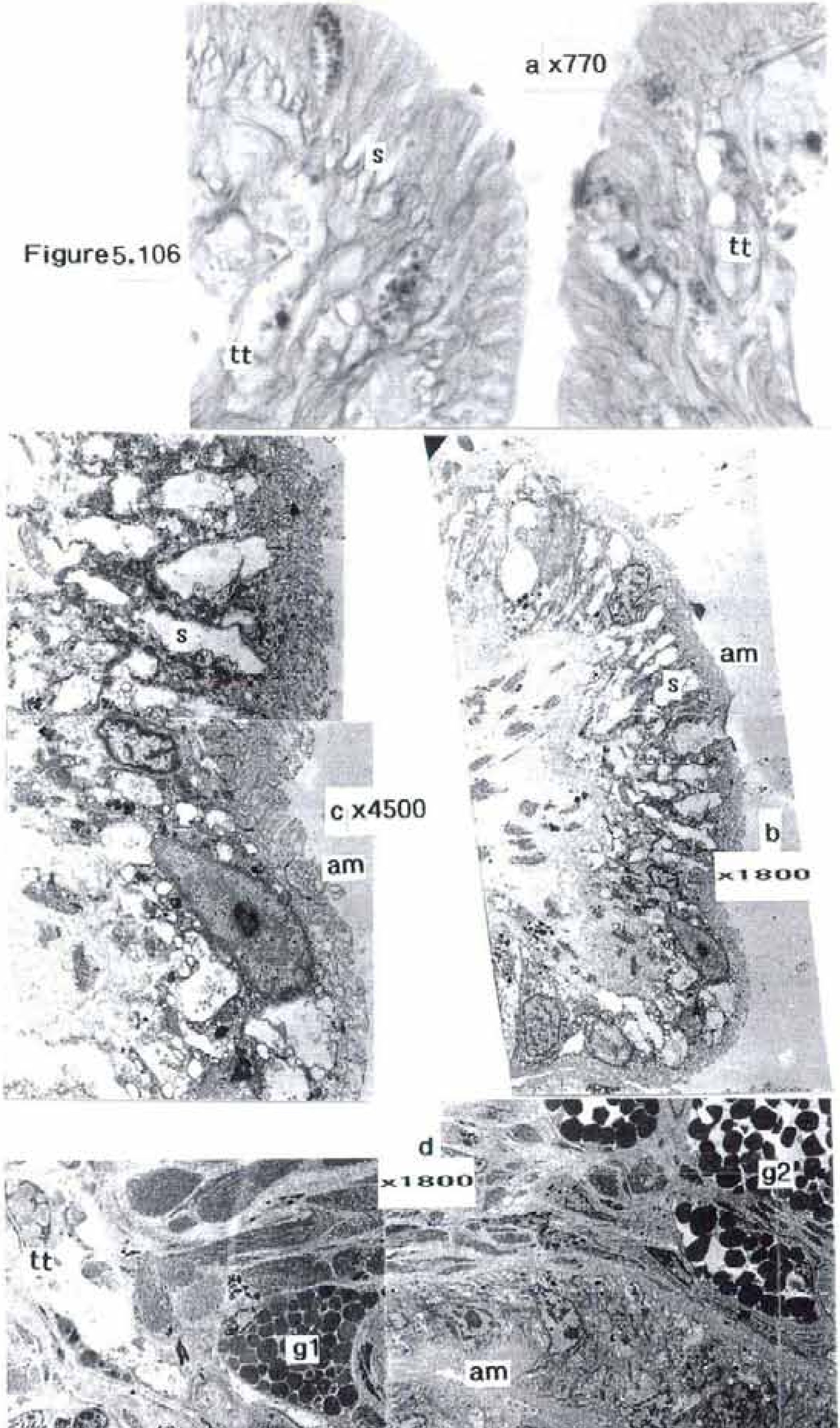
There is a quite dense population of both Trabecular Turquoise Glands (tt in Figure 5.106. a and d), and a variety of Granular Cytoplasm Secretory Glands (g1 and g2 in Figure 5.106. d).

The surface epithelium bears secretory apical microvilli (am in Figure 5.106. b, c and d).

FIGURE 5.106. *Pinctada margaritifera* – Anterior Distal Folded Region.

a.	L.M. x 770 A.B./M.S.B.	Surface epithelium and subepithelial secretory glands and sinus.
b.	T.E.M. x 1800	Surface epithelium with apical microvilli, subepithelial glands and sinus.
c.	T.E.M. x 4500	As for b above at higher magnification.
d.	T.E.M. x 1800	Surface epithelium and subepithelial secretory glands around an infolding.

Figure5.106



5.5.2.6.8. Mantle Margin Regions.

5.5.2.6.8.1. Anterior LatF1..

This is divided into three histological Regions - Proximal LatF1., Mantle Edge Gland, and Terminal LatF1..

Proximal LatF1. has lower foldings than the Distal Folded Region and lacks the specialised subepithelial sinus of the latter but has a similar relatively heavy population of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands (pLl in Figure 5.103. b).

The Mantle Edge Gland is a specialised elongate columnar epithelium (meg in Figure 5.107. a and b) with a dense mat of secretory apical microvilli (am in Figure 5.107. a) and a system of small sinus beneath the pronounced basement membrane (s and bm in Figure 5.107 b). There is a conspicuous shortage of unicellular secretory glands compared with any other Region but the Isthmus on the External Mantle and Mantle Margin. Vesicles from the extremities of the apical microvilli coalesce to form a lamina secretion which is very electron dense (ls in Figure 5.97a) and stains turquoise with A.B./M.S.B. (ls in Figure 5.107. b).

Terminal LatF1. has a lower (than the Mantle Edge Gland), but still elongate columnar epithelium and lacks the system of subepithelial sinus. The unicellular secretory glands of Terminal F1. underlie this surface epithelium but mostly secrete through that of Distal MedF1. and will therefore be described in detail with that Region. They include Ovoid Blue Glands and distal diffuse glands (ob and dd in Figure 5.107. a and b).

FIGURE 5.107. *Pinctada margaritifera* - Anterior Mantle Edge Gland and Terminal LatF1..

a. T.E.M. x 18000 Apical Mantle Edge Gland

b. L.M. x 770 A.B./M.S.B. Mantle Edge Gland.

c. L.M. x 770 A.B./M.S.B. Terminal F1..

d. L.M. x 777 A.B./M.S.B. Terminal F1..

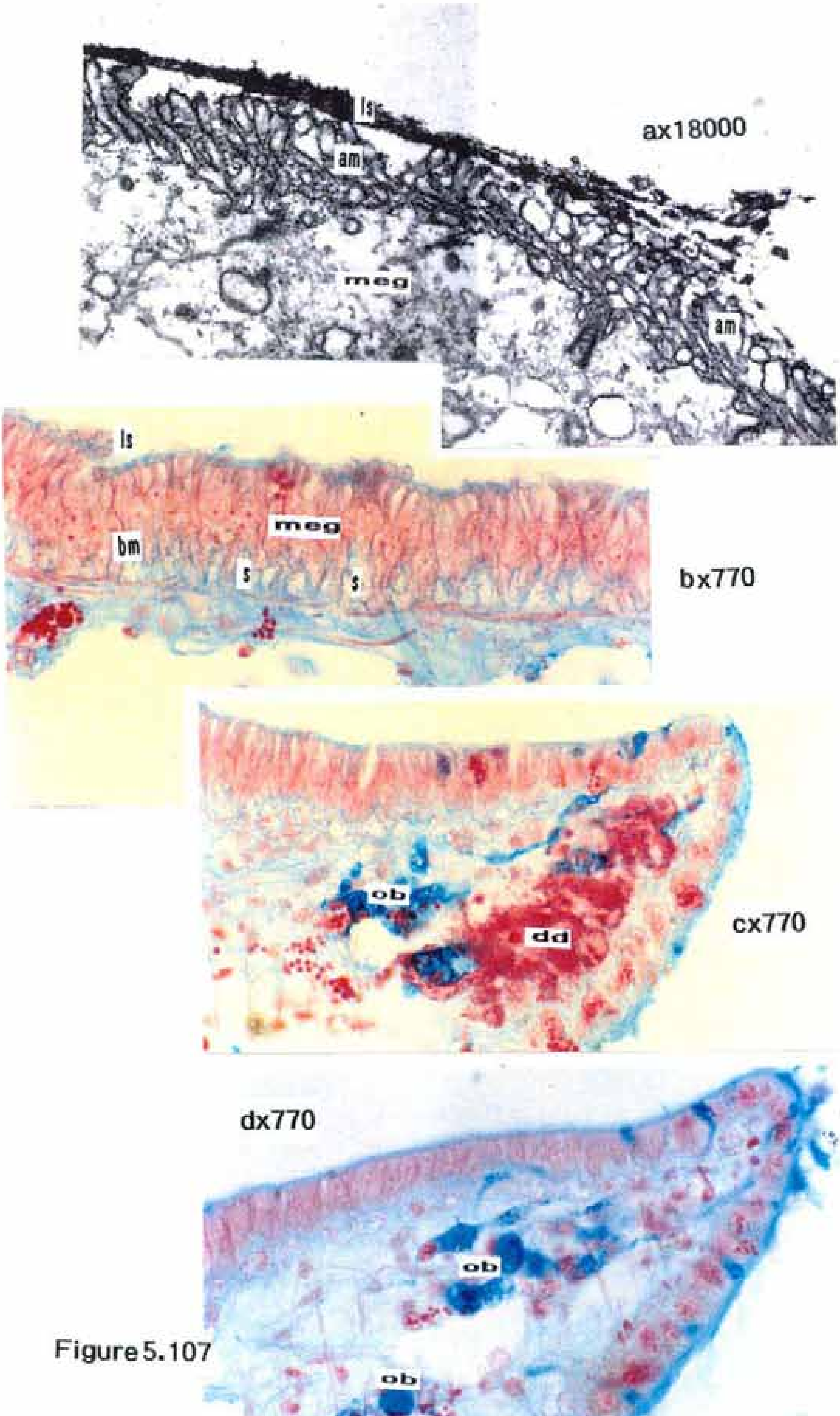


Figure 5.107

5.5.2.6.8.2. Anterior and Byssal Embayment Distal and Middle MedF1..

The Anterior Distal MedF1. has a low columnar surface epethelium with occassional pigmented cells, (p in Figure 5.108. a).

There are a number of Granular Cytoplasm Secretory Glands in the subepithelium (g in Figure 5.108. a). Almost unique to this location are the Ovoid Blue Glands and Distal Diffuse Glands (ob and dd respectively in Figure 5.107. c and d and 5.108. a).

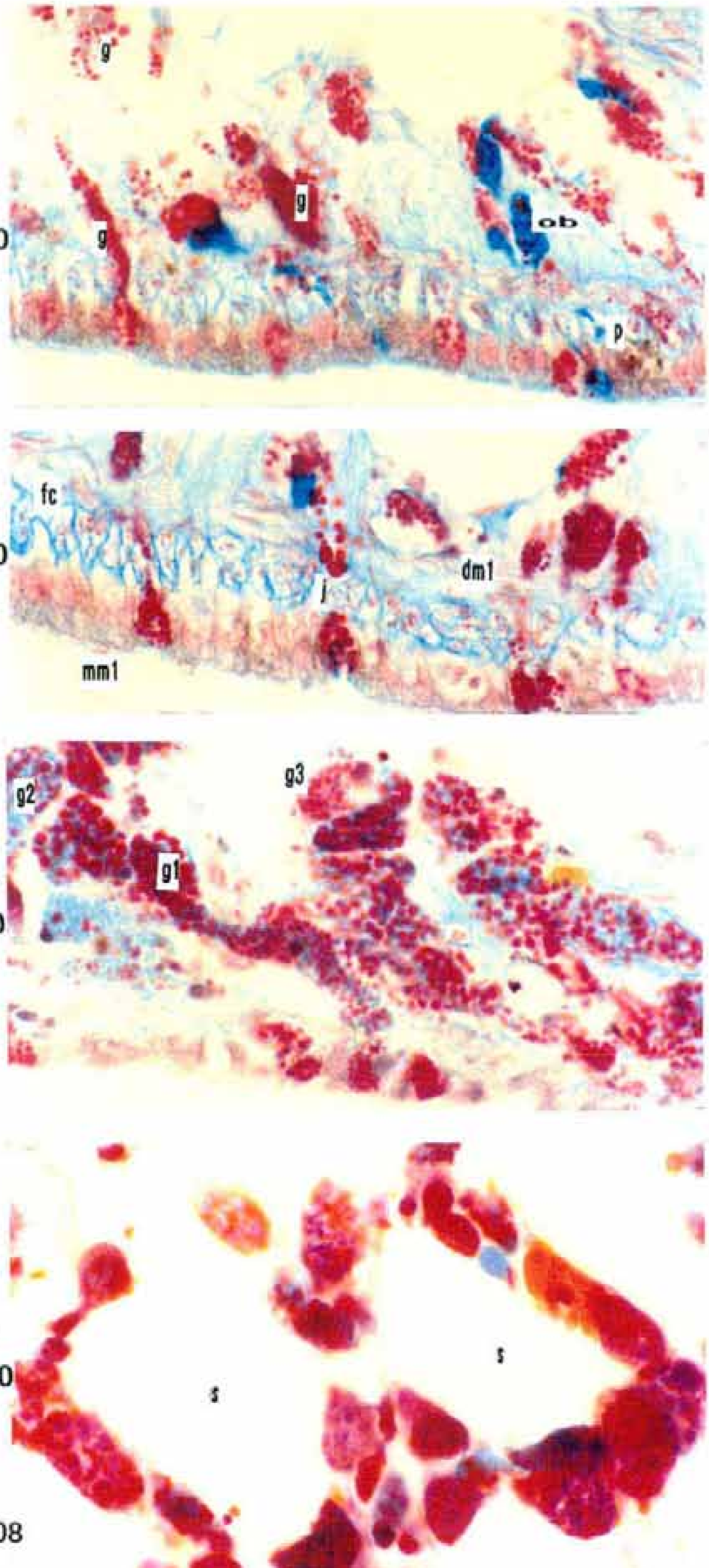
The junction (j) of the Distal MedF1. and Middle MedF1. (dm1 and mm1 respectively) is illustrated in Figure 5.108. b. The latter has a unique arrangement of subepithelial fibrous connective tissue and muscles, (fc in Figure 5.108. b).

In the Byssal Embayment there is a quite different array of Granular Cytoplasm Secretory Glands from those seen anteriorly (g1, g2 and g3 in Figure 5.108. c).

Figure 5.108. d illustrates the relationship between the cell bodies of a variety of Granular Cytoplasm Secretory Glands to each other and to the parenchymal sinus (s in Figure 5.108. d) in the Anterior Distal MedF1..

FIGURE 5.108. *Pinctada. margaritifera* –Anterior and Byssal Embayment Distal and Middle MedF1..

a. L.M. x 770 A.B./M.S.B. Distal MedF1..
b. L.M. x 770 A.B./M.S.B. Junction of Distal and Middle MedF1..
c. L.M. x 770 Mallorys. Byssal Embayment Distal MedF1..
d. L.M. x 1200 Mallorys. Sagittal Section, subepithelial Anterior Distal Med.F1..



a
x770

b
x770

c
x770

d
x1200

Figure5.108

5.5.2.6.8.3. Anterior Proximal MedF1..

The Anterior Proximal MedF1. surface epithelium is an elongate columnar epithelium relative to the other regions of MedF1. and has apical microvilli.

The subepithelial Granular Cytoplasm Secretory Glands are predominantly of two kinds - one with a cytoplasm consisting mostly of granules which stain red with A.B./M.S.B. and the other with red granules in a blue cytoplasmic matrix with A.B./M.S.B. (g1 and g2 respectively in Figure 5.109. a and b).

The Pleated Secretion of Groove F1F2. (ps in Figure 5.109. a, b and c) initially stains turquoise with A.B./M.S.B. but usually changes its staining affinity distally. It is a continuous sheet of material and hence forms a complete barrier between the materials secreted on either side of it (Figure 5.109. c).

FIGURE 5.109. *Pinctada margaritifera* - Anterior Proximal MedF1..

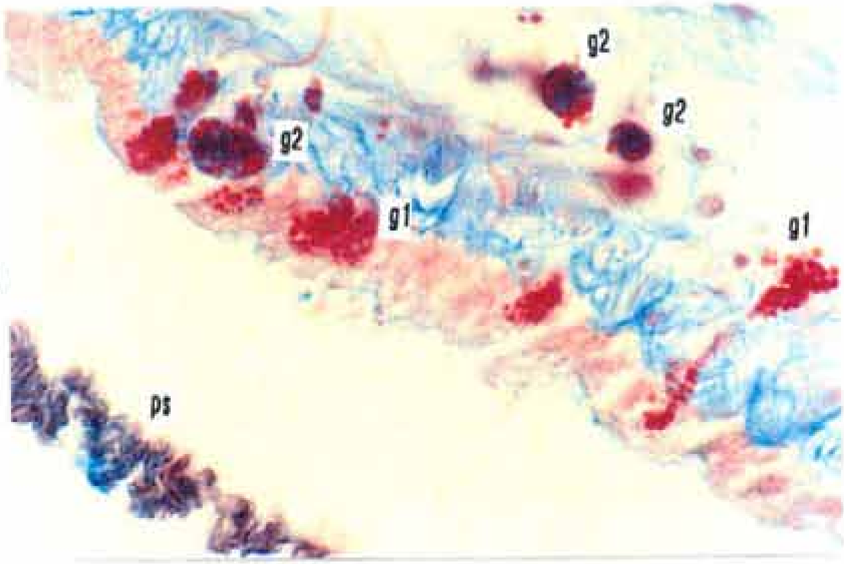
a. L.M. x 770 A.B./M.S.B. Surface epithelium, subepithelial glands and Pleated Secretion of Groove F1F2.

b. L.M. x 770 A.B./M.S.B. As for a above.

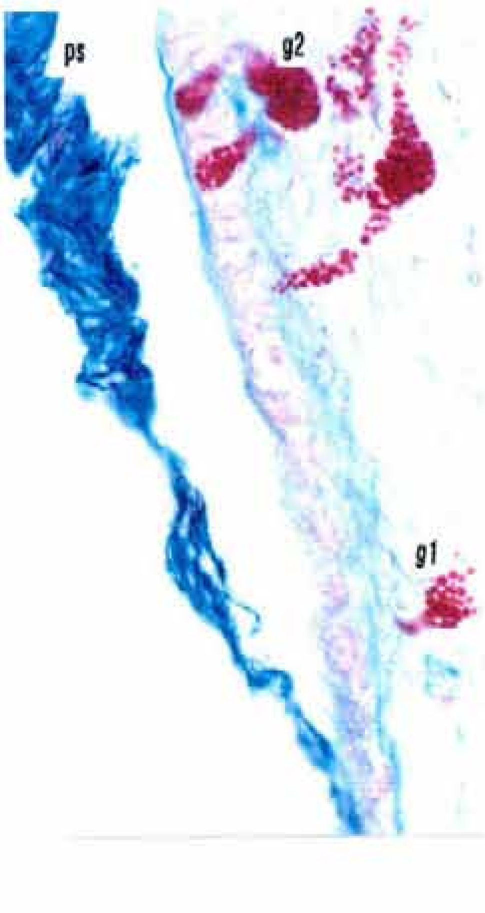
c. T.E.M. x 1800 As for a and b above.

a

x770



b x770



c x1800

Figure 5.109

5.5.2.6.8.4. Anterior Apical Groove F1F2..

In Figure 5.110, L is lateral and M is medial.

The Pleated Secretion of Groove F1F2. (ps in Figure 5.110. a and b) issues from the Apical Channel (ac in Figure 5.110. a and b). Lateral to the Apical Channel are the Distal and Proximal parts of the Omega Glands (do and po respectively in Figure 5.110. a and b). The Proximal Part of the Omega Gland comprises Celtic Scroll Cells superficially. (cs in Figure 5.110). and deeper, the Fenestrated Cells and the Multivesiculate Cells. (fc and mc respectively in Figure 5.110)..

Medial to the apical Channel are proximally, the Dactylocytes with even length strong microvilli medially bordering the Apical Channel, and distal to them the Black Granule Secretory Cells, (d and bgsc respectively in Figure 5.110.).

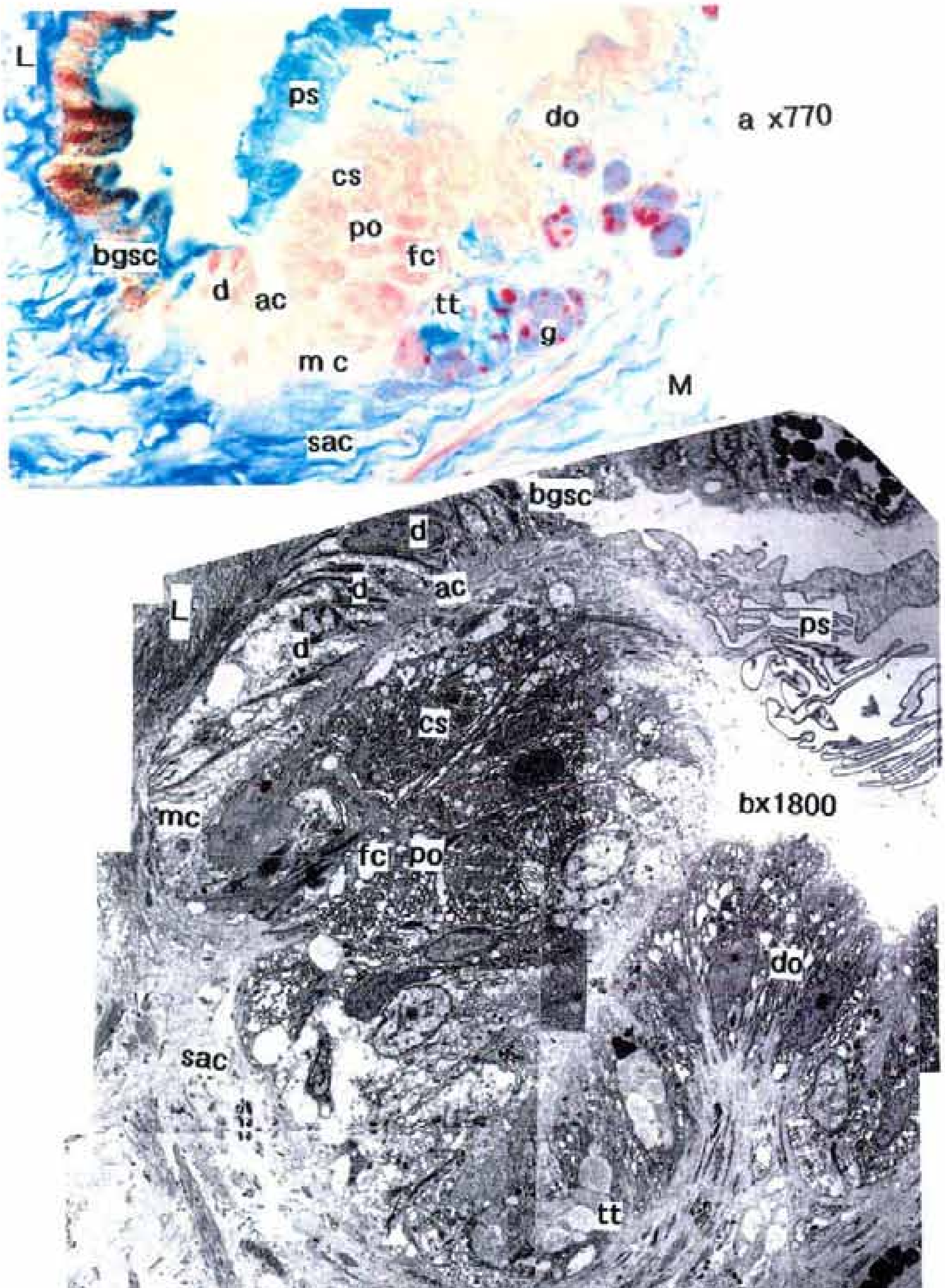
Beneath the Omega Gland is the dense Subapical Connective Tissue (sac in Figure 5.110. a and b).

Between the two parts of the Omega Gland are the Unicellular Glands of the Omega Gland - Trabecular Turquoise Glands and or Granular Cytoplasm Secretory Glands depending on location, (tt and g in Figure 5.110. a and b).

FIGURE 5.110. *Pinctada margaritifera* Anterior Apical Groove F1F2..

a. L.M. x 770 A.B./M.S.B. Omega Gland, Dactylocytes and other specialised secretory structures of Apical groove F1F2.

b. T.E.M. x 1800 As for a above.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tt
M

Figure 5.110

5.5.2.6.8.5. Cells of the Omega Gland and the Origin of the Pleated Secretion of Groove F1F2..

The Pleated Secretion of Groove F1F2. originates at the proximal end of the Apical Channel (pe in Figure 5.111. a and c).

At this point, the most proximal Dactylocyte joins the Multivesiculate Cell (pdc and mc respectively in Figure 5.111. a and c) and the Amorphous Membrane Bound Secretion elaborated by the Fenestrated Cells (mbs and fc respectively in Figure 5.111. a, b and c).

The Multivesiculate Cell contributes two distinct types of Vesicles to the location where the Pleated Secretion originates - a hollow visicle with internal adherances to its surrounding membrane (hv in Figure 5.111. a and c) and a multi membranous vesicle which appears to be synthesized in the vicinity of the Amorphous Membrane Bound Secretion of the Fenestrated Cell (mm in Figures 5.111. a, b and c).

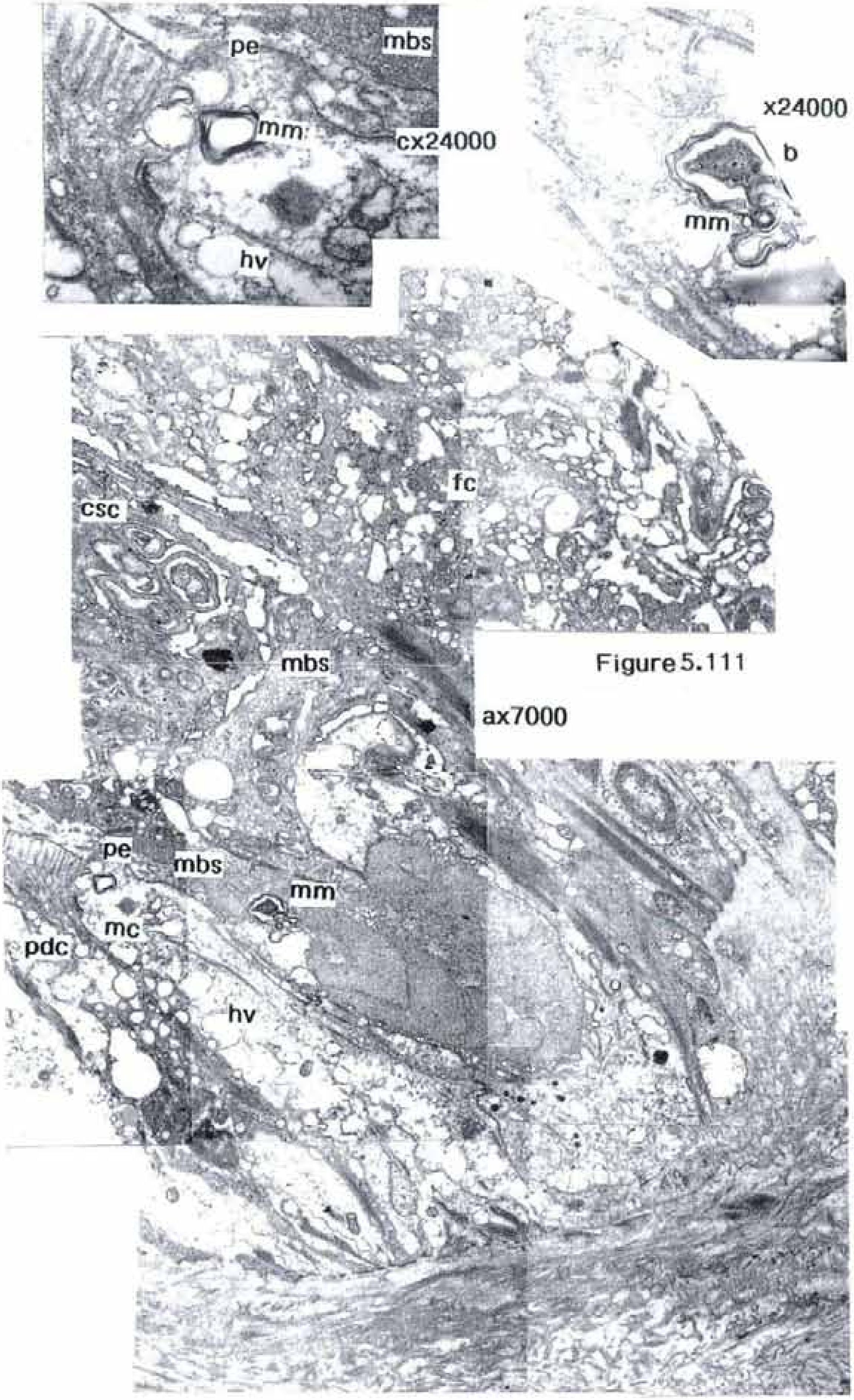
The Multivesiculate Cells and the Fenestrated Cells form the inner layer of the Proimal Part of the Omega Gland. The Outer Layer of the Omega Gland is occupied by the Celtic Scroll Cells (csc in Figure 5.111. a).

FIGURE 5.111. *Pinctada margaritifera* - Cells of the Anterior Omega Gland and site of origin of Pleated Secretion of Groove F1F2..

a. T.E.M. x 7000 Celtic Scroll Cell, Fenestrated Cells, Multivesiculate Cell, Dactylocyte and Amorphous Secretion at proximal end of the Apical Channel of Groove F1F2.

b. T.E.M. x 24000 Higher magnification of part of a.

c. T.E.M. x 24000 Higher magnification of part of a.



pe
mbs
mm
cx24000
hv
x24000
b
mm
fc
csc
mbs
ax7000
pe
mbs
mm
mc
pdc
hv

Figure 5.111

5.5.2.6.8.6. Distal Anterior Apical Channel of Groove F1F2..

The forming Pleated Secretion of Groove F1F2. lies in the Apical Channel between the Celtic Scroll Cells of the Omega Gland laterally and the microvilli of the Dactylocytes medially (cs and dc respectively in Figure 5.112.).

The Dactylocytes contribute electron dense material to the forming Pleated Secretion via their apical microvilli (ed, Figure 5.112.).

The Celtic Scroll Cells produce large membrane bound vesicles which are secreted into proximal Groove F1F2. lateral to the Pleated Secretion of Groove F1F2..

The Black Granule Secretory Cells are distal to the Dactylocytes (ie on Proximal LatF2.), and synthesize electron dense granules which are matured in vesicles (bg in Figure 5.112).

FIGURE 5.112. *Pinctada margaritifera* – Anterior Apical Channel, Celtic Scroll Cells, Dactylocytes and Black Granule Secretory Cells.
T.E.M. x 7000 Distal Anterior Apical Channel.

Figure5.112

x7000

5.5.2.6.8.7. Byssal Embayment and Anterior Proximal LatF2..

The Proximal LatF2. subepithelium of the Byssal Embayment has a massive concentration of several species of Granular Cytoplasm Secretory Glands (g1, g2, g3 of Figure 5.113 a and b) which secrete into Receptacle Glands (r.g. Figure 5.103 a and b) in the Proximal LatF2. epithelium.

The concentration of subepithelial unicellular glands in Proximal LatF2. is greatly decreased compared with the above at the Anterior Mantle Margin (Figure 5.113. c).

Light Blue Glands are mixed with the other Granular Cytoplasm Secretory Glands of the Proximal LatF2. subepithelium but secrete through the Middle LatF2. surface epithelium (lb in Figure 5.113. d).

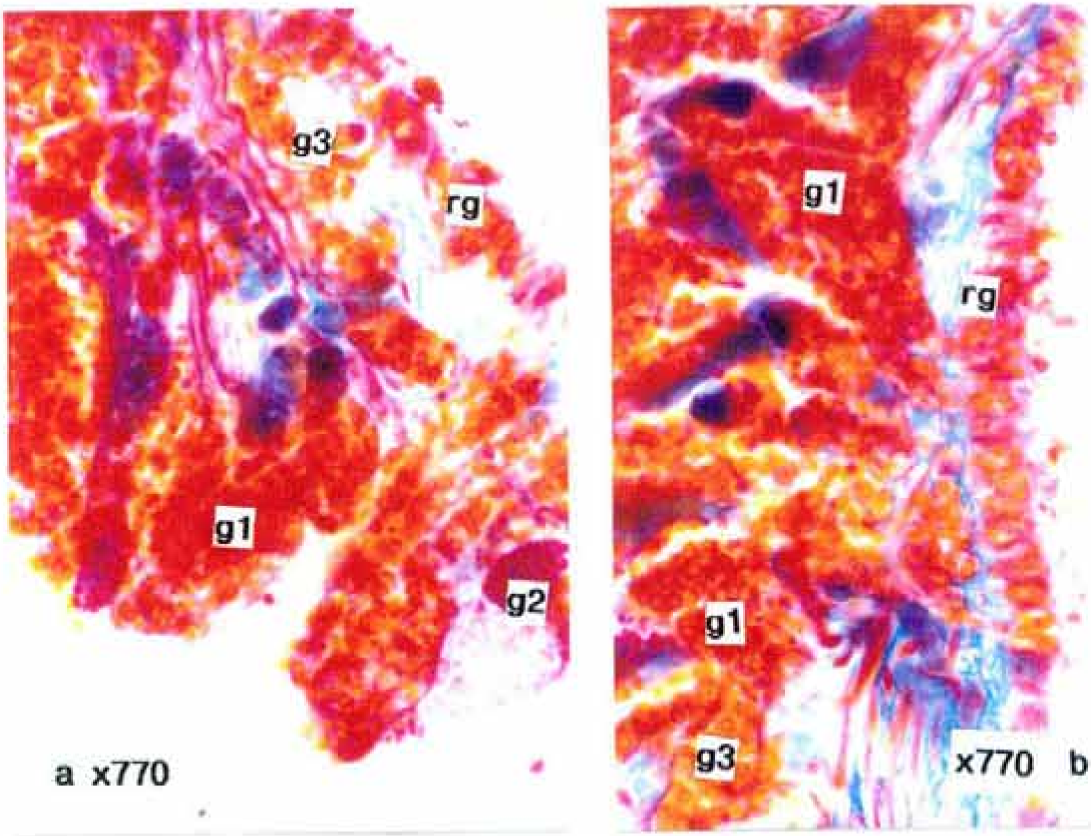
FIGURE 5.113. *Pinctada margaritifera* Anterior Proximal LatF2. and Byssal Embayment Proximal LatF2..

a. L.M. x 770 Mallorys Byssal Embayment.

b. L.M. x 770 Malllorys Byssal Embayment.

c. L.M. x 770 A.B./M.S.B. Anterior Mantle Margin.

d. L.M. x 770 A.B./M.S.B. Anterior Mantle Margin.



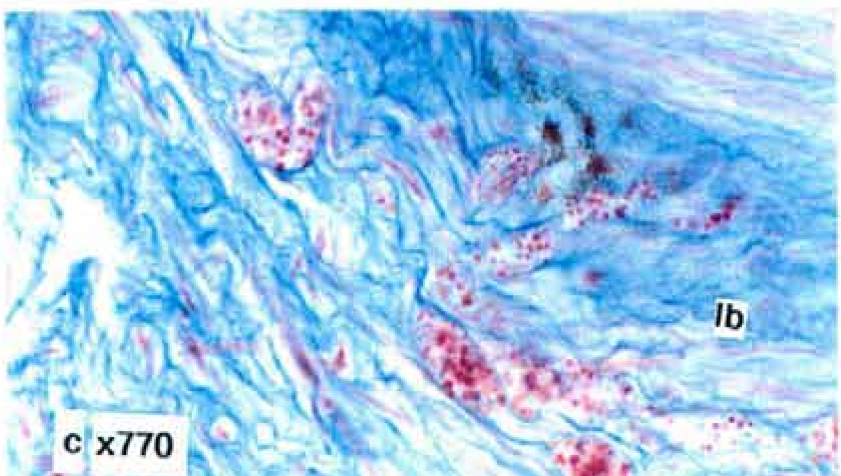
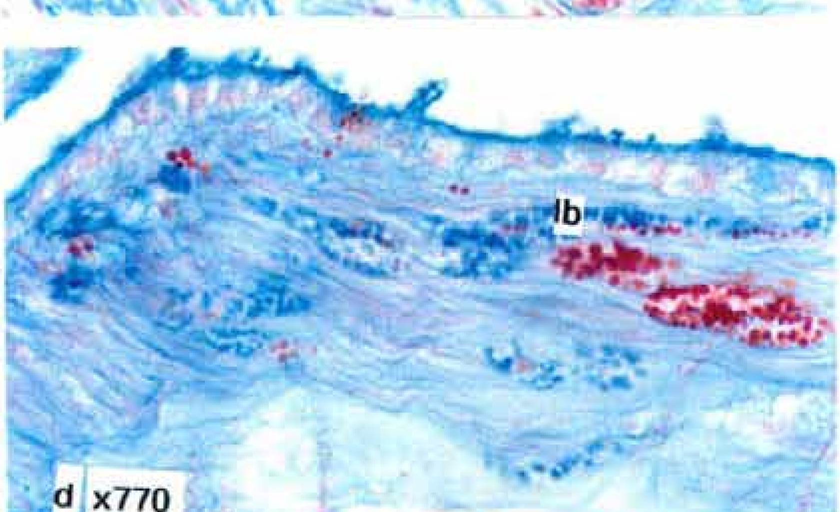


Figure5.113



5.5.2.6.8.8. Anterior Middle and Distal LatF2. and Terminal F2., Distal and Proximal MedF2..

A type of Trabecular Turquoise Gland distinct from that which occurs on the External Mantle and on F1. is common to all Regions of F2. and LatF3., (tt2 in Figure 5.114. b, c, d and e).

The dominant subepithelial unicellular Gland of Middle Lat F2. is the Light Blue Gland (lb of Figure 5.114. a and b).

The surface epithelium of the Middle LatF2. Region is similar throughout the *Pinctada* and is a heavily ciliated elongate columnar epithelium with basal nuclei (se in Figure 5.114. a). It exhibits the semicircular infoldings separated by flat plateaux typical of this Region (si in Figure 5.114. a).

The surface epithelium of Medial LatF2 is heavily pigmented. With A.B./M.S.B. the surface epithelial pigment granules of Distal MedF2. stain brown and those of Proximal MedF2 stain largely a tan colour. (Figure 555.114. d and e respectively).

FIGURE 5.114. *Pinctada margaritifera* – Anterior Middle and Distal LatF2., Terminal F2. and Distal and Proximal MedF2..

a. L.M. x 770 A.B./M.S.B. MiddleLatF2.

b. L.M. x 770 A.B./M.S.B. Distal LatF2.

c. L.M. x 770 A.B./M.S.B. Terminal F2.

d. L.M. x 770 A.B./M.S.B. Distal MedF2.

e. L.M. x 770 A.B./M.S.B. Proximal MedF2..

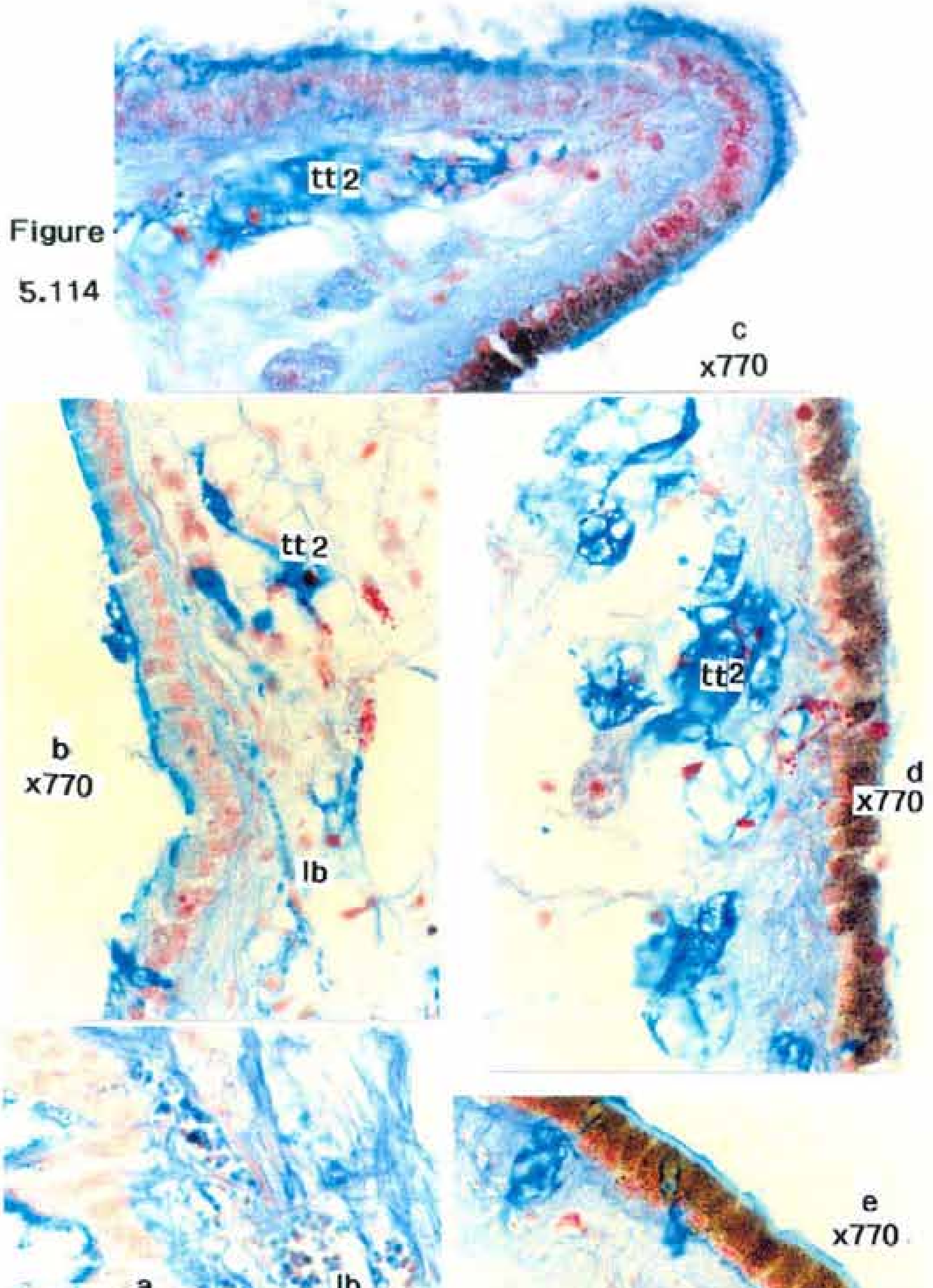


Figure
5.114

5.5.2.6.8.9. Anterior Proximal and distal LatF3. and MedF3.

Both regions of LatF3. have a columnar surface epithelium with basal nuclei and pigmented apical cytoplasm - that of the proximal MedF3. has granules which largely stain tan with A.B./M.S.B. and that of the Distal MedF3. granules which exclusively stain a greyish colour with this stain (Figure 5.115, a and b respectively).

The subepithelial unicellular glands are largely the F2. LatF3. type of Trabecular Turquoise Glands (tt2 in Figure 5.115, a and b).

The medial surface of F3. is covered with a mucosa which is very different from any of those described on the External Mantle and Mantle Margin. A major difference is that the Trabecular Turquoise Glands and the Granular Cytoplasm Secretory Glands are invariably intraepithelial. (Figure 5.115, c).

FIGURE 5.115. *Pinctada margaritifera* L.M. LatF3., MedF3..

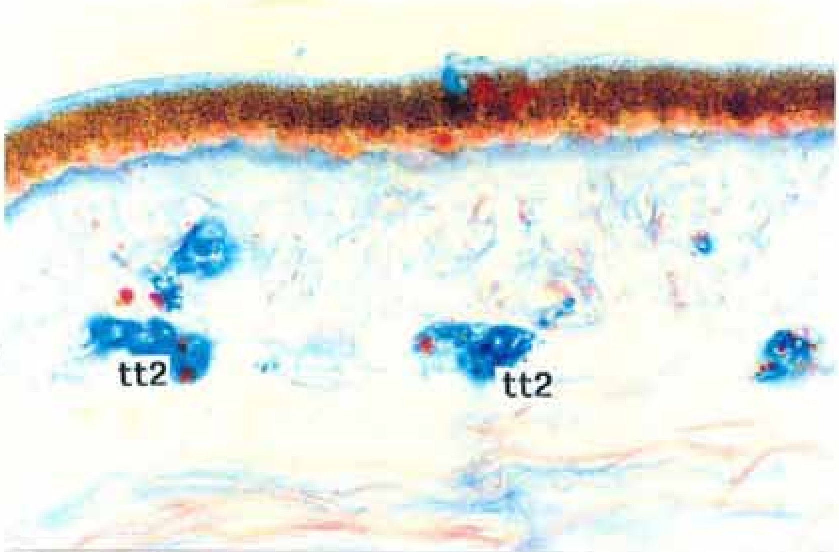
a. L.M. x 770 A.B./M.S.B. Proximal LatF3.

b. L.M. x 770 A.B./M.S.B. Distal LatF3.

c. L.M. x 770 A.B./M.S.B. MedF3.

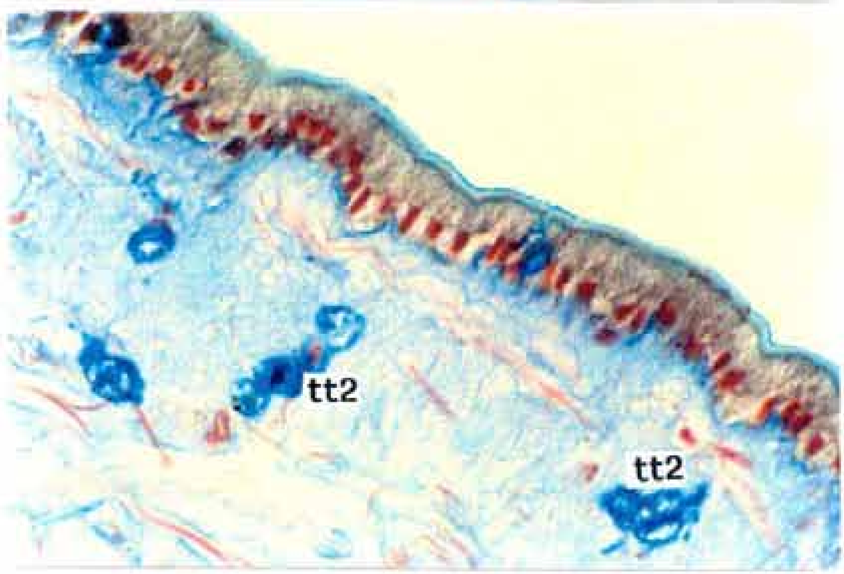
a

x770



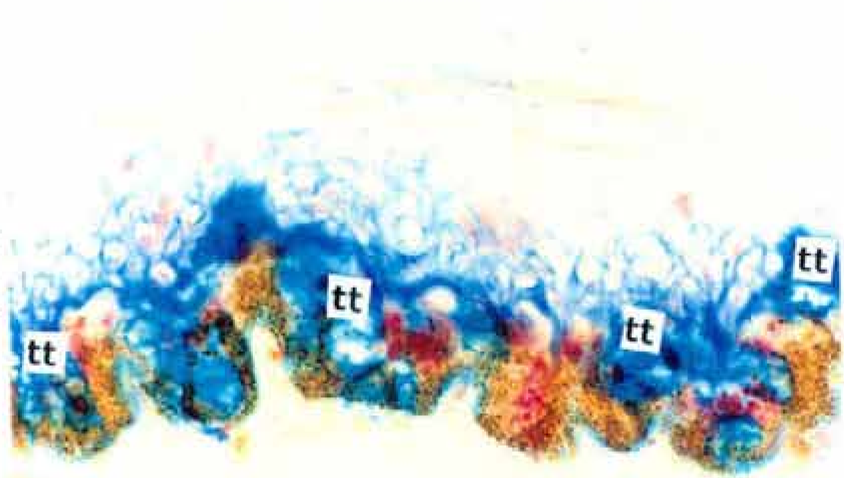
b

x770



c

x770



Figure

5.115

5.5.2.6.8.10. Ventral External Mantle and Mantle Margin.

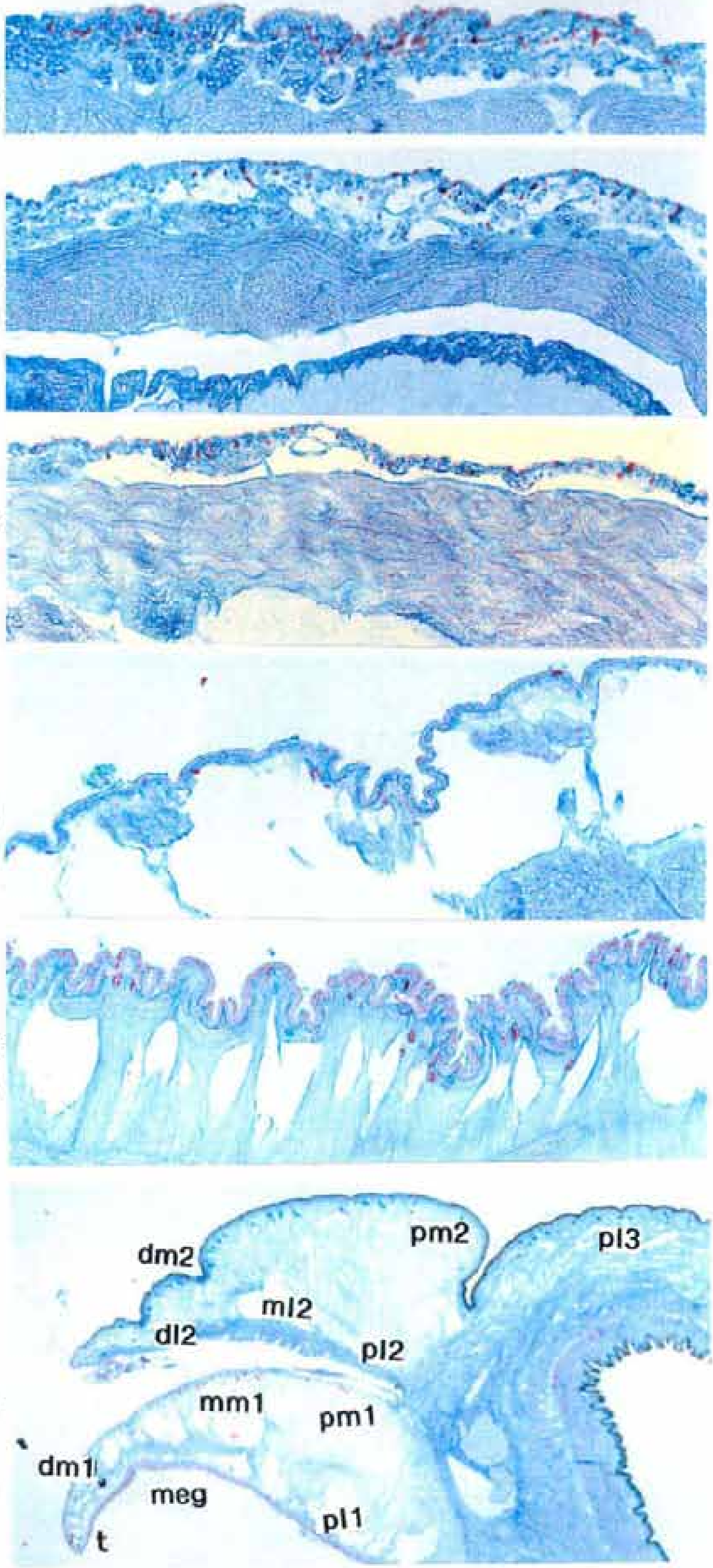
The Ventral Pallial Gland has a less dense population of either Trabecular Turquoise Glands or Granular Cytoplasm Secretory Glands than the Anterior Pallial Gland but is broader radially (Figure 5.116. a and b).

The Ventral Proximal Middle and Distal Pallial Regions are very similar to the equivalent Anterior Pallial Regions (Figure 5.116. c, d and e compared with Figure 5.102. b, c and d).

The histological divisions of the Anterior Mantle Margin (Figure 5.103.) are maintained in the Ventral Mantle Margin - again in Figure 5.116. pl1. meg and t indicate Proximal LatF1., Mantle Edge Gland and Terminal F1., dm1. mm1 and pm1 represent Distal MedF1., Middle MedF1. and Proximal MedF1. respectively; pl2. ml2. and dl2 indicate Proximal LatF2., Middle LatF2. and Distal LatF2. respectively and dm2 and pm2 indicate Distal MedF2. and Proximal MedF2. respectively. pl3 locates Proximal LatF3..

FIGURE 5.116. *Pinctada margaritifera* - Ventral Anterior Pallial Gland and Proximal Middle and Distal Ventral Pallial Regions and Ventral Mantle Margin.

a.	L.M. x 120	A.B./M.S.B.	Proximal Part of Ventral Pallial Gland.
b.	L.M. x 120	A.B./M.S.B.	Distal Part of Ventral Pallial Gland.
c.	L.M. x 120	A.B./M.S.B.	Ventral Proximal Pallial Region.
d.	L.M. x 120	A.B./M.S.B.	Ventral Middle Pallial Region.
e.	L.M. x 120	A.B./M.S.B.	Distal Middle Pallial Region.
f.	L.M. x 40	A.B./M.S.B.	Ventral Mantle Margin.



a x120

b x120

c x120

d x120

e x120

f x40

Figure 5.116

5.5.2.6.8.11. Ventral Distal Folded Region and Ventral Mantle Edge Gland and Terminal F1..

The Ventral Distal Folded Region has a lower columnar epithelium than the Anterior Distal Folded Region but a similarly dense population of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands (tt and g in Figure 5.117. a – c.f. Figure 5.106. a).

The Ventral Mantle Edge Gland (meg in Figure 5.117. b) is composed of very uniform elongate columnar epithelial cells with a surface secretion which stains turquoise with A.B./M.S.B. and is similar in all respects to the Anterior Mantle Edge Gland (Figure 5.107. b).

The terminal epithelium (t in Figure 5.117. c) is a low columnar epithelium with basal nuclei. In the terminal parenchyma of Ventral Fold F1. are Ovoid Blue Glands and Distal Diffuse Glands (ob and dd respectively in Figure 5.117 c) as in Anterior Terminal F1. (Figure 5.107. c and d).

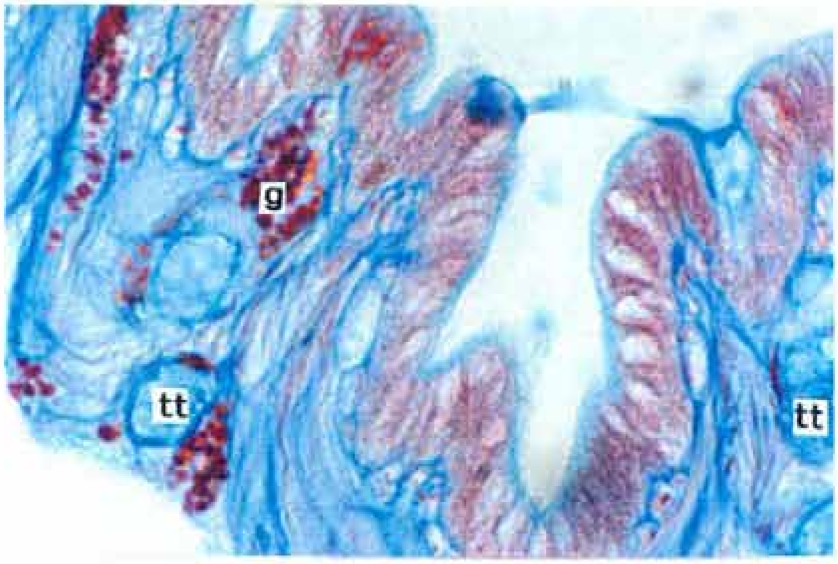
FIGURE 5.117. *Pinctada margaritifera* – Ventral Distal Folded Region, Mantle Edge Gland and Terminal F1..

a. L.M. x 1200 A.B./M.S.B. Distal Folded Region.

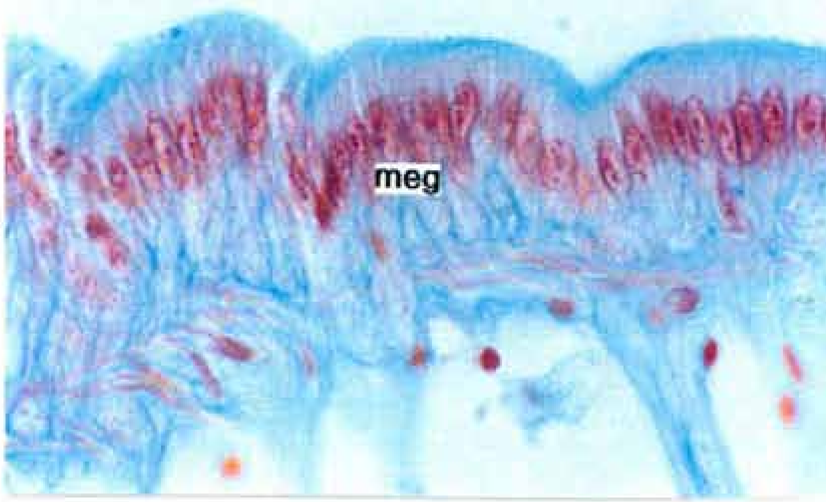
b. L.M. x 1200 A.B./M.S.B. Mantle Edge Gland.

c. L.M. x 1200 A.B./M.S.B. Terminal F1..

a
x1200



b
x1200



c
x770

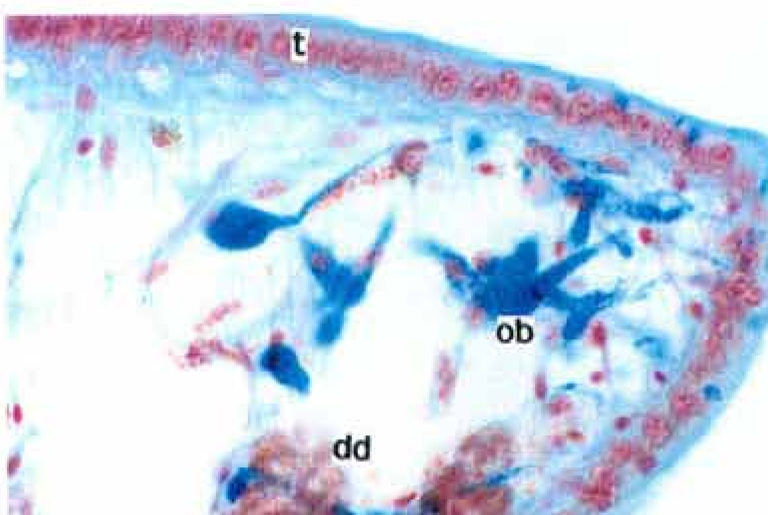


Figure
5.117

5.5.2.6.8.12. Ventral Medial and Apical F1..

Ventral Distal MedF1. has a low columnar surface epithelium with basal nuclei and Granular Cytoplasm Secretory Glands Ovoid Blue Glands (g and ob in Figure 5.118.) and Distal Diffuse Glands (dd in Figure 5.117). in the subepithelium.

The Ventral Middle MedF1. is again characterised by the distinctive subepithelial fibrous connective tissue unique to this location of the Mantle and Mantle Margin surface (c in Figure 5.118. b. compare with fc in Figure 5.108, b). The subepithelium of this Region hosts Trabecular Turquoise glands which are similar to those of the External Mantle and a variety of Granular Cytoplasm Secretory Glands (tt and g. g and g respectively in Figure 5.118. b).

The Ventral Proximal MedF1. has relatively few subepithelial Trabecular Turquoise or Granular Cytoplasm Secretory Glands (g in Figure 5.118. c.

Ventral Apical Groove F1F2. has an Omega Gland o in Figure 5.118. d) with Trabecular Turquoise Glands a common feature of the Unicellular Glands of the Omega Gland (tt in Figure 5.118. d).

The Dactylocytes (d in Figure 5.118. d). line the medial side of the Apical Channel from which issues the Pleated Secretion of Groove F1F2. (ps in Fiugre 5.118. d). The Black Granule Secretory Cells are distal to the Daclytocytes (bg in Figure 5.118. d).

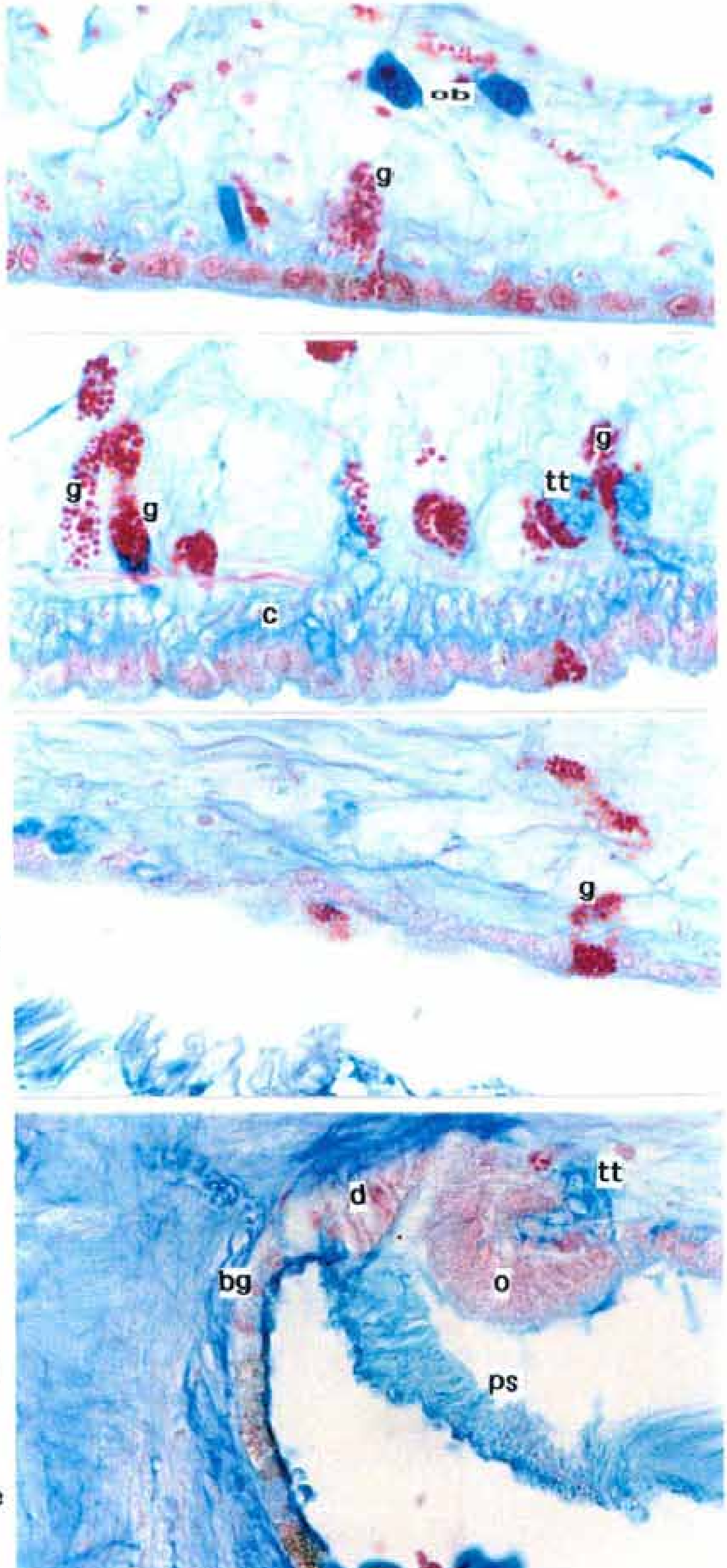
FIGURE 5.118. *Pinctada margaritifera* - Ventral MedF1. and Apical Groove F1F2..

a L.M. x 770 A.B./M.S.B. Ventral Distal MedF1..

b. L.M. x 770 A.B./M.S.B. Ventral Middle MedF1..

c. L.M. x 770 A.B./M.S.B. Ventral Proximal MedF1..

d. L.M. x 770 A.B./M.S.B. Ventral Apical Groove F1F2..



a
x770

b
x 770

c
x770

d
x770

Figure

5.118

5.5.2.6.8.13. Ventral Circumpallial Nerve and LatF2..

The Circumpallial Nerve, especially in the vicinity of the origins of the F1. and F2. rami, has numerous nuclei inside the nerve cord. Some of these are large pale nuclei each with a prominent nucleolus, in large pale staining cells with cytoplasmic extensions. That is, they are morphologically indistinct from neurons - n in Figure 5.119. a. Others are strong staining small nuclei in cells like the astrocytes of mammalian neuroglia (a in Figure 5.119. a). The Proximal LatF2. runs from the Black Granule Secretory Cells (bg in Figure 5.119. b) to the commencement of the ciliated epithelium of Middle LatF2. (c in Figure 5.119. d). The secretion of the subepithelial Granular Cytoplasm Secretory Glands (g in Figure 5.119. b) combine with the surface secretions of the epithelia (e in Figure 5.119. b and c) and the secretion of the Black Granule Secretory Glands (bg in Figure 5.119. b), to form the Vesiculate Secretion of Groove F1F2. (vs in Figure 5.119. c). This, together with the secretions of the F2F3. type Trabecular Turquoise Glands, Light Blue Glands and Granular Cytoplasm Secretory Glands of Middle and Distal LatF2. (tt2, lb and g respectively in Figure 5.119. d and e) produces what is thought to be part of the forming Organic Matrices of the Prismatic Layer (PL in Fiugre 5.119. e). The ciliated epithelium, (c in Figure 5.119. d) is that typical of Middle LatF2., as are the semicircular invaginations separated by short plateaux (Figure 5.119. d compare with Figure 5.114. a). The Ventral Distal LatF2. has a ciliated columnar surface epithelium (c in Figure 5.119. e) with rounded basal nuclei.

FIGURE 5.119. *Pinctada margaritifera* - Ventral Circum-pallial Nerve,LatF2..

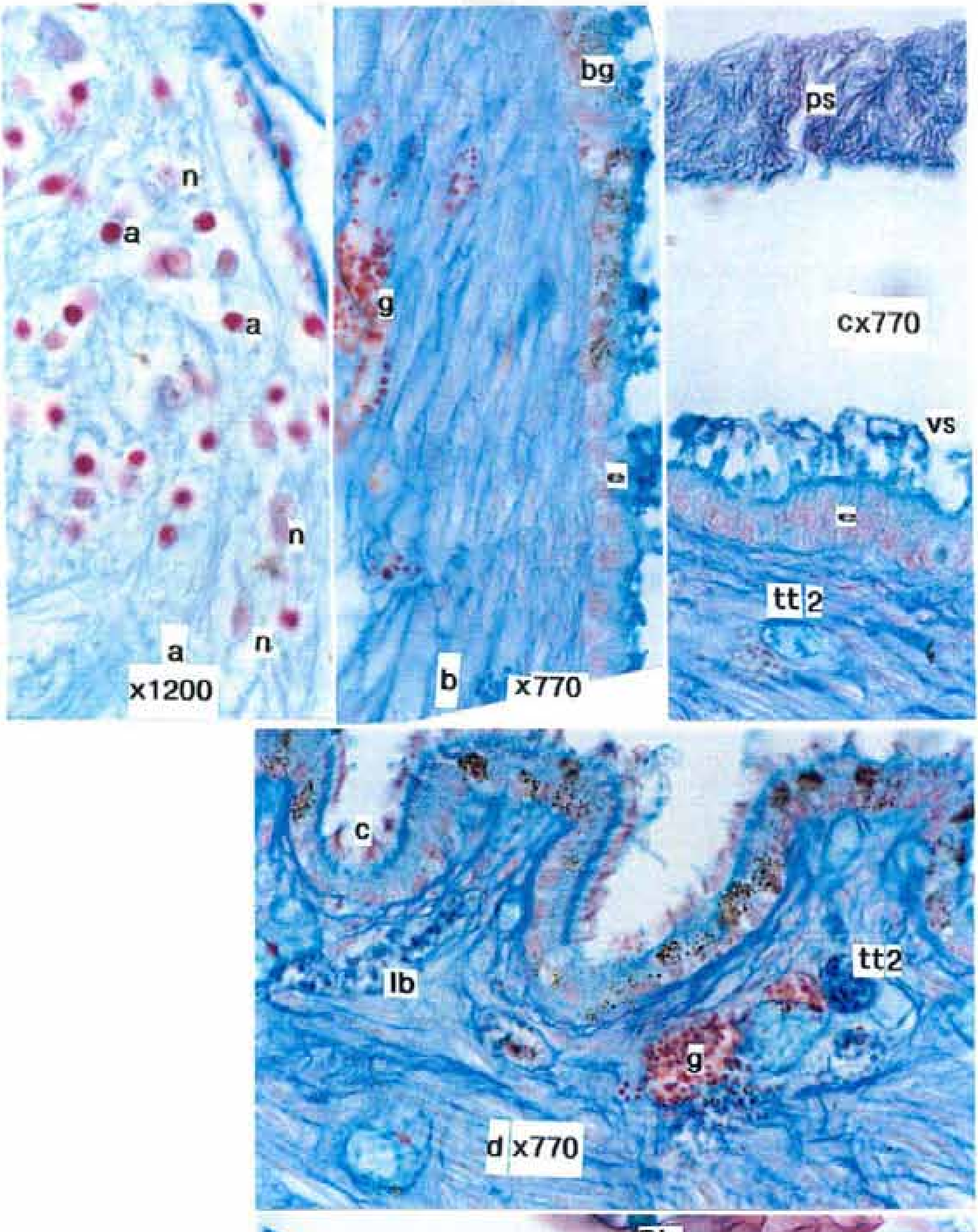
a. L.M. x 1200 A.B./M.S.B. Nuclei of cells in Circum-pallial Nerve.

b. L.M. x 770 A.B./M.S.B. Proximal LatF2.

c. L.M. x 770 A.B./M.S.B. Pleated Secretion of Groove F1F2. and Vesiculate Secretion of Groove F1F2. in Prox. Part of Groove.

d. L.M. x 770 Middle LatF2.

e. L.M. x 770 Distal LatF2.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g
d x770
PL
c
lb
tt2
e x770

Figure 5.119

5.5.2.6.8.14. Ventral MedF2. and LatF3..

Distal and Proximal MedF2. and Proximal and Distal LatF3. all have columnar surface epithelia with basal nuclei and pigmented apical cytoplasms (p in Figure 5.120. a, b c and d).

The pigment granules change from predominantly brown granules with A.B./M.S.B. in the Distal MedF2. epithelium to a mixture of amber and brown granules in Proximal MedF2.. almost exclusively amber granules in Proximal LatF3. and almost exclusely brown granules in Distal LatF3. The columnar epithelium is longer and the granules far denser in Proximal LatF3. than the other Rgions.

Each of these four Regions displays F2.-F3. type Trabecular Turquoise Glands (tt2 in Figure 5.120 a, b and c), and there are Granular Cytoplasm Secretory Glands in all four Regions (g in Figure 5.120.a, b, c and d), some of which, as those shown as g in Figure 5.120. a, are unique to that region.

FIGURE 5.120. *Pinctada margaritifera* -Ventral Distal and Proximal MedF2.. Proximal and Distal LatF3..

a. L.M. x 770 Distal MedF2..
b. L.M. x 770 Proximal MedF2..
c. L.M. x 770 Proximal LatF3..
d. L.M. x 770 Distal LatF3..

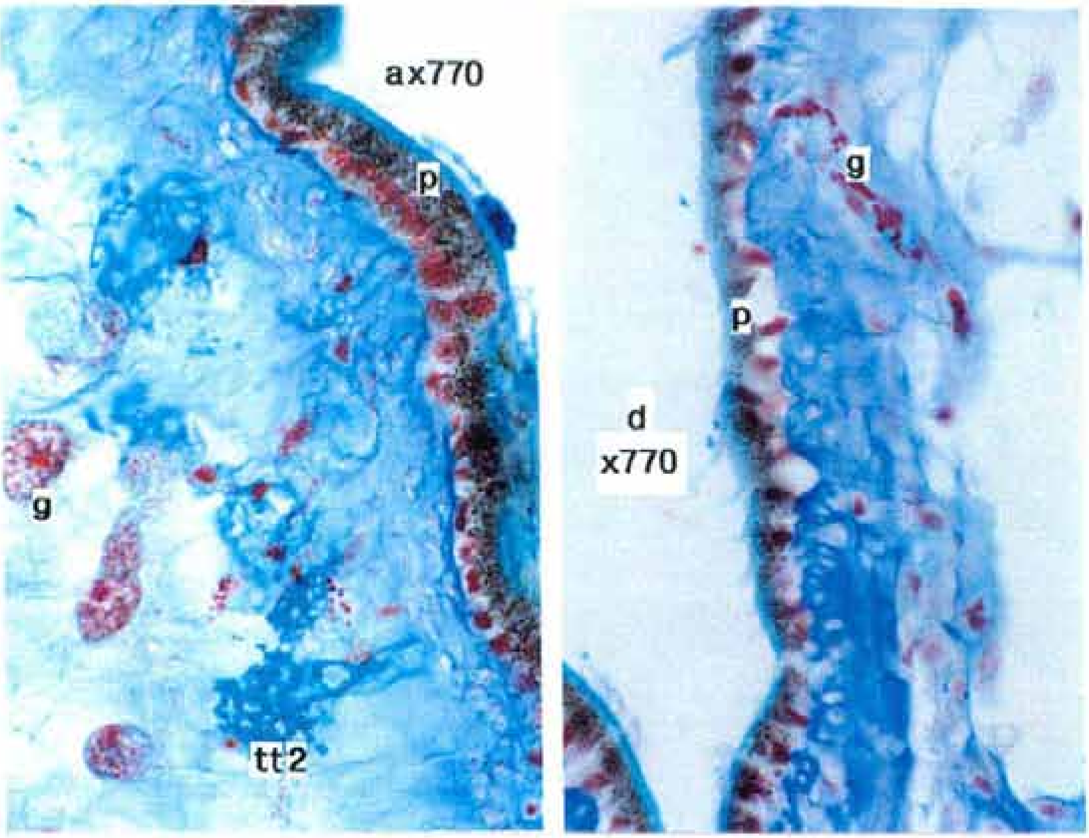
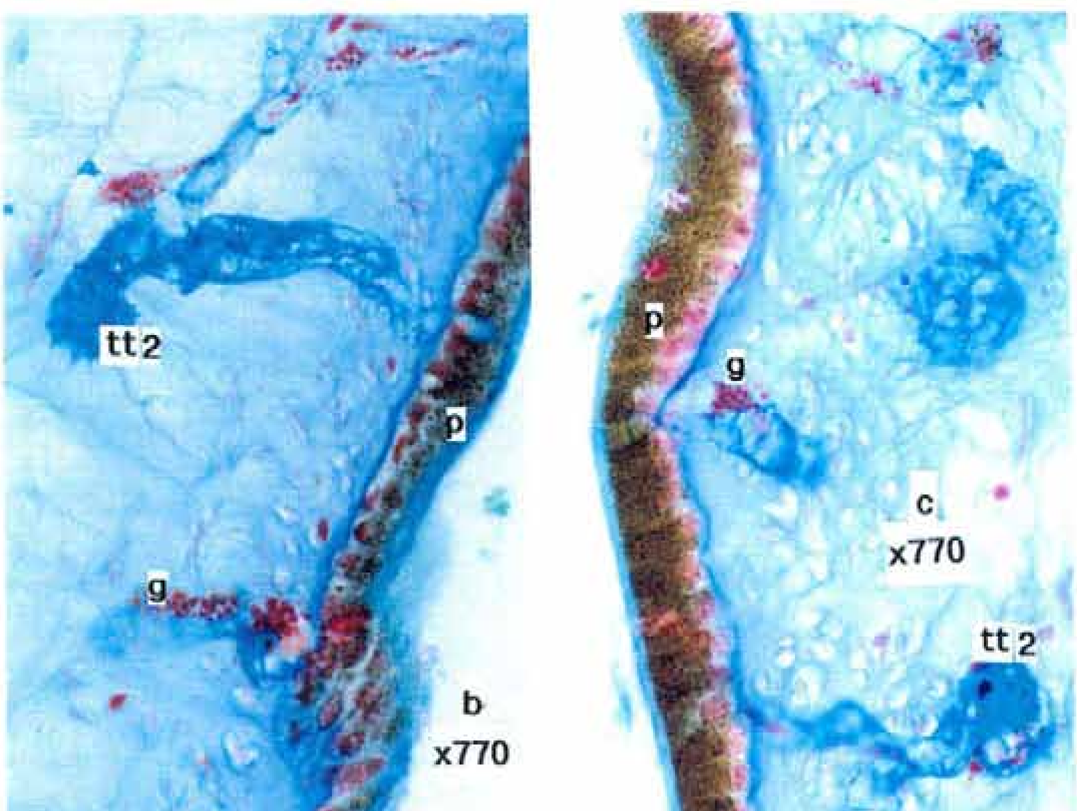


Figure 5.120



5.5.2.6.8.15. The Posterior Pallial Gland and Pallial Regions.

The Posterior Pallial Gland has a dense subepithelial fibrous connective tissue layer, but a greatly decreased glandular tissue of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands (tt and g respectively in Figure 5.121. a), compared with the Ventral and more especially the Anterior Pallial Glands (Figure 5.121. a, compared with Figures 5.116. a. and Figure 5.102. a).

The Trabecular Turquoise Glands of the Posterior Proximal, Middle and Distal Pallial Regions (Figure 5.121. b, c and d respectively) are largely intraepithelial or immediately subepithelial (tt in Figure 5.121. b, c and d).

The Granular Cytoplasm Secretory Glands of Proximal Middle and Distal Pallial Regions are, relatively, both small and sparse (g in Figures 5.121. b, c and d).

FIGURE 5.121. *Pinctada margaritifera* - Posterior Pallial Gland and Proximal, Middle and Distal Pallial Regions.

a. L.M. x 1200 A.B./M.S.B. Posterior Pallial Gland.

b. L.M. x 1200 A.B./M.S.B. Posterior Proximal Pallial Region.

c. L.M. x 1200 A.B./M.S.B. Posterior Middle Pallial Region.

d. L.M. x 1200 A.B./M.S.B. Posterior Distal Pallial Region.

a
x1200

b
x1200

c
x1200

d
x1200

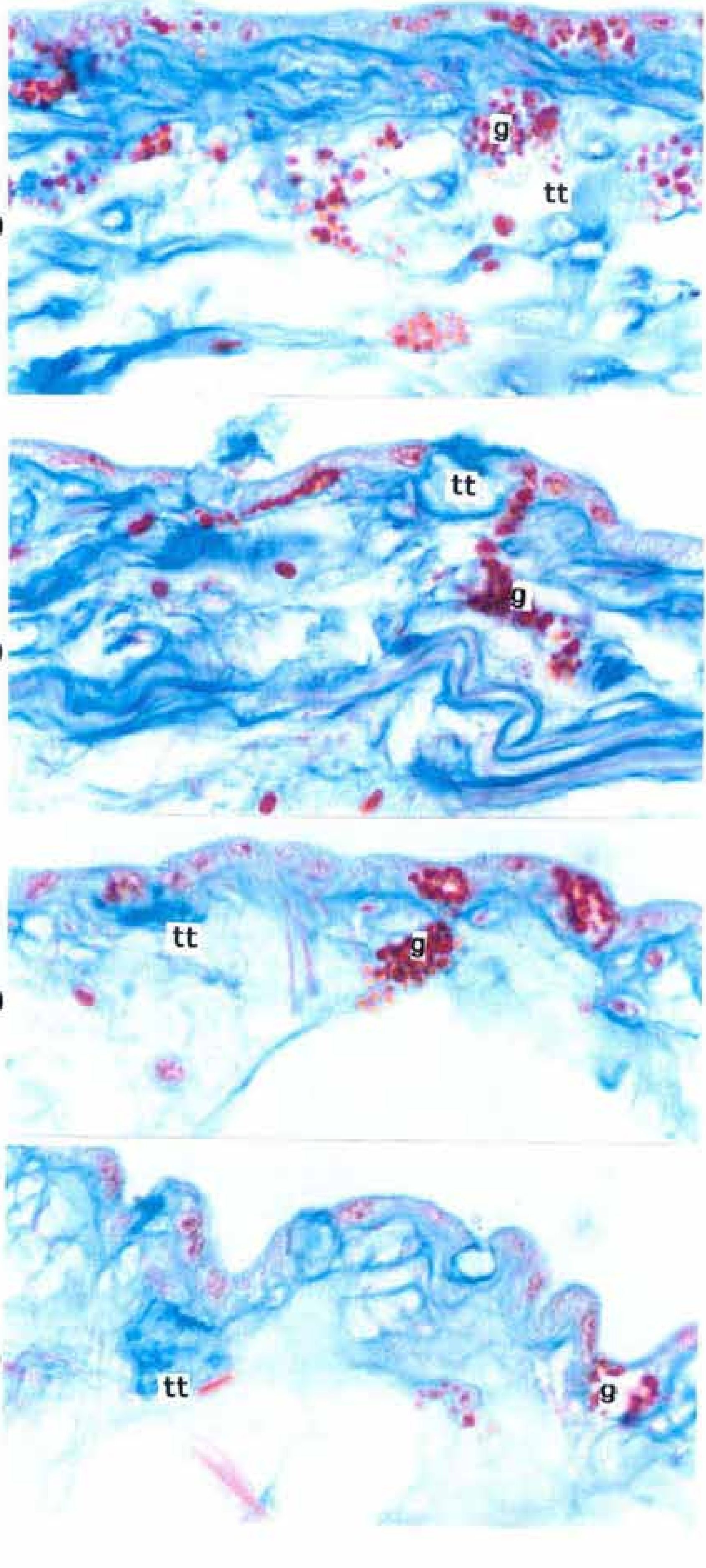


Figure
5.121

5.5.2.6.8.16. Posterior Distal Folded Region, Mantle Edge Gland and Terminal F1..

The Posterior Distal Folded Region has a relatively low columnar epithelium with relatively large apical nuclei, (n in Figure 5.122. a). There are here a heavy concentration of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands. (tt and g respectively in Figure 5.122. a).

The Mantle Edge Gland is a far lower columnar epithelium than that of the Ventral and Anterior Mantle Margins (Figure 5.122. b compared with Figures 5.107. b and 5.117. b).

The Terminal Epithelium of Posterior LatF1. (t in Figure 5.122. c), and the Terminal F1. subepithelial unicellular glands (consisting especially of Ovoid Blue Glands, and Distal Diffuse Glands (ob and dd respectively in Figure 5.122. c)), are very similar to those of the Ventral and Anterior Mantle Margins (Figure 5.122. c compared with Figures 5.107. c and d and 5.117. e).

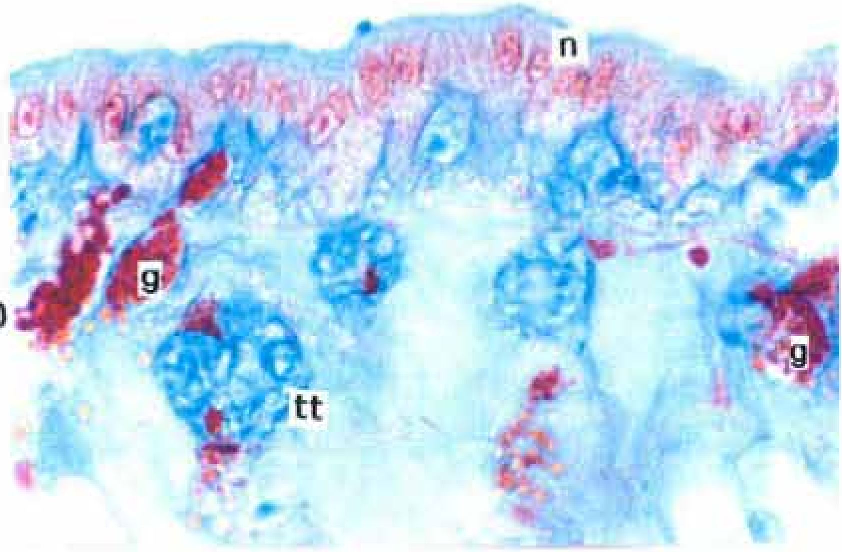
FIGURE 5.122. *Pinctada margaritifera* - Posterior Distal Folded Region Mantle Edge Gland and Terminal F1..

a. L.M. x 1200 A.B./M.S.B. Distal Folded Region.

b. L.M. x 1200 A.B./M.S.B. Posterior Mantle Edge Gland.

c. L.M. x 1200 Posterior Terminal F1..

a
x1200



b
x1200

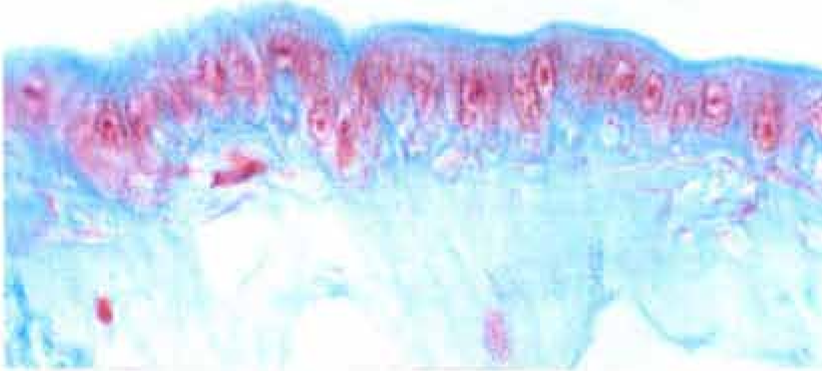
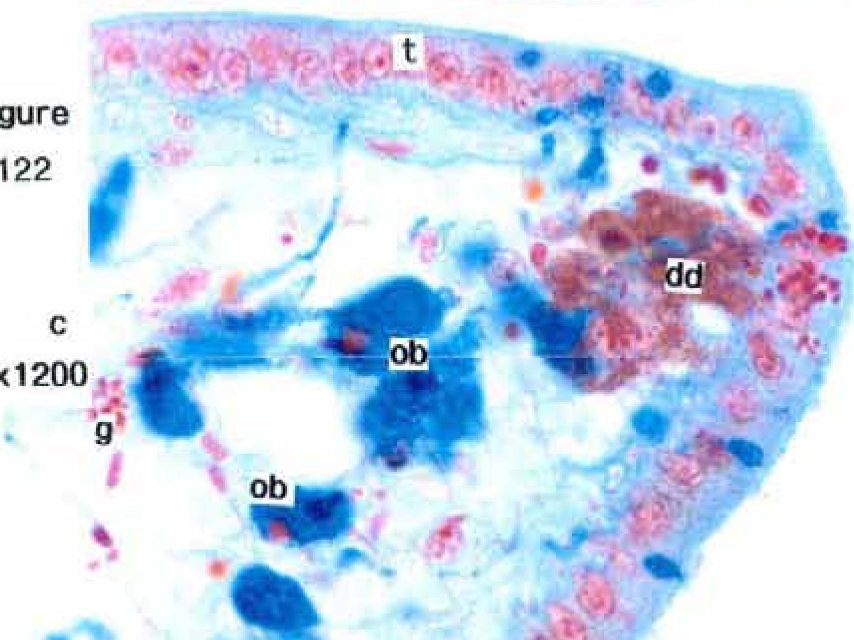


Figure
5.122

c
x1200



5.5.2.6.8.17. Posterior MedF1. and Apical Groove F1F2..

Posterior Middle MedF1. has the subepithelial fibrous connective tissue unique to this location for the entire periphery except for the byssal embayment (c in Figure 5.123. a, compared with Figures 5.108. b and 5.118. b). The subepithelium of the Middle and Proximal MedF1. bears Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands (tt and g in Figures 5.123. a and b).

The Distal Part of the Omega Gland is separated from the Proximal Part (do and po respectivel;y in Figure 5.123. b), by the Unicellular Glands of the Omega Gland (ug in Figure 5.123. b).

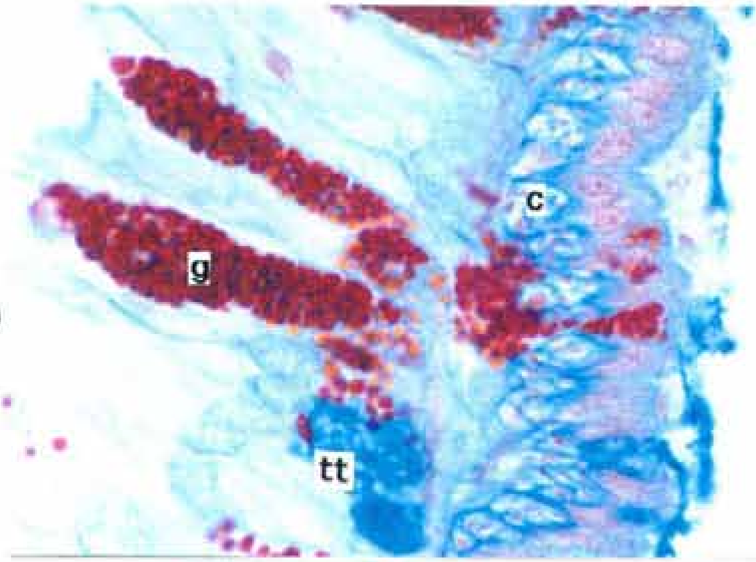
The Pleated Secretion of Groove F1F2., (ps in Figure 5.123. b) issues from the Posterior Apical Channel between the Proximal Part of the Omega Gland and the Dactylocytes (po and d respectively in Figure 5.123. b). The Black Granule Secretory Cells of the Posterior Apical Groove F1F2. are, as in the Ventral and Anterior Mantle Margins, located distal to the Dactylocytes on Proximal LatF2. (bg in Figure 5.123. b).

FIGURE 5.123. *Pinctada margaritifera* - Posterior MedF1. and Posterior Apical Groove F1F2..

a. L.M. x 1200 A.B./M.S.B. Middle MedF1..

b. L.M x 1200 A.B./M.S.B. Posterior Proximal MedF1. and Posterior Apical Groove F1F2..

a
x1200



b
x1200

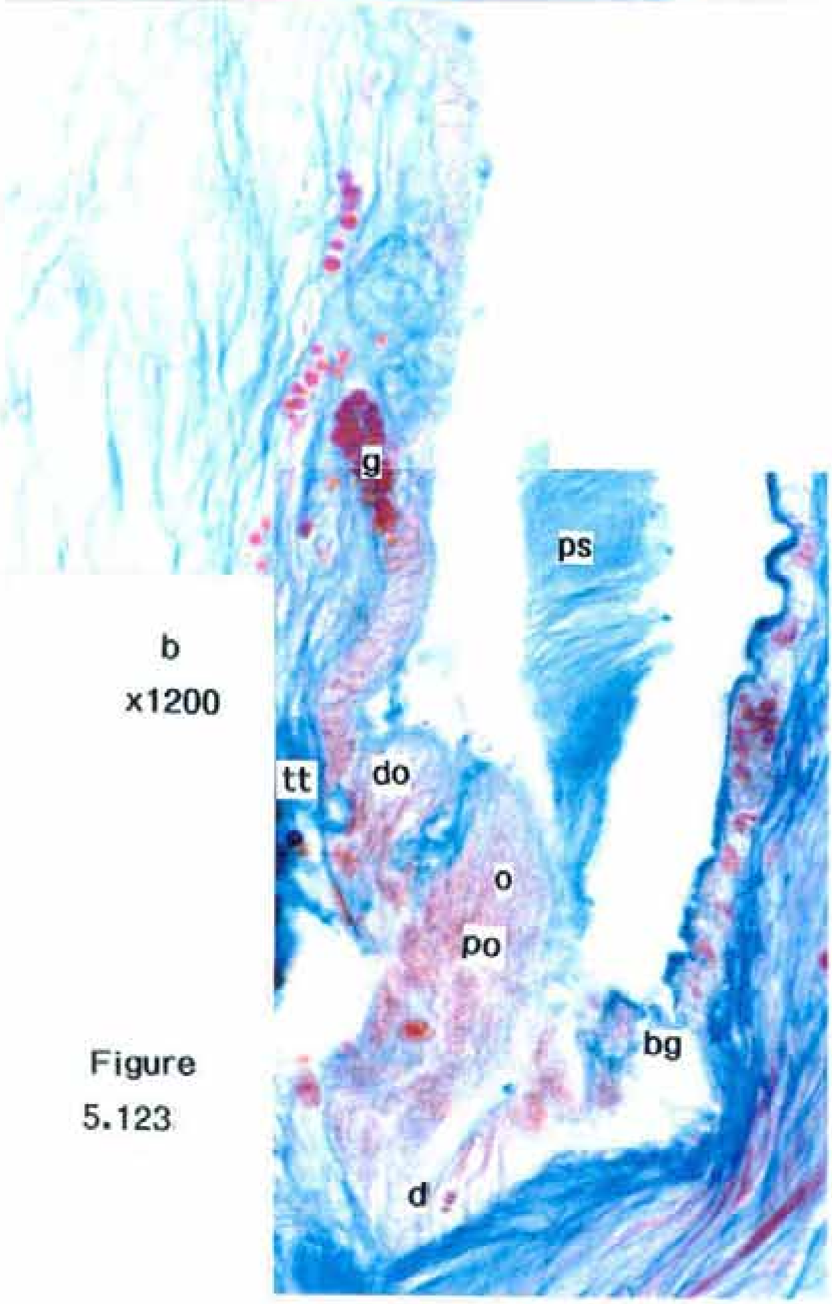


Figure
5.123

5.5.2.6.8.18. Posterior LatF2., MedF2. and LatF3..

There are a variety of species of Granular Cytoplasm Secretory Glands (g in Figure 5.124. a), in the subepithelium of Proximal LatF2. which secrete into Groove F1F2. via Receptacle Glands in the columnar surface epithelium (e in Figure 5.124. a).

The Middle LatF2. has a ciliated elongate columnar epithelium (c in Figure 5.124. b). The Vesiculate Secretion of Groove F1F2. is borne by the cilia, (vs in Figure 5.124. b). The Middle LatF2. epithelium has semicircular infoldings separated by short plateaux (Figures 5.124. b). There are numerous Type F2. - F3. Trabecular Turquoise Glands and Light Blue Glands in the subepithelium (tt2 and lb respectively in Figure 5.124. b).

The pigment granules of the surface epithelium apical cytoplasm of Regions Distal MedF2., Proximal MedF2., Proximal LatF3. and Distal LatF3. (p in Figure 5.124. c, d, e and f), are mostly grey-brown, a mixture of grey-brown and amber, predominantly amber and predominatly grey-brown respectively (Figure 5.124. c, d, e and f).

All four regions have an evenly spread population of Type F2. - F3. Trabecular Turquoise Glands in their subepithlium (tt2 in Figure 5.124. c, d, e and f) as well as Granular Cytoplasm Secretory Glands which are commonly located with the Trabecular Turquoise Glands (g in Figure 5.124. c, d and e).

FIGURE 5.124. *Pinctada margaritifera* - Posterior LatF2., MedF2., LatF3..

a. L.M. x 1200 A.B./M.S.B. Proximal LatF2..

b. L.M. x 1200 A.B./M.S.B. Middle LatF2..

c. L.M. x 1200 A.B./M.S.B. Distal MedF2..

d. L.M. x 1200 A.B./M.S.B. Proximal MedF2..

e. L.M. x 1200 A.B./M.S.B. Proximal LatF3..

f. L.M. x 1200 A.B./M.S.B. Distal LatF3..

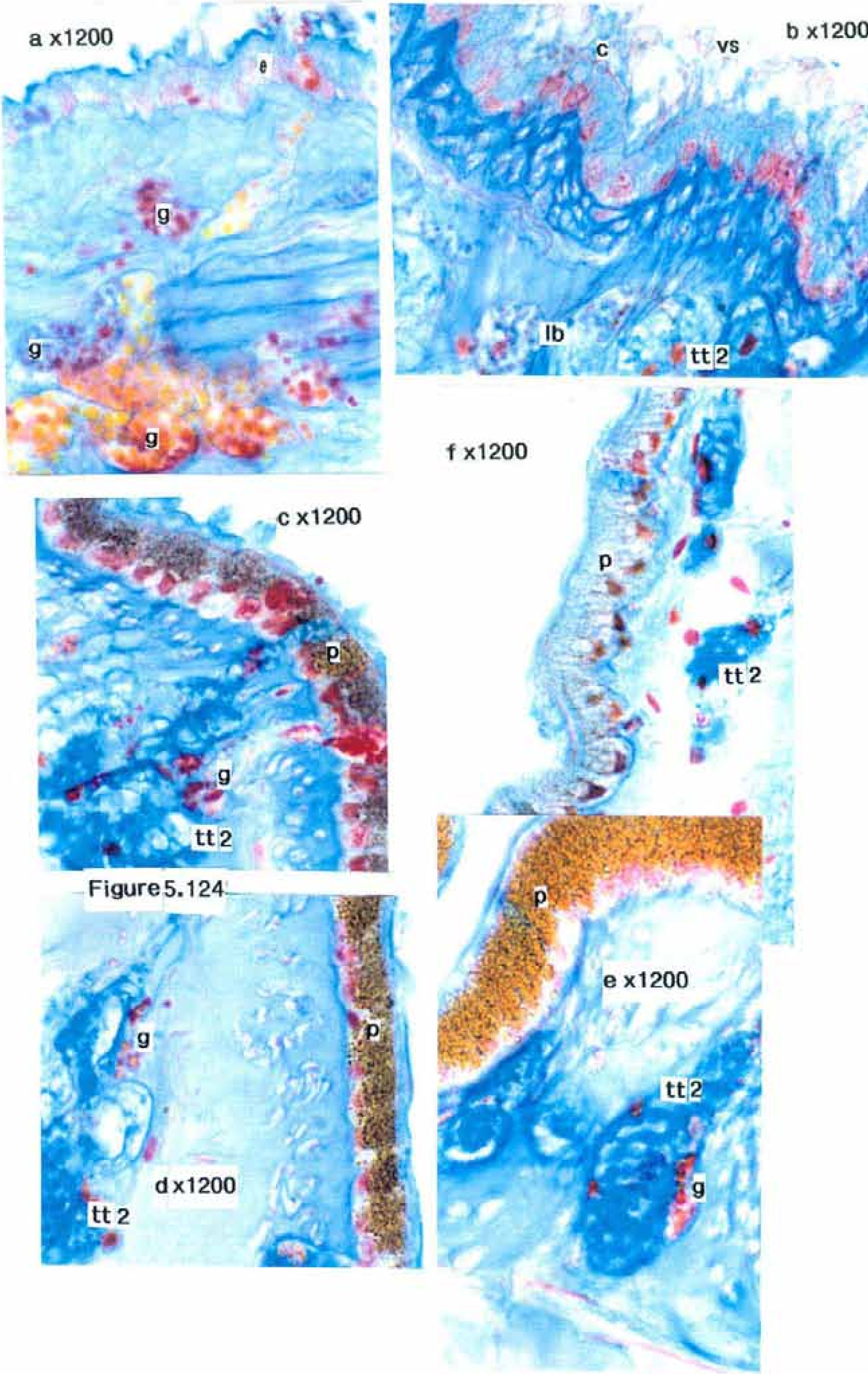

a x1200
e
g
g
g
b x1200
c
vs
lb
tt 2
f x1200
p
tt 2
c x1200
p
g
tt 2
d x1200
g
p
tt 2
e x1200
p
tt 2
g

Figure 5.124

5.5.2.6.8.19. Neural Supply to Pallial Gland.

The unicellular secretory glands of the Pallial Gland are directly supplied by branches of large nerves (b in Figure 5.125.) which run in the submucosal connective tissue (n in Figure 5.125.).

Within the structure of the Pallial Gland the large nerve branch shown appears to have two types of neurosecretory granules - larger ovoid granules (o in Figure 5.125.) which are concentrated in the vicinity of the Granular Cytoplasm Secretory Glands (g in Figuere 5.125.) and very small spherical granules (s in Figure 5.125.) concentrated in the vicinity of the Trabecular Turquoise Glands (tt in Figure 5.125.).

FIGURE 5.125. *Pinctada margaritifera* - Nerve Supply to Unicellular Glands of Pallial Gland.

T.E.M. x 3500.

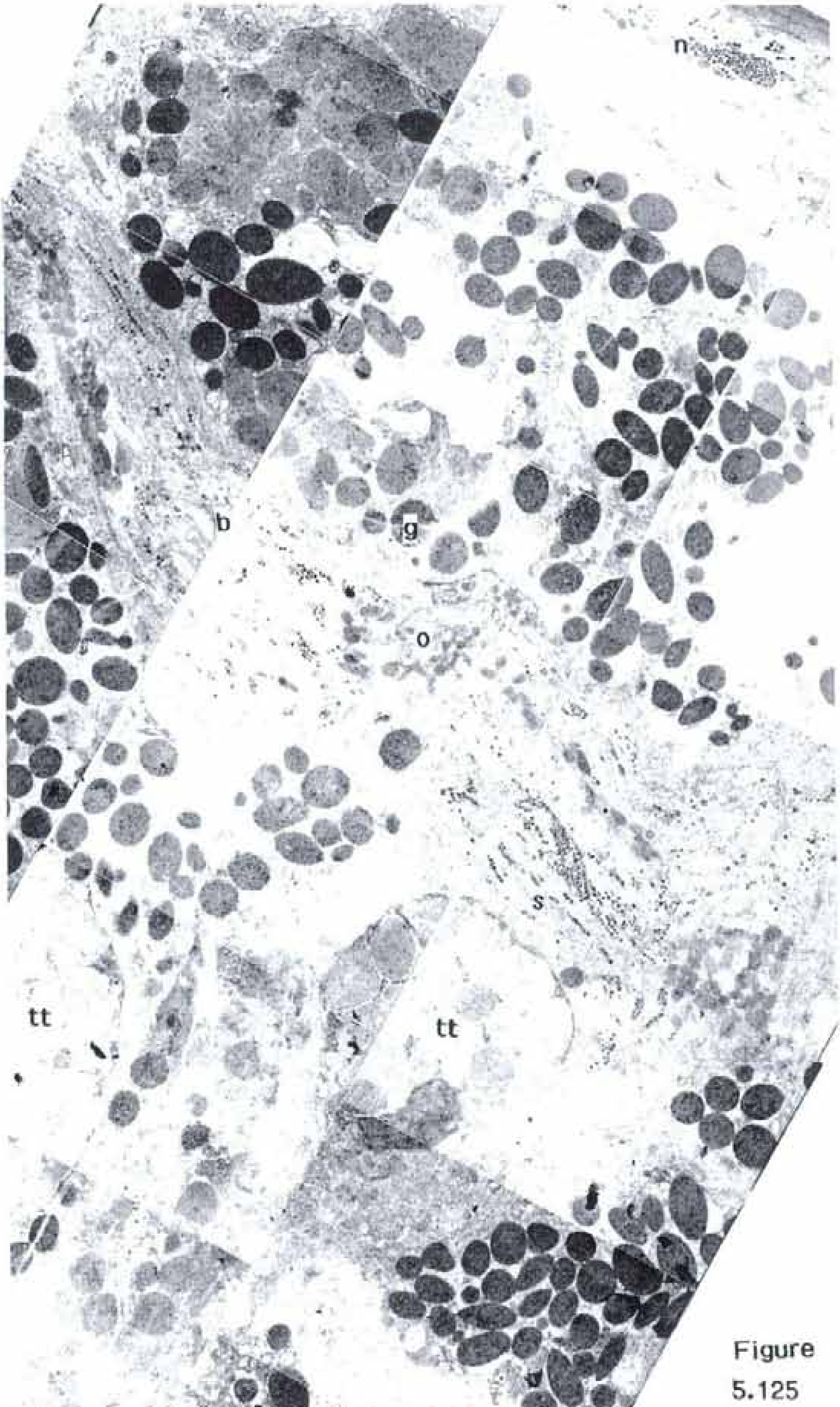


Figure
5.125
x3500

5.5. *PTERIACEA*. (Continued)

5.5.3. *Pinctada fucata*.

5.5.3.1. Medial View of Nacreous Layers and Nacreo-prismatic Junction.

The Patterns of edges of Nacre Sheets in the Shoulder Region and the Pallial Gland Region are commonly concentric (c in Figure 5.126. a and b) circular or spiral (s in Figure 5.126 b) joined by tightly curved patterns with numerous anastomoses. The partly-bound and free Nacre Tiles are elongate hexagons (pb and f respectively in Figure 5.126. c).

The medial surface of the Inner Nacreous Layer in the Ventral Pallial Region in general has patterns which are tightly curved with numerous anastomoses proximally and become progressively closer to parallel linear pattern going distally. Figure 5.126. d shows the transformation from curved patterns proximally (c on right) to parallel linear patterns distally (p on left).

The distance between edges of Nacre Sheets on the Inner Nacreous Layer decreases with decrese in curvature from about 45 µm apart in the concentric circular and spiral patterns of the Shoulder and Pallial Gland Regions (Figure 5.126. a and b) to about 35 µm apart in the strongly curved patterns of the Proximal Pallial Region (c in Figure 5.126. d, and e) to about 25 µm apart in the parallel linear patterns of the Distal Pallial Region (p in Figure 5.126. d and l and Figure 5.126. g and i). The distance bewtween the edges of Nacre Sheets in the parallel linear patterns of the Outer Nacreous Layer is about 12 µm (Figure 5.126. j).

In both the Inner Nacreous Layer and the Outer Nacreous layer the Partly-bound and Free Nacre Tiles (pb and f in Figure 5.126. c, e, f, g, i, j and l) tend towards a regular geometric shape. The bound Nacre Tiles are irregular polygons with bent side walls (Figure 5.126. k).

The partly-bound and free Nacre Tiles of both the Inner Nacreous Layer and the Outer Nacreous Layer decrease in size with distance from the bound edge of the Nacre Sheet to which they are related (Figure 5.126. e, f, g, i, j and l).

The Partly-bound and Free Nacre Tiles at the Nacreo-prismatic Junction lie in the grooves between the medial ends of the Prismatic Layer Prisms. (Figure 5.126. l).

Near the Nacreo-prismatic Junction the medial ends of the prisms are not covered by an Inner Fibrous Sheath. (Figure 5.126. l).

FIGURE 5.126. *Pinctada fucata* - Medial View, Nacreous Layers and Nacreo-prismatic Junction.

a. S.E.M. x 79 Inner Nacreous Layer - mid-Shoulder Region.

b. S.E.M. x 79 Inner Nacreous Layer - Pallial Gland Region.

c. S.E.M. x 6300 Inner Nacreous Layer - Pallial Gland Region.

d. S.E.M. x 79 Inner Nacreous Layer - Edges of Nacre Sheets Middle-Distal, Ventral Pallial Region.

e. S.E.M. x 1600 Inner Nacreous Layer - Bound, Partly-bound and Free Nacre Tiles.

f. S.E.M. x 3400 Inner Nacreous Layer - Middle Ventral Pallial Nacre.

g. S.E.M. x 1600 Inner Nacreous Layer - Distal Ventral Pallial Nacre.

h. S.E.M. x 160 Inner Nacreous Layer - Distal Ventral Pallial Nacre Parallel line patterns and embayment.

i S.E.M. x 1600 Inner Nacreous Layer Parallel Line Pattern at left of h. above.

j. S.E.M. x 1600 Outer Nacreous Layer, Parallel Line Pattern just proximal to Nacreo-prismatic Junction.

k. S.E.M. x 6300 Inner Nacreous Layer - broken edge of Nacre Sheet.

l. S.E.M. x 390 Partly-bound and Free Nacre Tiles at Nacreo-Prismatic Junction.

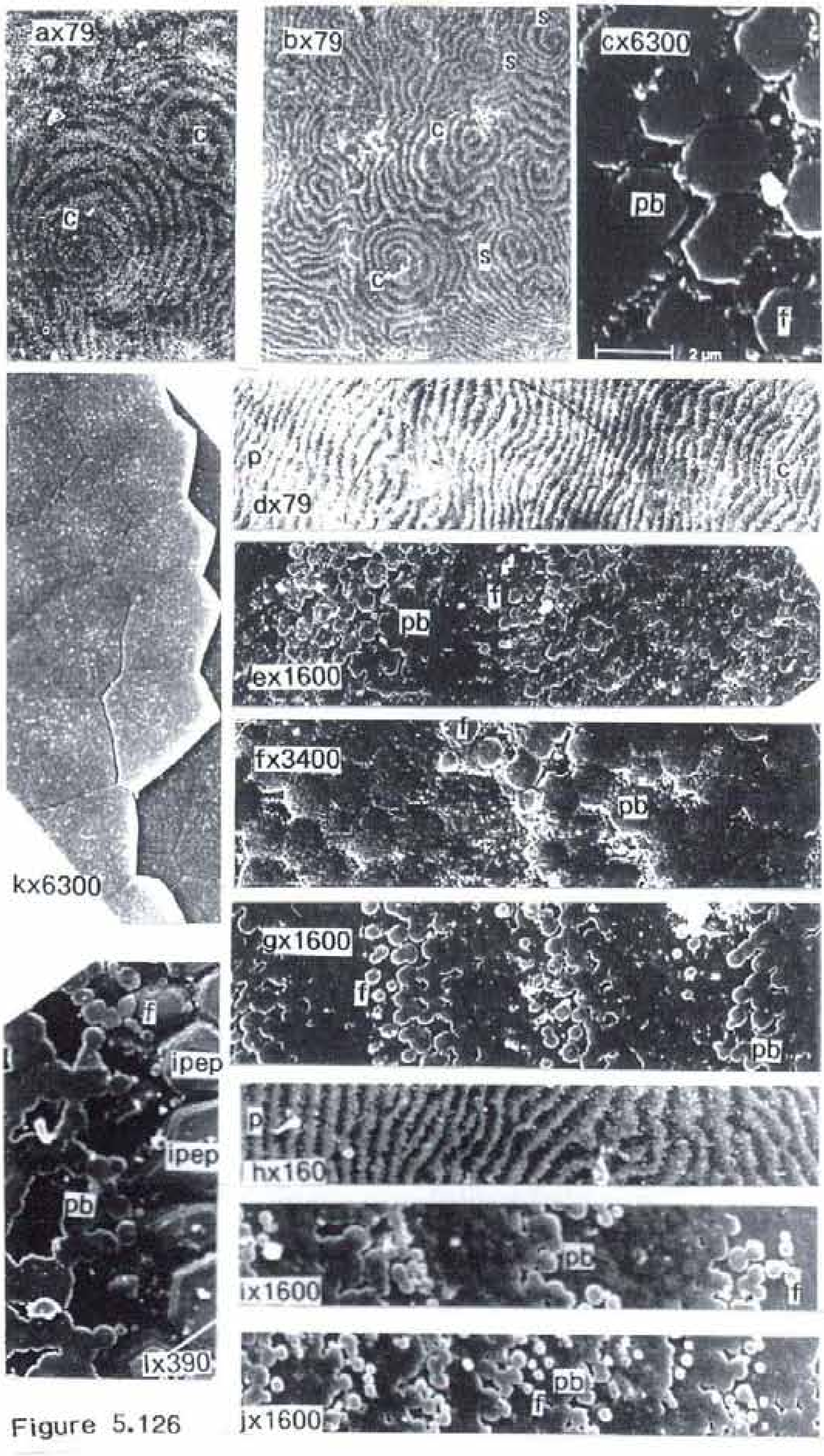

ax79
c
c
bx79
s
s
c
s
c
cx6300
pb
f
2 µm
p
c
dx79
f
pb
ex1600
f
fx3400
pb
kx6300
gx1600
f
pb
f
ipep
ipep
pb
lx390
p
hx160
pb
ix1600
f
pb
f
jx1600

Figure 5.126

5.5.3.2. Prismatic Layer - L.M. of Radial Section and S.E.M. of Medial and Lateral Surfaces.

The decalcified Prismatic Layer of *P. fucata* in radial section sometimes appears as if there is no physical limit between the prisms of successive Growth Scales (as at j in Figure 5.127. a) and at other places the successive Growth Scales are delimited by Inner Prismatic End Plates and/or an Inner Fibrous Sheath on the medial surface of the more lateral Growth Scale, and on an Outer Fibrous Sheath and Outer Prismatic End Plates on the lateral surface of the more medial Growth Scale (s in Figure 5.127. a). In other places the line of demarcation between the Growth Scales is sometimes marked by the presence in the lateral end of the Major Prisms of the more medial Growth Scale of Inner lateral Structure. In Figure 5.127. a, parts of two successive Growth Scales are shown. The earlier (more lateral) is marked 1 just proximal to where the Growth Process has been broken off, and the latter is marked 2 towards its proximal end. In Figure 5.127. a, P is proximal, D distal and L lateral.

The medial surface of both the distal Growth Scale (Figure 5.127. b) and Growth Processes (t Figure 5.127. c) are covered by Inner Fibrous Sheath (ifs in Figure 5.127. b and c) which is lacking just distal to the Nacreo-prismatic Junction (Figure 5.126. l).

The fibres of the Outer Fibrous Sheath (ofs in Figure 5.127. d and e) which covers the lateral surface of both the exposed Prismatic Layer and Growth Processes are far coarser than those of the Inner Fibrous Sheath and are directed around the surface directly over the centre of the underlying Outer Prismatic End Plates (Figure 5.127. d) and over the centre of these the fibres run transversely (t in Figure 5.127. e).

FIGURE 5.127. *Pinctada fucata* – L.M. Radial Section of Prismatic Layer and S.E.M. of Medial and Lateral Surfaces of Prismatic Layer.

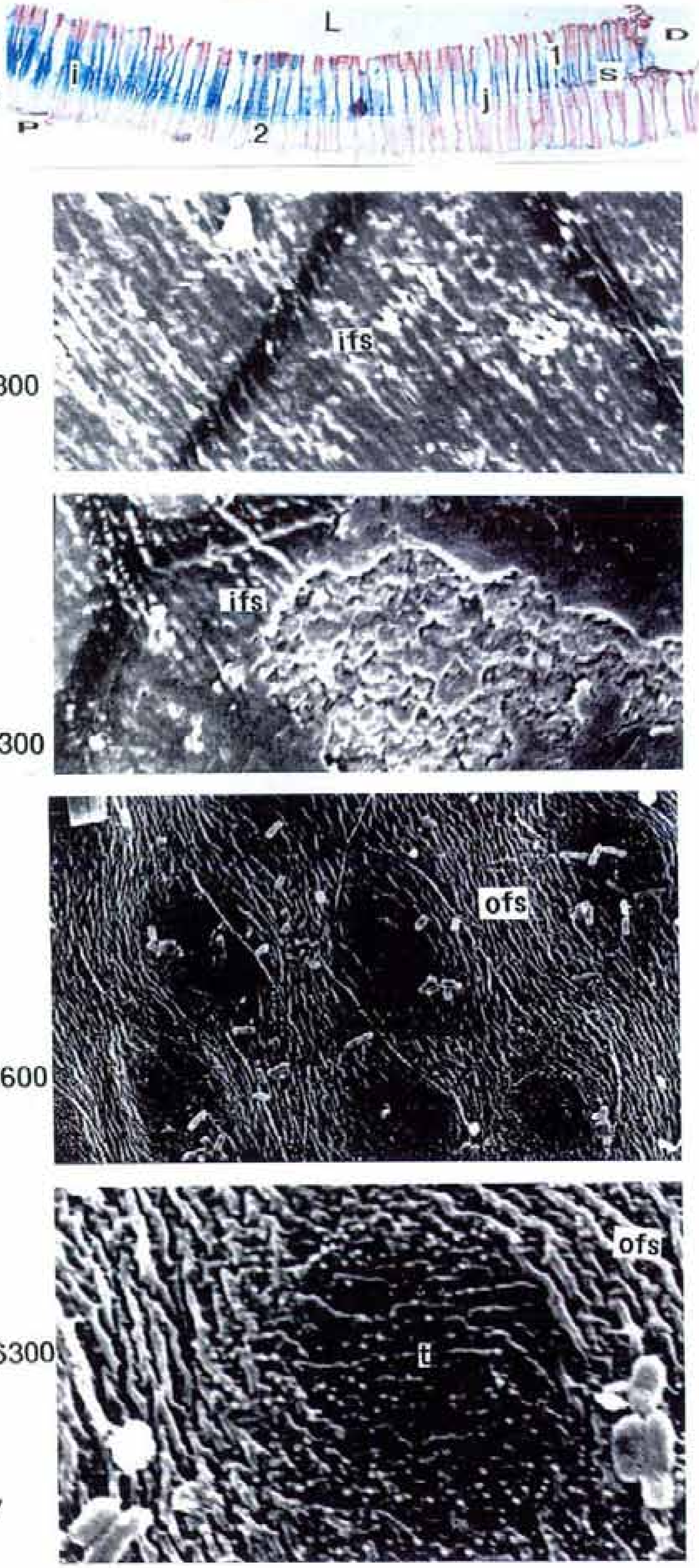
a. L.M. x 120 Azan Radial Section of Prismatic Layer.

b. S.E.M. x 6300 Inner Fibrous Sheath covering Distal Medial Surface of Prismatic Layer Growth Scale.

c. S.E.M. x 6300 Eroded Inner Fibrous Sheath, Medial Surface of Growth Process.

d. S.E.M. x 1600 Outer Fibrous Sheath, Lateral Surface of Growth Scale.

e. S.E.M. x 6300 Transverse Fibres over central part of Outer Prismatic End Plate – Lateral Surface of Prismatic Layer.



ax120

bx6300

cx6300

dx1600

ex6300

Figure 5.127

5.5.3.3. Radial Broken Surface of Prismatic Layer.

The Interprismatic Organic Matrices separate the Major Prisms of the Prismatic Layer (ip and mp respectively in Figure 5.128. a and b). The long axes of the Major Prisms are normal to the plane of junction with the Outer Nacreous Layer (on in Figure 5.128. a). The Interprismatic Organic Matrix consists of sheets of perforated material which lie in the Major Prisms in parallel planes normal to the long axis of each Major Prisms (in. in Figure 5.128. a and b).

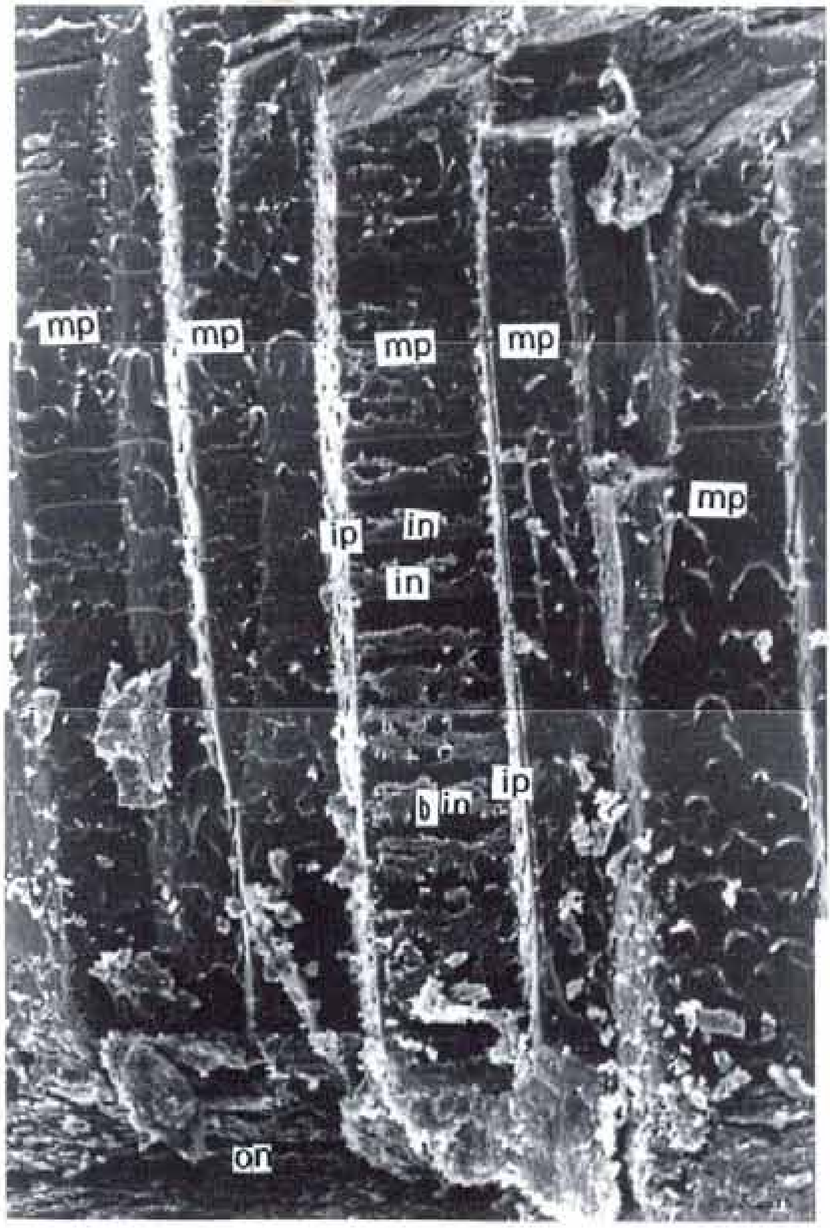
The Intraprismatic Organic Matrices are joined to the Reteform Interprismatic Organic Matrices, thus dividing the Major Prisms into compartments (c in Figure 5.128. a and b). The calcite tablets lie in these compartments.

FIGURE 5.128. *Pinctada fucata* – S.E.M. of Radial Broken Surface of Prismatic Layer.

a. S.E.M. x 680 Major Prisms of Prismatic Layer and Outer Nacreous Layer.

b. S.E.M. x 3400 Higher magnification near b in Figure 5.128. a.

a x680



b x3400

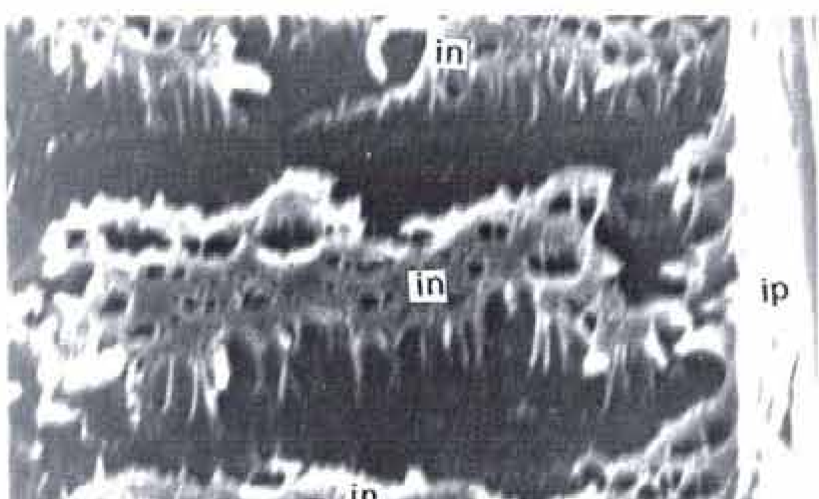


Figure 5.128

5.5.3.4. Pallial Gland, Pallial Region, Inner Nacreous Organic Matrix, Mantle

Margin, Fold F1. and Outer Nacreous Organic Matrix.

The Pallial Gland has Trabecular Turquoise Glands which are superficial to a dense mass of Granular Cytoplasm Secretory Glands (tt and g respectively in Figure 5.129. a).

These two kinds of glands are also found in the Pallial Region where there are relatively fewer Granular Cytoplasm Secretory Glands (tt and g in Figure 5.129. b).

On the surface of the Pallial Epithelium and in the adjacent Pallial Space (pl and ps respectively in Figure 5.129. a, b, c, d and e) the secreted granules from the Granular Cytoplasm Glands appear to at first coalesce and lose their identity in a secretory ball (s in Figure 5.129. c and d). These then appear to disintegrate laterally (d in Figure 5.129. c and d) and join with the secretions of the Trabecular Turquoise Glands to form Inner Nacreous Layer Organic Matrix (in. in Figure 5.129. c and d).

The Mantle Margin has Folds F1., F2. and F3., (f1, f2, and f3 in Figure 5.129. e and f).

LatF1. has a Mantle Edge Gland (meg in Figure 5.129. f). Apical Groove F1F2. has an Omega Gland on the lateral side and, just proximally the Circum-pallial Nerve (o and n respectively in Figure 5.129. e). There is a mass of glandular cells subepithelial to MedF1. (g mfi in Figure 5.129. e) which produce a secretion which stains turquoise with A.B./M.S.B. and may be the organic matrix of the Outer Nacreous Layer (onl in Figure 5.129. f). The Pleated Secretion of Groove F1F2. (ps in Figure 5.129. e) seperates MedF1. and its secretions from those of the subepithelial Glands of LatF2. (g in Figure 5.129. e).

FIGURE 5.129. *Pinctada fucata* - External Mantle, Mantle Margin and their Secretions.

a. L.M. x 770 Pallial Gland

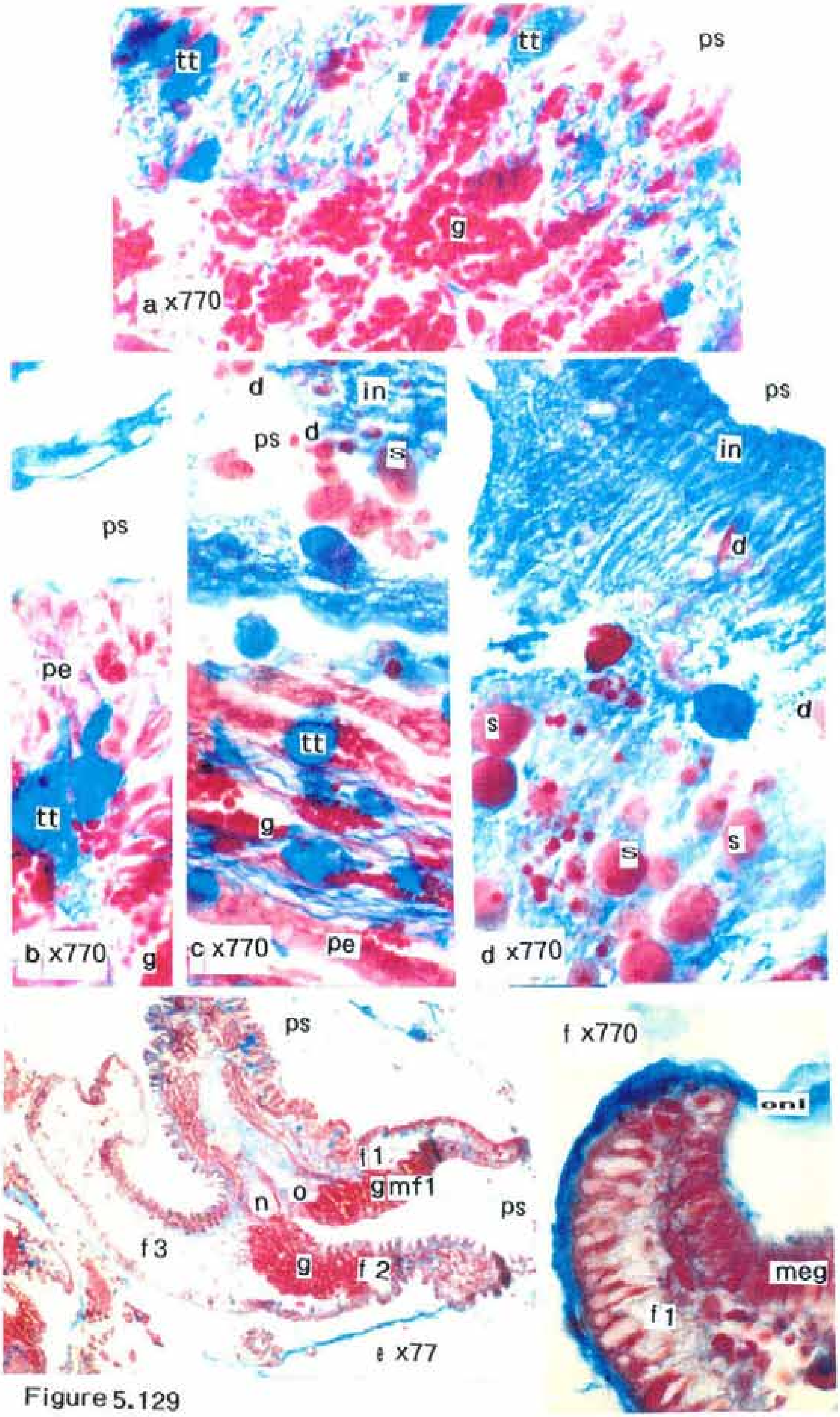
b. L.M. x 770 Pallial Region secretory structures.

c. L.M. x 770 Pallial Region surface and secretions.

d. L.M. x 770 Secreted material in Pallial Space.

e. L.M. x 77 Mantle Margin material secreted by MedF1.

f. L.M. x 770 Terminal F1.


tt
tt
ps
g
a x770
d
in
ps
d
s
ps
in
d
pe
tt
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s
tt
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b x770
g
c x770
pe
d x770
ps
f x770
onl
f1
n
o
g
mf1
ps
f3
g
f2
meg
f1
e x77

Figure 5.129

5.5.3.5. Apical Groove F1F2..

A tangential section of the apex of Groove F1F2. shows that in this plane the Apical Channel is folded, (ac in Figure 5.130.).

The Celtic Scroll Cells line the lateral side of the Apical Channel and the Dactylocytes the medial side, (cs and d respectively in Figure 5.130.).

The Unicellular Glands of the Omega Gland lie immediately lateral to the Omega Gland epithelial cells (u in Figure 5.130.). Granular Cytoplasm Secretory Glands occur amongst the muscles and fibrous connective tissue between the Dactylocytes and the Circum-pallial Nerve.

Apart from the nuclei of Sheath Cells (s in Figure 5.129.) at the edge of the Circum-pallial Nerve, (cn in Figure 5.130.), two different types of nuclei occur within the body of the nerve. One is a small dense nucleus (a in Figure 5.129.). The other type are large pale nuclei with prominent nucleoli in large pale cells (n in Figure 5.130.). The latter are thought to be those of peripheral neurones - large pale staining nuclei with prominent nucleoli, (n in Figure 5.130).

FIGURE 5.130. *Pinctada fucata* Apical Groove F1F2.
L.M. x 770 Azan.

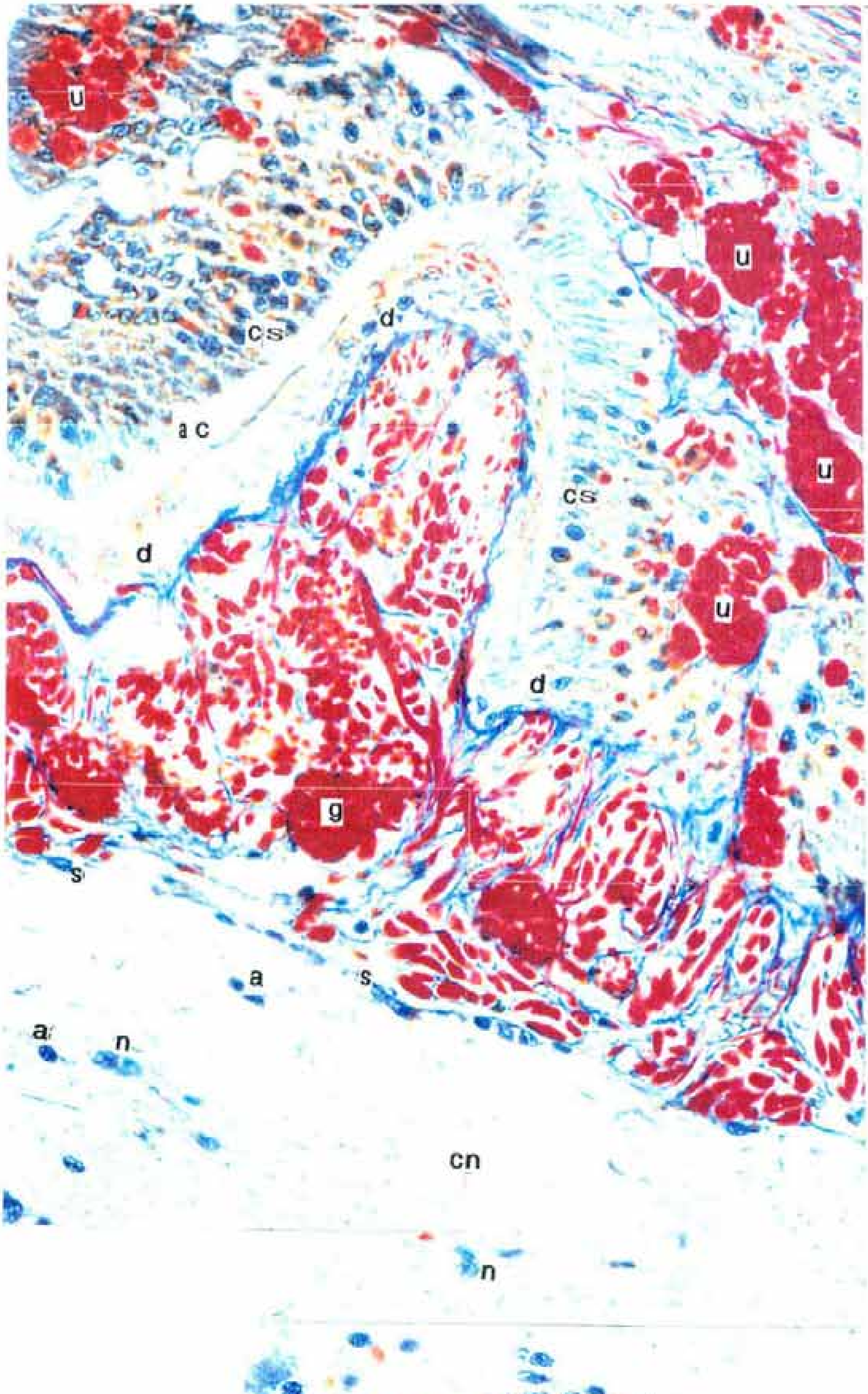


Figure 5.130

5.5.4. *PINCTADA SP. 1.*

5.5.4.1. Nacreous Layers and Prismatic Layer of Valve.

The medial surface of the Inner Nacreous Layer displays concentric circular patterns of edges of Nacre Sheets in the Shoulder Region (c in Figure 5.131. a), and concentric ovoid, (o in Figure 5.131. b), ones in the Pallial Gland Region. Going distally across the Pallial Region the patterns of edges of Nacre Sheets progressively approach parallel linear patterns (Figure 5.131. c). Throughout the Inner Nacreous Layer medial surface the distance between edges of Nacre Sheets is greater where they are more strongly curved and becomes smaller as the patterns approach parallel linearity. The parallel linear edges of Nacre Sheets of the Outer Nacreous Layer (Figure 5.131. d) are about half the distance apart of the most narrowly spaced patterns in the Inner Nacreous Layers (cf Figure 5.131 c).

Throughout the medial surface of the Nacreous Layers the Partly-bound and Free Nacre Tiles lie on about half the distance between the bound edges of Nacre Sheets and become progressively smaller with distance from the bound edge of the Nacre Sheet.

At the Nacreo-prismatic Junction the Partly-bound and Free Nacre Tiles lie in the Grooves surrounding the Inner Prismatic End Plates (ipep in Figure 5.131. e) which are not covered with an Inner Fibrous Sheath adjacent to the Nacreo-prismatic Junction.

More distally the Inner Prismatic End Plates are covered with an Inner Fibrous Sheath (ipep and ifs respectively in Figure 5.131. f). The periodicity of the surface fibres of this is less than half that of those of the Outer Fibrous Sheath (ofs in Figure 5.131. g) which invests the outer surface of the Prismatic Layer.

Near the edge of the Growth Processes the Inner Prismatic End Plates of the Growth Process Prisms are constrained into parallelograms instead of the usual irregular polygons (p in Figure 5.131. h and i).

FIGURE 5.131. *Pinctada sp. 1.* Nacreous Layers and Prismatic Layer of Valve.

a. S.E.M. x 100 Inner Nacreous Layer. Shoulder Region Medial View.

b. S.E.M. x 100 Inner Nacreous Layer. Pallial Gland Region Medial View.

c. S.E.M. x 100 Inner Nacreous Layer. Pallial Region Medial View.

d. S.E.M. x 400 Outer Nacreous Layer just proximal to Nacreo-Prismatic Junction.

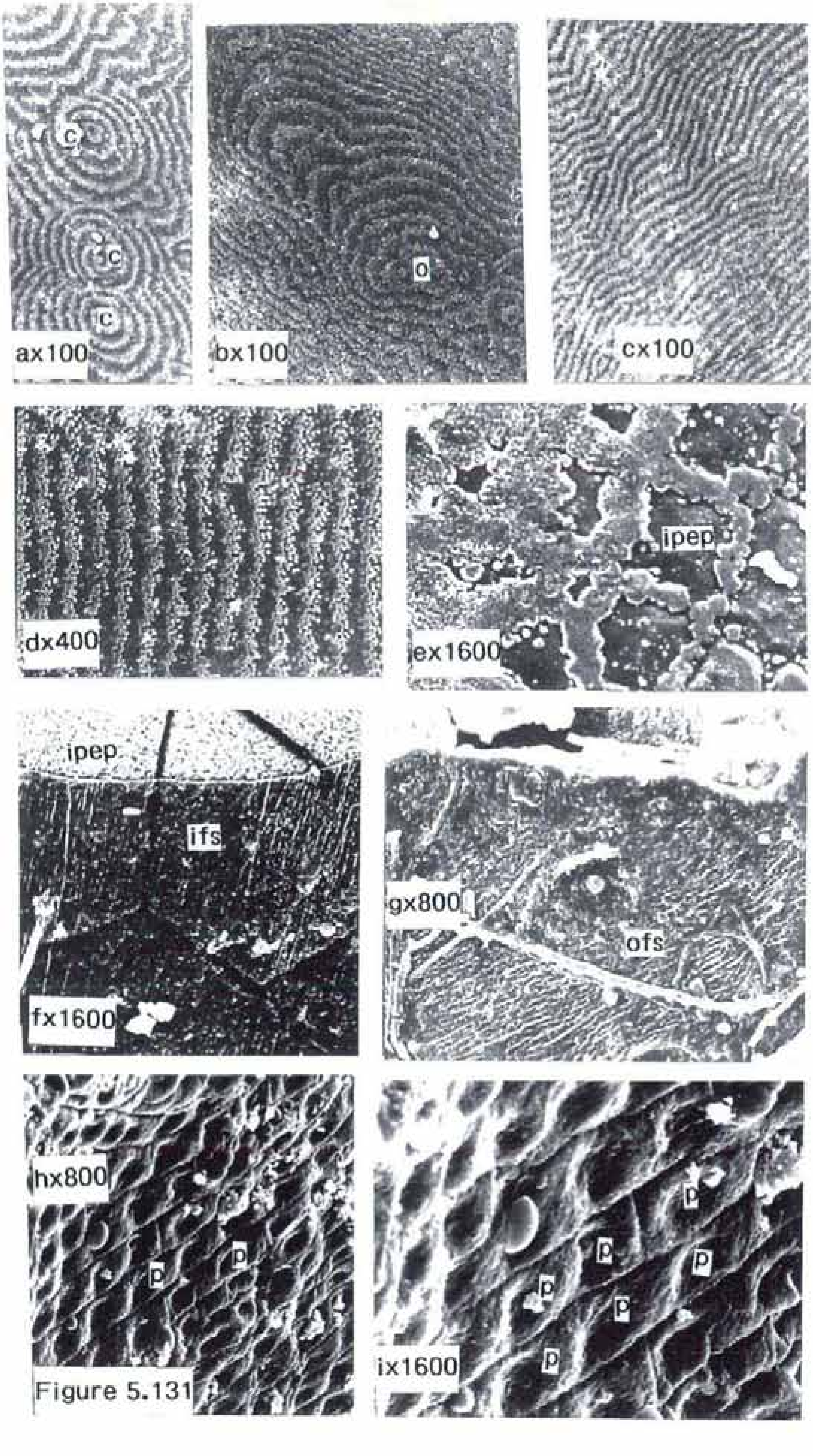
e. S.E.M. x 1600 Nacreo-prismatic Junction - medial view.

f. S.E.M. x 1600 Inner Fibrous Sheath - Distal Medial Growth Scale.

g. S.E.M. x 800 Outer Fibrous Sheath - Lateral Surface Prismatic Layer.

h. S.E.M. x 800 Growth Process - Inner Prismatic End Plates near edge.

i. S.E.M. x 1600 Growth Process - Inner Prismatic End Plates near edge.


c
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o
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cx100
dx400
ipep
ex1600
ipep
ifs
fx1600
gx800
ofs
hx800
p
p
p
p
p
p
p
p
ix1600

Figure 5.131

5.5.4.2. Mantle Margin and Secretory Structures of LatF2..

The Mantle Margin has three Folds. F1., F2. and F3. in Figure 5.132. a.

Proximal LatF1. has a subepithelium with numerous Trabecular Turquoise Glands (tt1 in Figure 5.132. a). Middle LatF1. has a Mantle Edge Gland (m in Figure 5.132. a). There are Ovoid Blue Glands in the parenchyma of Terminal F1. (o in Figure 5.132. a).

Trabecular Turquoise Glands of the External Mantle-F1. type are scattered throughout the MedF1. epithelium and subepithelium. (tt1 in Figure 5.132. a).

Apical Groove F1F2. has an Omega Gland which produces the Pleated Secretion of Groove F1F2.. (o and ps respectively in Figure 5.132. a). The Circum-pallial Nerve lies beside the Circum-pallial Sinus (n and si respectively in Figure 5.132. a).

The subepithelial glands of Proximal and Middle Lateral F2. form a continuous mass from the Circum-pallial Nerve to the middle of Middle LatF2.. (g in Figure 5.132. a and b).

The secretory glands of the subepithelium of Proximal LatF2.. (g in Figure 5.132. a), and Middle LatF2.. (g in Figure 5.132. b), secrete a material which stains bright red with Azan (s in Figure 5.132. b and c) and comes to lie in the epithelial folds of the middle and distal parts of Middle LatF2. medial to the Pleated Secretion of Groove F1F2. (ps in Figure 5.132. c).

Trabecular Turquoise Glands of the F2F3. type are rare on LatF2. but occur throughout the subepithelium of MedF2. and LatF3.. (tt 2 in

Figure 5.132. a).

The Mantle Margin of *Pinctada sp. 1* is distinguished from that of *Pinctada fucata* by the relatively greatly reduced concentration of Granular Cytoplasm Secretory Glands in the MedF1. subepithelium (Figure 5.132. a, compared with Figure 5.129. e). The secretory structures of LatF2. are invested by branches of the F2. ramis of the Circum-pallial Nerve (F2. rn in Figure 5.132. b).

FIGURE 5.132. *Pinctada sp. 1.* L.M. Mantle Margin and Secretion of Prismatic Layer Organic Matrices.

a. L.M. x 77 A.B./M.S.B. Mantle Margin.

b. L.M. x 770 Azan Proximal Part of Middle LatF2..

c. L.M. x 770 Azan Distal Part of Middle LatF2..

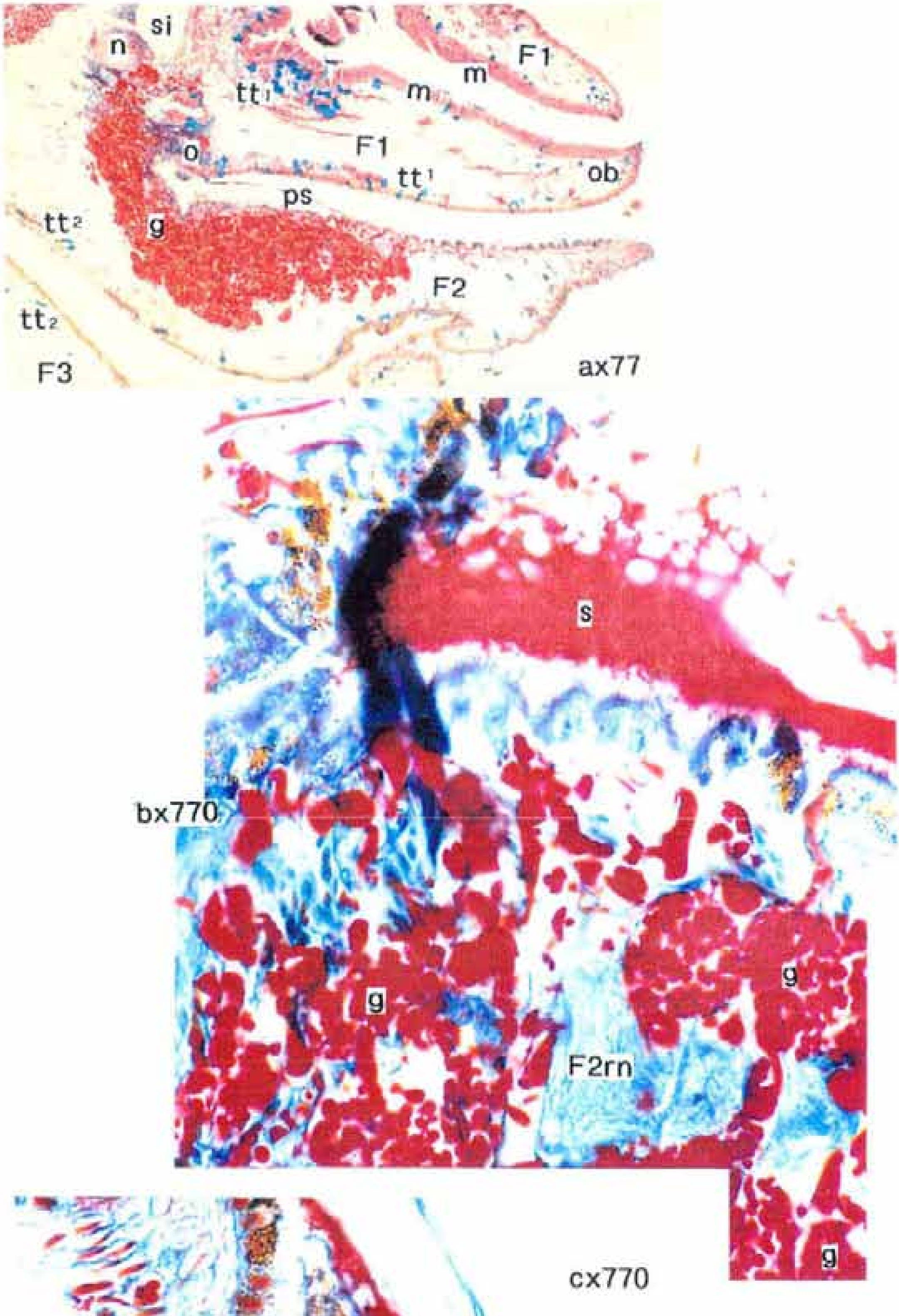

si
n
F1
m
tt1
m
o
F1
ob
tt1
ps
tt2
g
F2
tt2
F3
ax77
s
bx770
g
g
F2rn
g
cx770
s
ps

Figure 5.132

5.5.5. *PINCTADA SP. 2.*

5.5.5.1. Nacreous and Prismatic Layers of Valves.

The shapes of the Partly-bound and Free Nacre Tiles of the medial surface of the Inner Nacreous Layer may vary from truncated orthorhombic shapes in the Proximal Pallial Region (Figure 5.133. a) to approach regular hexagons in the Distal Pallial Region (Figure 51.33. b) on the same valve. Again, on the same valve, the medial surface of the Outer Nacreous Layer may have extended hexagon shapes of Nacre Tiles (Figure 5.133. c). Irrespective of shape the equivalent axes of the Nacre Tiles are aligned at a set angle to the direction of the edges of the Nacre Sheets in any area. (Figure 5.133. a, b and c).

The Outer Prismatic End Plates (opep in Figure 5.133 a) of the Major Prisms of the Prismatic Layer in this species bear concentric rings. (r. in Figure 5.133. d and e) which can be discerned through the Outer Fibrous Sheath (ofs in Figure 5.133. d).

In Figure 5.133. d the Outer Fibrous Sheath (ofs in Figure 5.133. e) has been eroded.

Near the edge of the Growth Processes the Inner Prismatic End Plates of the Growth Process Prisms have been constrained so that they are parallelograms in shape (p in Figure 5.133. g) and lie between parallel lines (Figure 5.133. f).

FIGURE 5.133. *Pinctada sp. 2.* Nacreous Layers and Prismatic Layer.

a. S.E.M. x 3200 Inner Nacreous Layer Proximal Pallial Region medial surface.

b. S.E.M. x 3200 Inner Nacreous Layer Distal Pallial Region medial surface.

c. S.E.M. x 3200 Outer Nacreous layer, medial surface.

d. S.E.M. x 1600 Lateral surface Prismatic Layer.

e. S.E.M. x 2500 Lateral surface Prismatic Layer.

f. S.E.M. x 200 Growth Process. Medial surface near edge.

g. S.E.M. x 1600 Growth Process. Medial surface near edge.

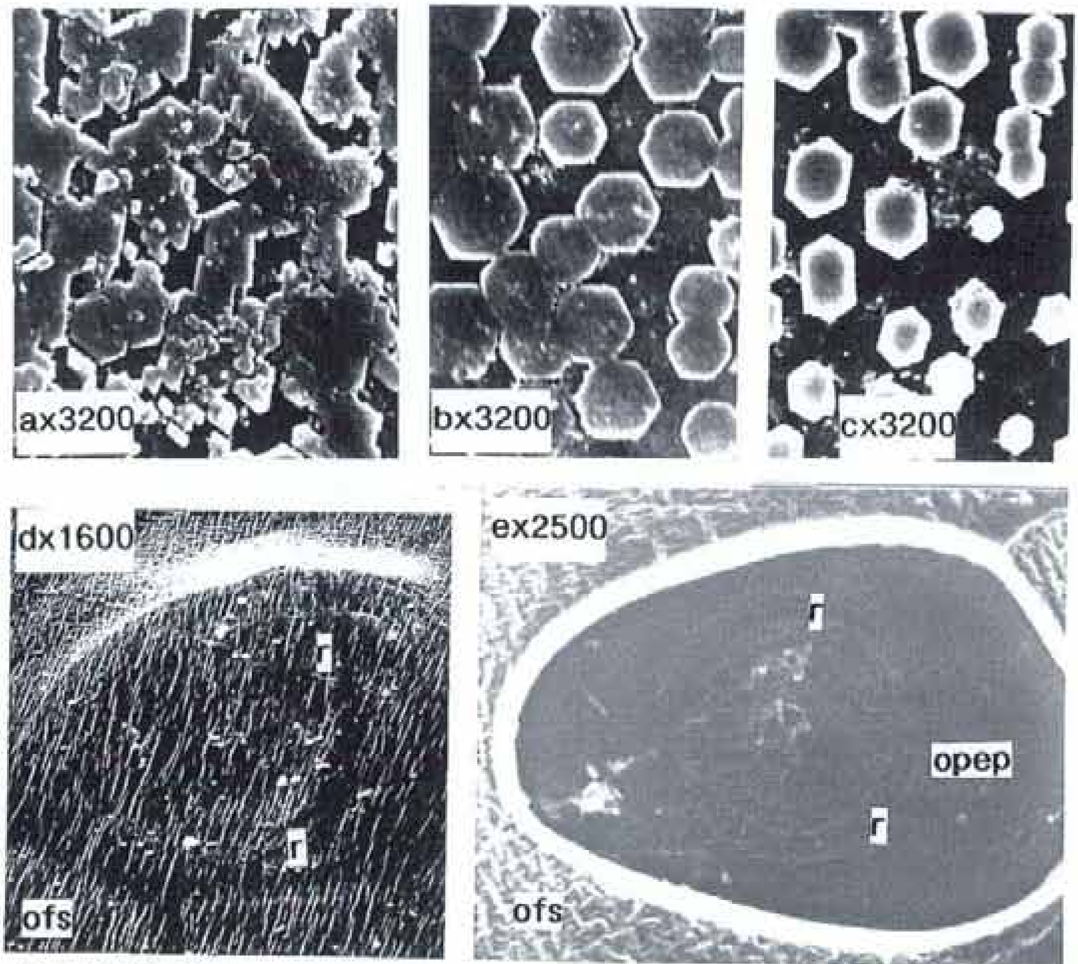
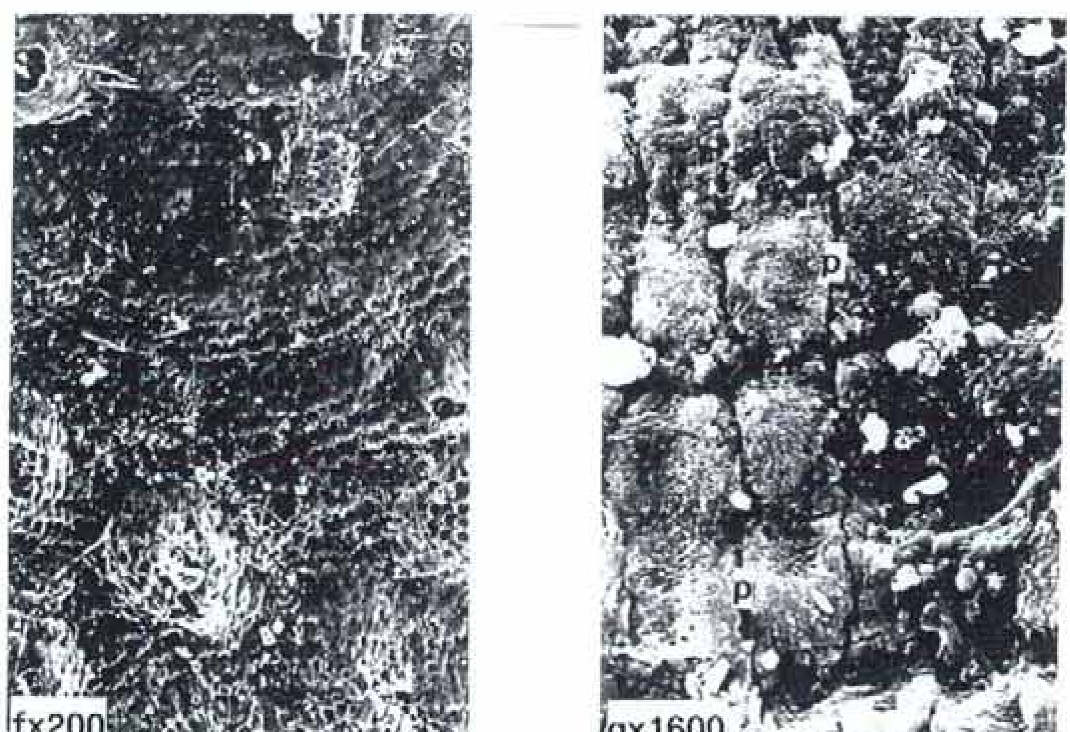

ax3200
bx3200
cx3200
dx1600
r
r
ofs
ex2500
r
opep
r
ofs

Figure 5.133


fx200
p
p
gx1600

5.5.5. *PINCTADA SP. 2*

5.5.5.2. Mantle Margin and Nacreous Organic Matrix Secretion by External Mantle and Distal Folded Region.

The External Mantle, Distal Folded Region, and Proximal LatF1.. (em, dfr and LF1. respectively in Figure 5.134. a) bear External Mantle-F1 Type Trabecular Turquoise Glands tt1 in Figure 5.134. a, c and d).

The LatF1. has a Mantle Edge Gland (m in Figure 5.134. a).

There is a relatively moderate density of Granular Cytoplasm Secretory Glands in the subepithelium of both MedF1. and LatF2.. (g, g in Figure 5.134. a, where F1. and F2. are marked F1. and F2).

Apical Groove F1F2. has an Omega Gland laterally and Dactylocytes medially (o and d respectively in Figure 5.134. a). Proximal to Apical Groove F1F2. are the Circum-pallial Nerve and Circum-pallial Sinus (n and si respectively in Figure 5.134. a).

The secretions of the Trabecular Turquoise Glands joins with agglomerated secretions from the Granular Cytoplasm Secretory Glands (TS and gs respectively in Figure 5.134. b, c and d) to form the Nacreous Organic Matrices above the Pallial Region (Figure 5.134. b) and the Distal Folded Region (Figure 5.134. e and d). The Nacreous Organic Matrices stain blue with Azan (nom in Figure 5.134. c) and turquoise with A.B./M.S.B. (nom in Figure 5.134 b and d).

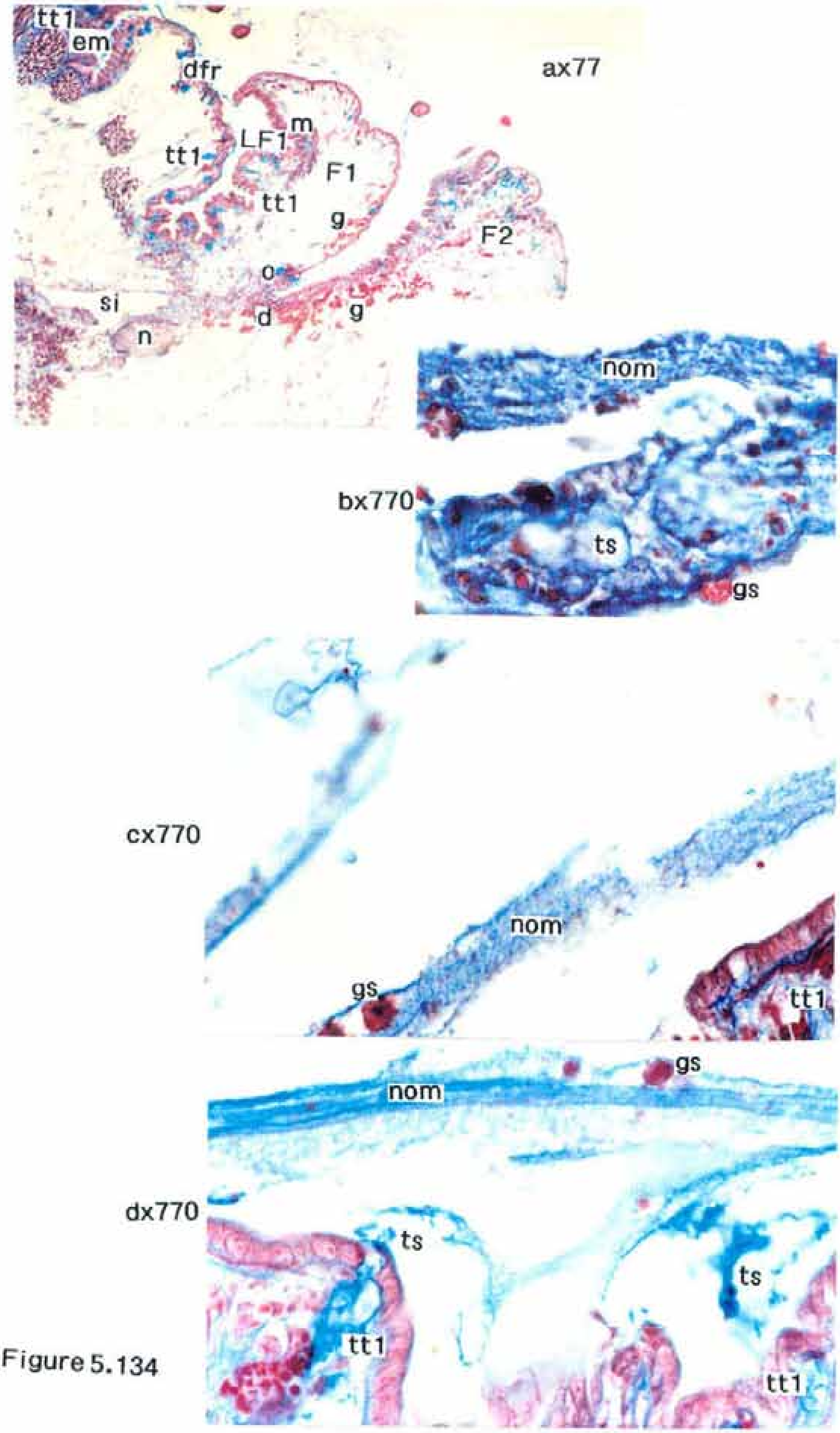
FIGURE 5.134. *Pinctada sp. 2*.- L.M. External Mantle, Mantle Margin and Secretion of Nacreous Organic Matrices.

a. L.M. x 77 A.B./M.S.B. Distal Folded Region and Mantle Margin Folds F1., F2.

b. L.M. x 770 A.B./M.S.B. Forming Nacreous Organic Matrix Pallial Region.

c. L.M. x 770 Azan Forming Nacreous Organic Matrix Distal Folded Region.

d. L.M. x 770 A.B./MS.S.B. Secretion of Trabecular Turquoise Glands of Distal Folded Region.


tt1
em
dfr
ax77
m
LF1
tt1
F1
tt1
g
F2
o
si
d
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n
nom
bx770
ts
gs
cx770
nom
gs
tt1
gs
nom
dx770
ts
ts
tt1
tt1

Figure 5.134

5.5.5.3. T.E.M. of Nacre Formation, Distal Folded Region.

The Organic Matrices in which the aragonite Nacre Tiles form are synthesized on the surface of the tissues which secrete the organic matrix precursors. At the edge of where the brickwork patters are forming are amorphous material, (presumably from Trabecular Turquoise Glands), granules and disintegrating granules from Granular Cytoplasm Secretory Glands, and cilia, (a, g and c respectively in Figure 5.135. a, b and c).

FIGURE 5.135. *Pinctada sp. 2.* T.E.M. of Formation of Nacreous Organic Matrix on Distal Folded Region.

a. T.E.M. x 7000

b. T.E.M. x 1800

c. T.E.M. x 11000 Higher magnification compared with area 1 in b above.

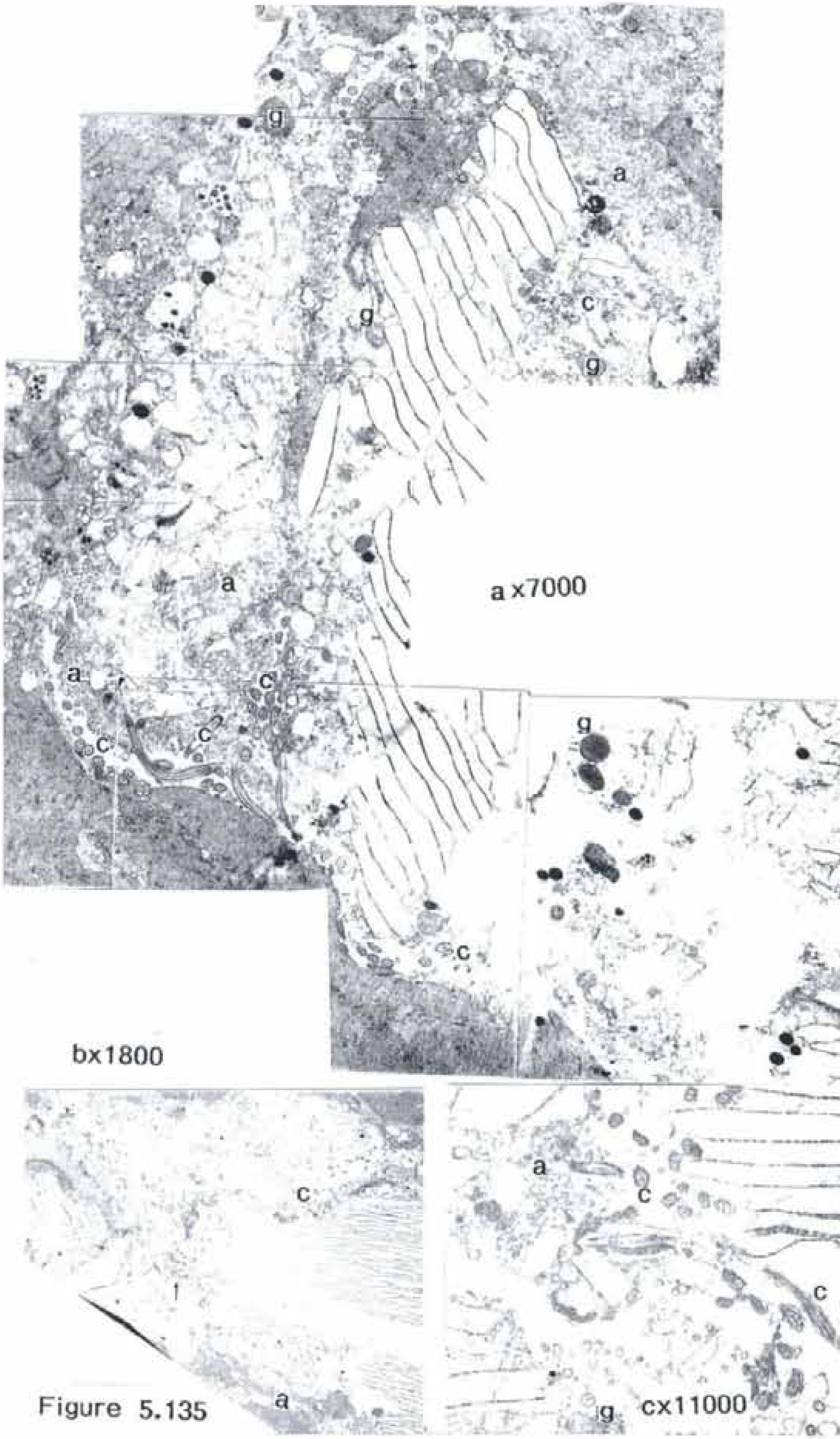


Figure 5.135

5.5.5.4. T.E.M. Nacreous Organic Matrix Formation - Middle MedF1..

Nacreous Organic Matrix (nom in Figure 5.136. a and b) forms on the surface of Middle MedF1. between the microvilli on the apical membrane of the surface epithelial cells and the Pleated Secretion of Groove F1F2. (m and ps respectively in Figure 5.136. a).

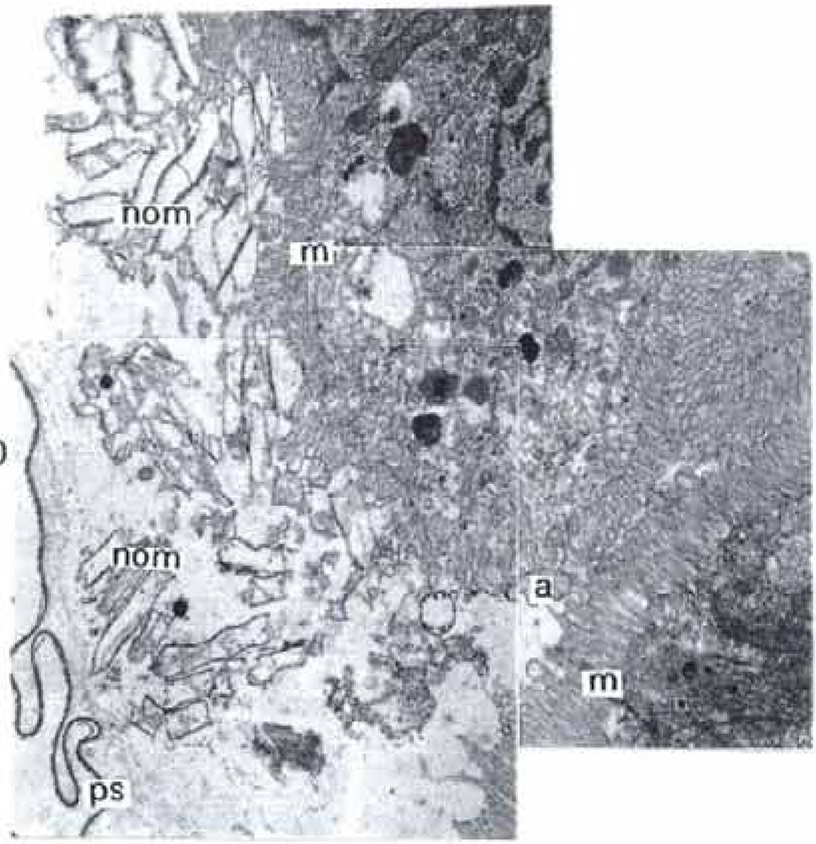
The Nacreous Organic Matrix appears to form in the presence of amorphous secreted material. disintegrating granules from Granular Cytoplasm Secretory Glands and in places secreted pigment granules (a. g and p respectively in Figure 5.136. b).

FIGURE 5.136. *Pinctada sp. 2* - Nacreous Organic Matrix Formation. Middle MedF1..

a. T.E.M. x 7000

b. T.E.M. x 7000

ax7000



b
x7000

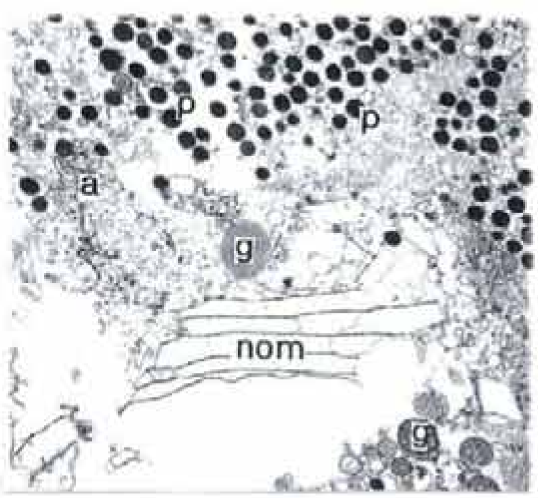


Figure 5.136

5.5.5.5. Pleated Secretion of Groove F1F2. and Vesiculate Secretion of Groove F1F2..

The Pleated Secretion of Groove F1F2. which issues from the Apical Channel between the Omega gland laterally and the Dactylocytes medially has a lighter middle part between two thinner electron dense outer lamina (Figure 5.137. a). Electron dense Granules from the Black Granule Secretory Cells are attached at regular intervals to the medial surface (bg in Figure 5.137. a).

The Vesiculate Secretion of Groove F1F2. is formed from vesiculate secretions of Proximal LatF2. (v in Figure 5.137c) and carries electron dense granules from the Black Granule Secretory Cells on its lateral surface. (bg in Figure 5.137. c).

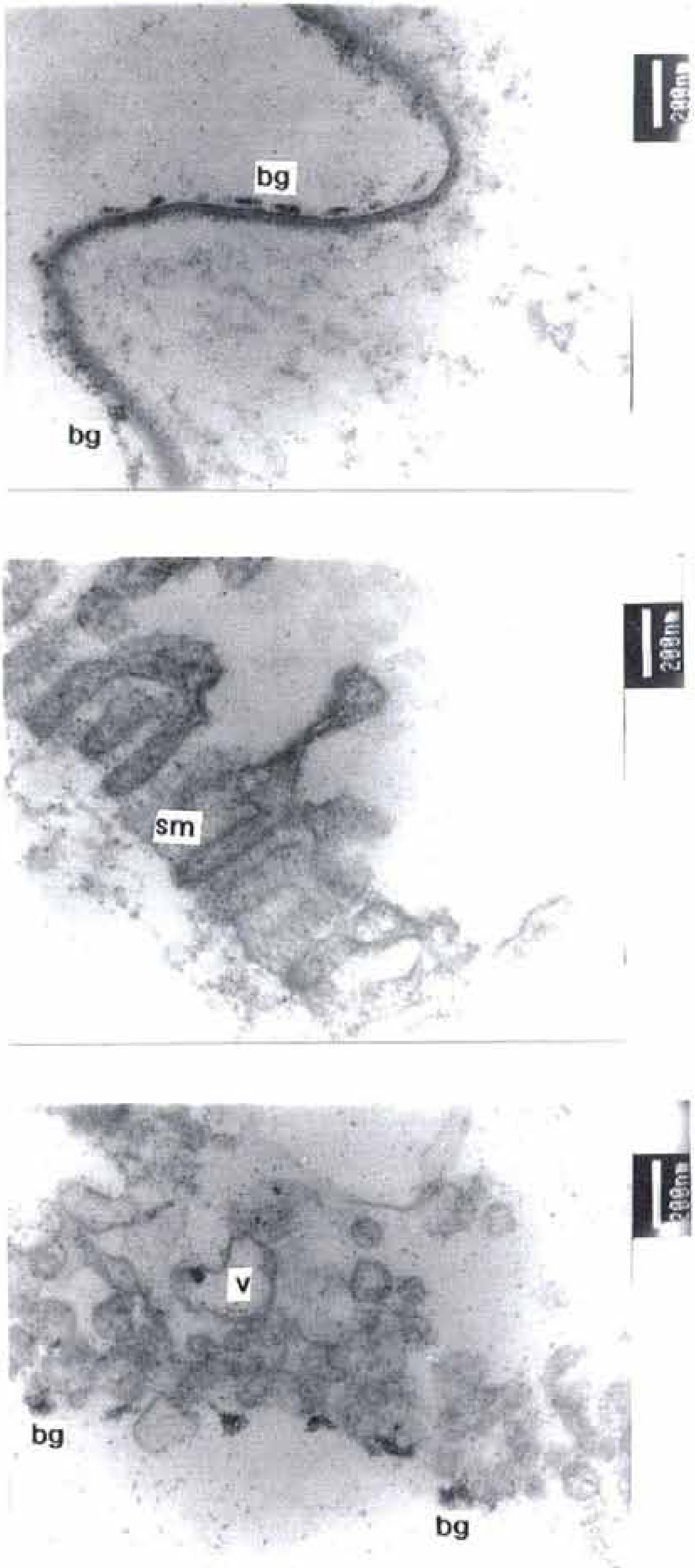
As the secretion passes distally down groove F1F2. the large vesicles are incorporated in a secretory matrix. (sm. Figure 5.137. b).

FIGURE 5.137. *Pinctada sp. 2.* - T.E.M. of Pleated Secretion of Groove F1F2., and Vesiculate Secretion of Groove F1F2..

a. T.E.M. x 53000 Pleated Secretion of Groove F1F2..

b. T.E.M. x 53000 Vesiculate Secretion of Groove F1F2..

c. T.E.M. x 53000 Vesiculate Secretion of Groove F1F2..



a
x53000

b
x53000

c
x53000

Figure 5.137

5.5.6. *PINCTADA SP. 3.*

5.5.6.1. Mantle Margin of *Pinctada sp. 3.*

Pinctada sp. 3 is a relatively small pearl oyster with a dark coloured Prismatic Layer and purple coloured nacre, especially the Outer nacreous Layer (Figure 3.3. a and c).

There are three Marginal Mantle Folds. F1. and F2. are shown in Figure 5.138. a.

External Mantle-F1. type Trabecular Turquoise Glands are conspicuous in the subepithelium of the Distal Folded Region and Middle MedF1., (tt in Figure 5.138. a and b). The surface epithelium of Proximal medF1. is heavily pigmented (P in mfs in Figure 5.138. a and b). LatF2. surface epithelium is less conspicuously pigmented, as is MedF2. (lf2 and mf2 respectively in Figure 5.138. a).

5.5.7. *PINCTADA SP. 4.*

5.5.7.1. Mantle Margin of *Pinctada sp. 4.*

The Valves of *Pinctada sp. 4.* are distinguished from *Pinctads sp. 3.* partly by the more silvery nacre, especially the Outer Nacreous Layer (Figure 3.3. c).

The most obvious difference in the histology of the Mantle Margin of *Pinctada sp. 4.* compared with *Pinctada sp. 3* is the almost total lack of pigment granules in the surface epithelium of Proximal MedF1. (mfi in Figure 5.138. c).

The surface epithelium of LatF2. is again pigmented (P in Figure 5.138. c).

FIGURE 5.138. Mantle Margins of *Pinctada sp. 3* and *Pinctada sp. 4.*

a. L.M. x 150 A.B./M.S.B. *Pinctada sp. 3.* Mantle Margin.

b. L.M. x 770 A.B./M.S.B. *Pinctada sp. 3.* MedF1..

c. L.M. x 770 A.B./M.S.B. *Pinctada sp. 4.* Proximal MedF1., Apical Groove F1F2., Proximal LatF2..

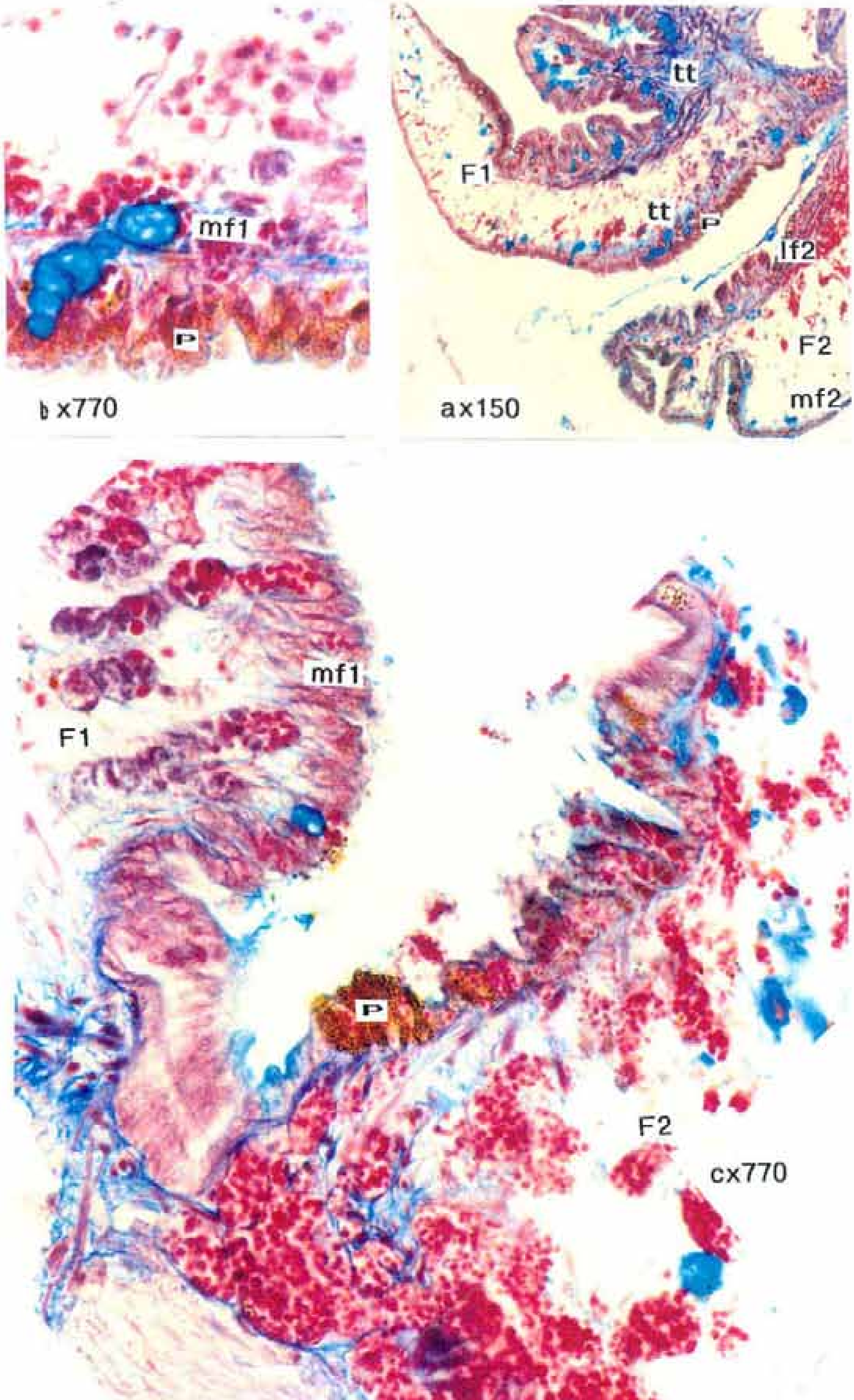


Figure 5.138

5.5.8. *PINCTADA CHEMNITZI*

The Valves of this species have a Prismatic Layer which is relatively very thin and is a uniform dull yellow-brown colour (PL in Figure 5.139. a). The Growth Processes are commonly so small as to be seen only with magnification (GP in Figure 3.3 g). The area of Nacreous Layer is noticeably greater in the left Valve than the Right Valve and in both cases commonly terminates distally well short of the Ventral periphery. (NL. Figure 5.139. a).

The Nacre has a clear silver lustre.

Both the External Mantle and more especially the Mantle Margin have a relatively diminished display of most kinds of secretory glands. (Figure 5.139. b and c show more abundant subepithelial glands than in some specimens of the Mantle Margin of this species.)

The Mantle Edge Gland (m.e.g. in Figure 5.139. b) is relatively inconspicuous as are the secretory strucutres of MedF1. The Omega Gland (O in Figures 5.139. b and c) and Dactylocytes (d in Figure 5.139. b and c) produce a copious Pleated Secretion of Groove F1F2. (ps in Figure 5.139. b and c).

There are relatively large numbers of Microgranular Glands in the Parenchyma of F2. and F3. (mg in Figure 5.139. b and c).

FIGURE 5.139. *Pinctada chemnitzi* - Valves and Mantle Margin.

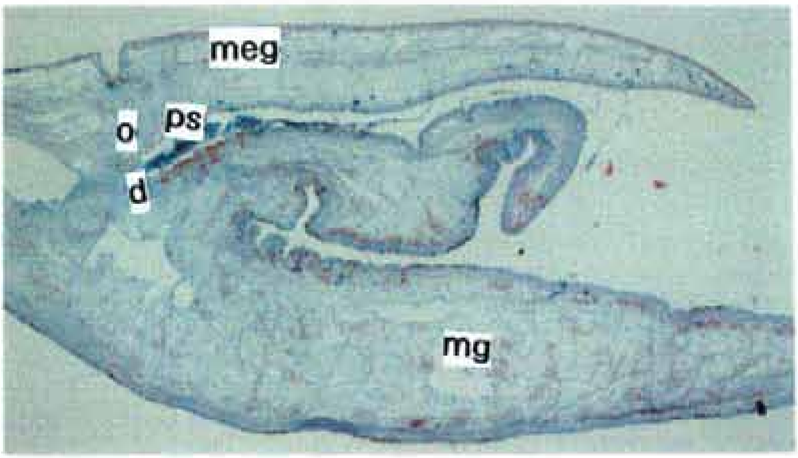
a. x 0.7 Medial surface of Valves.

b. L.M. x 77 A.B./M.S.B. Mantle Margin.

c. L.M. x 150 A.B./M.S.B. Groove F1F2., Groove F2F3..

ax0.7

bx77



cx150

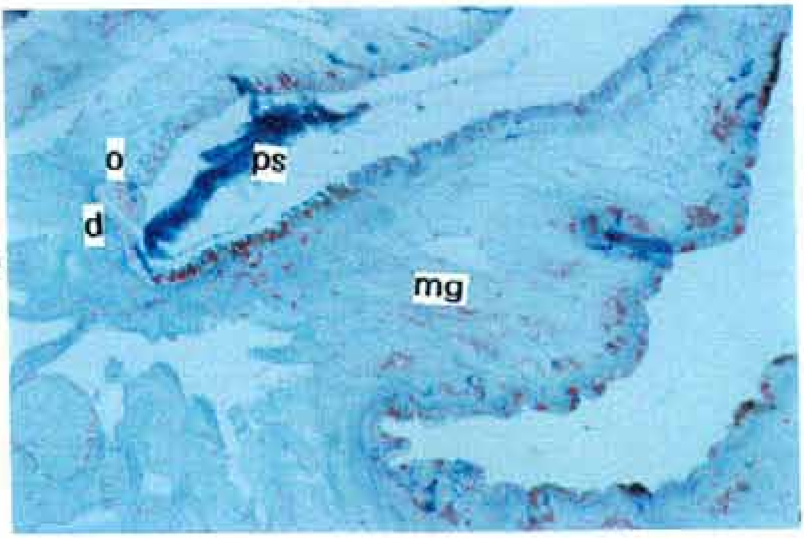


Figure
5.139

5..5.9. *PINCTADA SP. 5.*

The Valves of this species display relatively large coarse Prismatic Layer Growth Processes which are porcelain white in colour (gp in Figure 5.140. a).

The Nacreous Layers (nl in Figure 3.3 d) cover almost the entire medial surface of the Valves.

MedF1. has numerous subepithelial Granular Cytoplasm Secretory Glands and Trabecular Turquoise Gland (gc and tt in Figure 5.140. b). The Pleated Secretion (ps in Figure 5.140. b and c) issues from the Apical Channel (ac in Figure 5.140. a) and divides the forming Outer Nacreous Layer Organic Matrix (onom in Figure 5.140. c) from the Vesiculate Secretion of Groove F1F2. (vs in Figure 5.140. d).

Middle LatF2. is typical for the *Pinctada* (mLF2. in Filgure 5.140. d) and contributes to the Vesiculate Secretion. There is an unusually heavy concentration of Granular Cytoplasm Secretory Glands and Trabecular Turquoise Glands in the Subepithelium of MedF2. (gc and tt in Figure 5.140. e).

FIGURE 5.140. *Pinctada SP.5.* –Left Valve, Mantle Margin and Secretions.

a. x 1.5. Left Valve lateral view (repeated from Figure 3.3. e).

b. L.M.x 150 A.B./M.S.B. Apical Groove F1F2..

c. L.M. x 770 A.B./M.S.B. Forming Outer Nacreous Layer and Pleated Secretion.

d. L.M. x 770 A.B./M.S.B. Middle Lateral F2. and Vesiculate Secretion.

e. L.M. x 770 A.B./M.S.B. Middle MedF2..

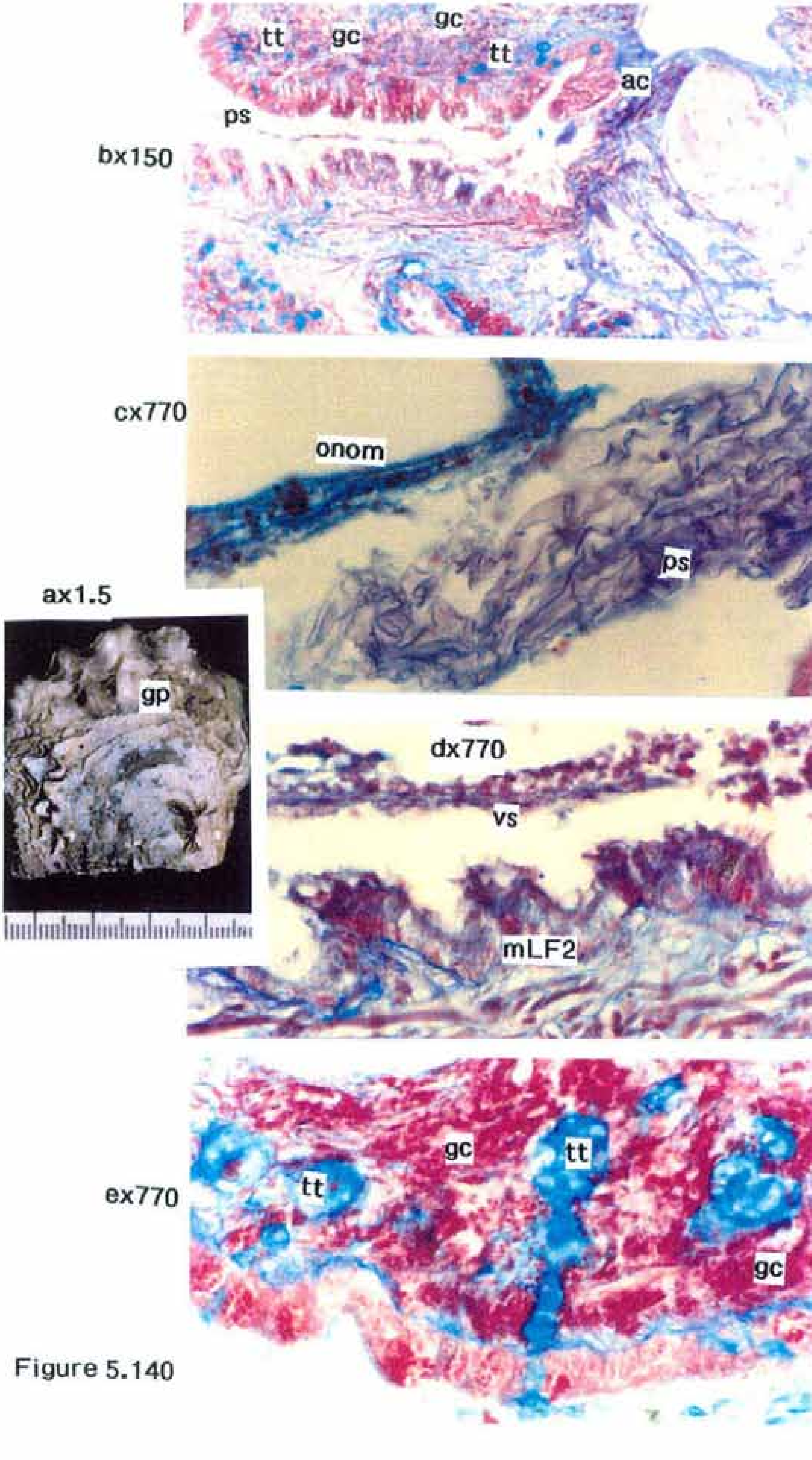

gc
tt
gc
tt
ac
ps
bx150
cx770
onom
ps
ax1.5
gp
dx770
vs
mLF2
gc
tt
tt
ex770
gc

Figure 5.140

5.5.10. *PINCTADA ALBINA SUGILLATA.*

In Figure 5.141, L is lateral and M medial.

In this species the secretory tissues of the External Mantle are unremarkable. The Mantle Margin is species specific.

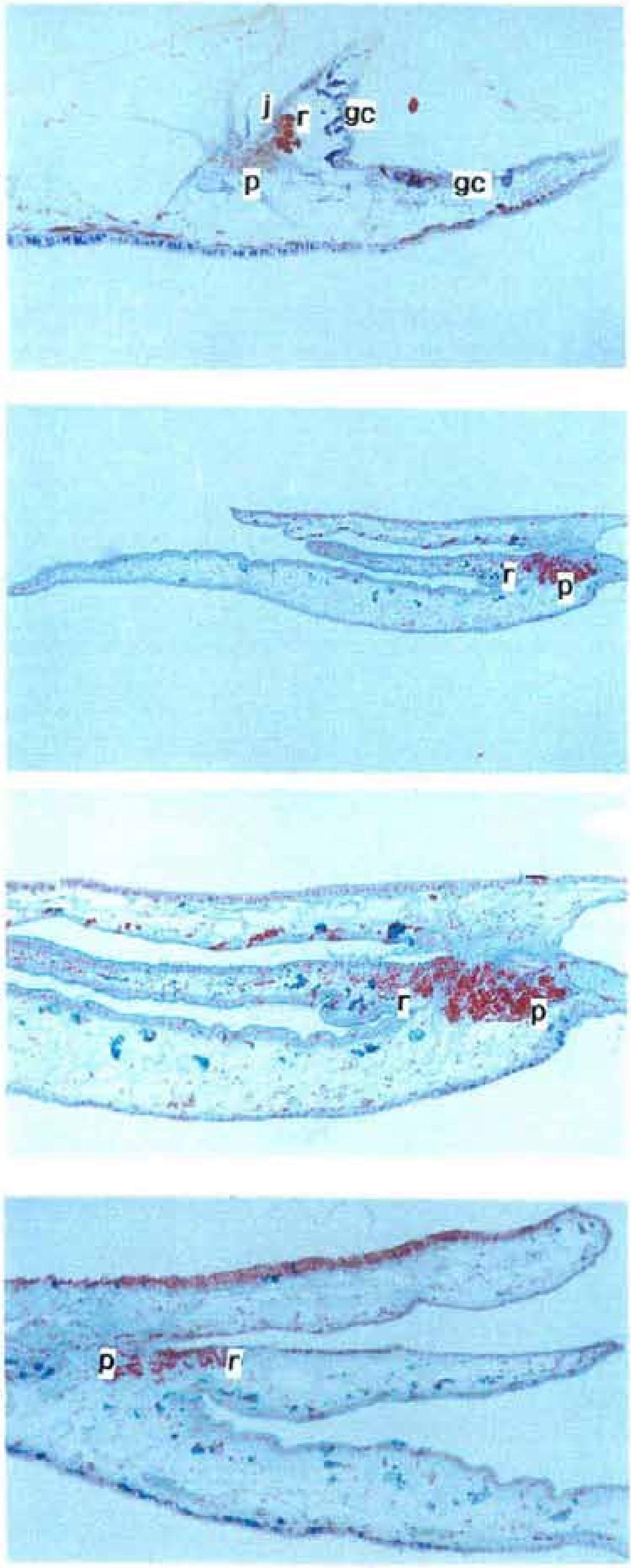
The subepithelial Granular Cytoplasm Secretory Glands of Proximal LatF2. stain pink with Azan (p in Figure 5.141. a) and secrete into Receptacle Glands in the Proximal LatF2. epithelium. This epithelium has few cilia (seen only with T.E.M.). With A.B./M.S.B. these glands stain brick red (p in Figure 5.141. b, c and d.).

At the line of junction between the Proximal LatF2. and the Middle LatF2. the surface epithelium abruptly changes to one with numerous elongate cilia as well as a dense mat of microvilli (j in Figure 5.141, a). Immediately distal to the proximal commencement of this ciliated epithelium it is crossed by the secretory tubules of large unicellular Granular Cytoplasm Secretory Glands which secrete directly into the Middle LatF2. part of Groove F1F2.. These Glands stain dark red with Azan (r in Figure 5.141. a). With A.B./M.S.B. these glands stain purplish-red (r in Figure 5.141. b, c and d).

There are large Granular Cytoplasm Secretory Glands in the subepithelim of MedF2. and LatF3. which stain blue and red with Azan (gc in Figure 5.141. a).

FIGURE 5.141. *Pinctada albina sugillata* - Mantle Margins.

a.	L.M. x 77	Azan	Three Marginal Mantle Folds.
b.	L.M. x 77	A.B./M.S.B.	As for a above.
c.	L.M. x 150	A.B./M.S.B.	As for a and b above.
d.	L.M. x 150	A.B./M.S.B.	As for a, b, and c above.



ax77

bx77

cx150

dx150

Figure 5.141

5.5.11. *PINCTADA SP. 6.*

5.5.12. *PINCTADA SP. 7.*

5.5.13. *PINCTADA SP. 8.*

5.5.14. *PINCTADA SP. 9.*

5.5.15. *PINCTADA SP. 10.*

These five pearl oysters are all distinguished from *Pinctada albina sugillata* by the histology of their Mantle Margin. *Pinctada sp 6.* (Figure 5.142. a), has numerous subepithelial Trabecular Turquoise Glands on MedF2. and LatF3. but lacks the Granular Cytoplasm Secretory Glands of these regions seen in *Pinctada albina sugillata* (Figure 5.141. a). *Pinctada sp 7.* (Figure 5.142. b), has a population of Granular Cytoplasm Secretory Glands throughout the subepithelium of F2. and LatF3. which stain red with Azan and lacks the pink staining glands of Proximal LatF2. and the red staining glands of Middle LatF2. seen in *P. albina sugillata* with this stain. The tissues of the Mantle Margin of *Pinctada sp. 8.* (Figure 5.142. c), are very similar to those of *P. albina sugillata* in MedF2. and LatF3. However the glands of Proximal LatF2. are nearly colourless with Azan in this species and the equivalent, to the Middle LatF2. Glands of *Pinctada albina sugillata* stain brown with Azan in this species. LatF2. of *Pinctada sp. 9.* (Figure 5.142. d), carries a species specific secretory tissue of elongate tubular secretory glands which stain dark red with Azan. The Proximal MedF1. of *Pinctada sp. 10* (Figure 5.142. e), has a dense population of Granular Cytoplasm Secretory Glands which, unique to this species of those studied, stain purple with A.B./M.S.B..

FIGURE 5.142. *Pinctada sp. 6, Pinctada sp. 7, Pinctada sp. 8, Pinctada sp. 9.* and *Pinctada sp. 10.* – L.M. Radial Sections of Mantle Margins.

a. L.M. x 77 A.B./M.S.B. *Pinctada sp. 6.*

b. L.M. x 77 A.B./M.S.B. *Pinctada sp. 7.*

c. L.M. x 150 Azan *Pinctada sp. 8.*

d. L.M. x 150 Azan MedF1. and LatF2. *Pinctada sp. 9.*

e. L.M. x 770 A.B./M.S.B. Middle Med F1., *Pinctada sp. 10.*

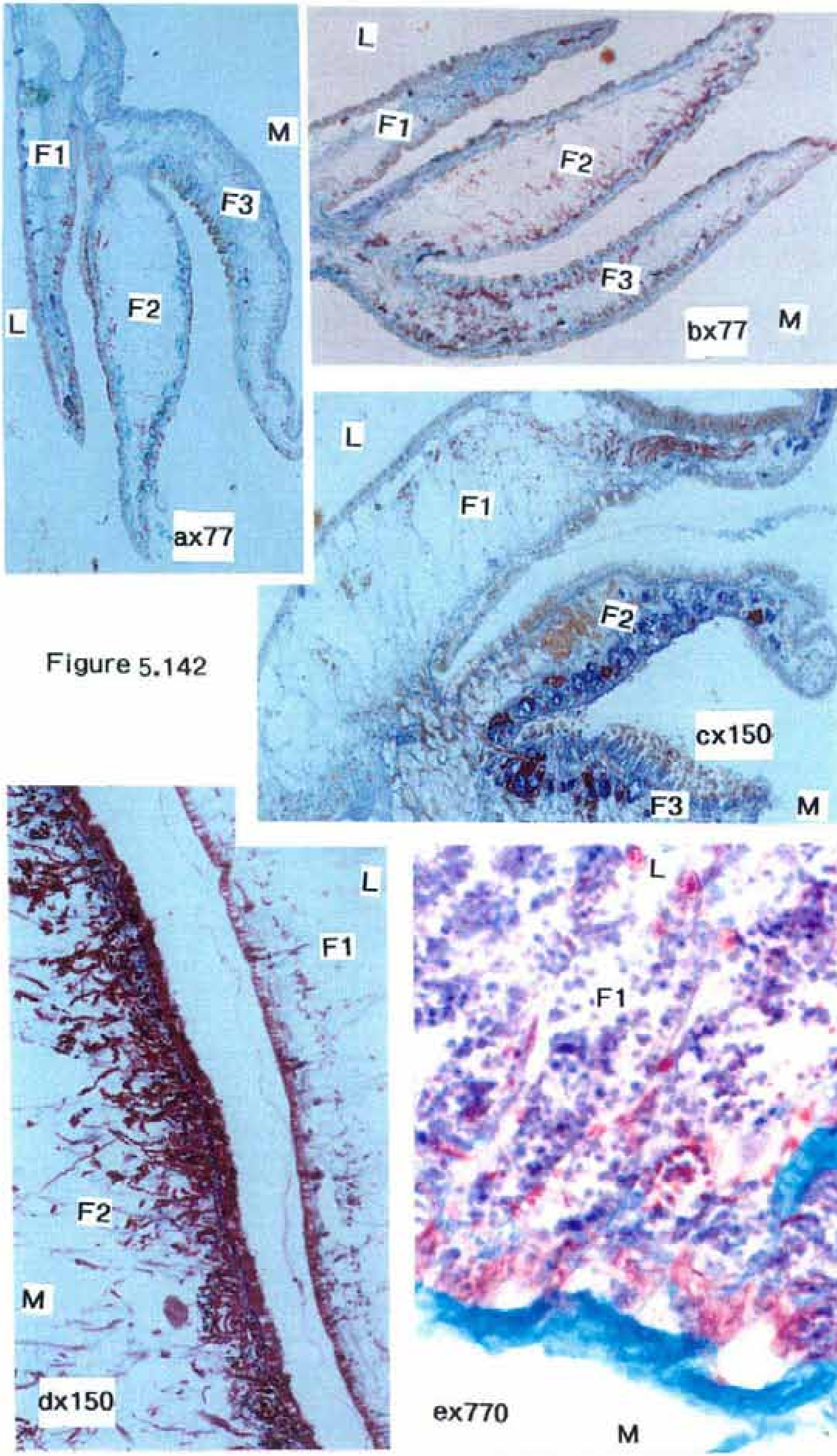

F1
M
F3
F2
L
ax77
L
F1
F2
F3
bx77
M
L
F1
F2
cx150
F3
M
L
F1
F2
M
dx150
L
F1
ex770
M

Figure 5.142

5.5.16. *PTERIA PENGUIN*

5.5.16.1. Inner and Outer Nacreous Layers.

The Inner Nacreous Layer of *Pteria penguin* has patterns to the edges of the Nacre Sheets which broadly reflect those seen in the *Pinctada*. That is, the Shoulder Region and Pallial Gland Region dispaly numerous concentric circular and less commonly spiral patterns and strongly curved curvilinear patterns with numerous anastomoses. The Anterior Pallial area and Proximal and Middle Ventral Pallial areas (Figure 5.143. a, b and c) display short strongly curved patterns with numerous anastomoses. Distally and Posteriorly from the above Regions the patterns more closely approach narrowly spaced parallel straight line patterns. However in this species as distinct from the species of the *Pinctada*, the parallel linear patterns, especially near the shell periphery often lie normal to the adjacent Nacreo-primsatic Junction.

The Inner Nacreous Layer Nacre Tiles are either diamond shaped or more usually, slightly truncated diamond shaped.

The Outer Nacreous Layer anteriorly displays a pattern of anastomosing and tightly curved edges of Nacre Sheets. However Ventrally and Posteriorly the Outer Nacreous Layer tends to closely spaced parallel linear patterns of edges of Nacre Sheets which are about parallel to the respective Nacreo-prismatic Junextions, (Figure 5.143. e and f).

The Nacre Tiles of the Outer Nacreous Layer in this species are usually diamond shaped (Figure 5.143. f) and the edges of the Free and Partly-bound Nacre Tiles are relatively rounded and indistinct (Figure 5.143. f and g).

The thicknesses of the Nacre Sheets of both the Outer and Inner Nacreous Layers are about 0.6 µm. (Figure 5.143. d and g).

FIGURE 5.143. *Pteria penguin* – S.E.M. Inner and Outer Nacreous Layers.

a.	S.E.M. x 400	Inner Nacreous Layer – Middle Ventral Pallial Region medial view.
b.	S.E.M. x 1600	As for a. above at higher magnification.
c.	S.E.M. x 3200	Middle Ventral Pallial Region.
d.	S.E.M. x 3200	Middle Ventral Pallial Region – radial broken surface.
e.	S.E.M. x 800	Outer Nacreous Layer – proximal to Ventral Nacreo -prismatic Junction medial view.
f.	S.E.M. x 6400	As for 3 above at higher magnification – medial view.
g.	S.E.M. x 3200	Outer Nacreous Layer – near Ventral Nacreo -prismatic Junction radial broken surface.

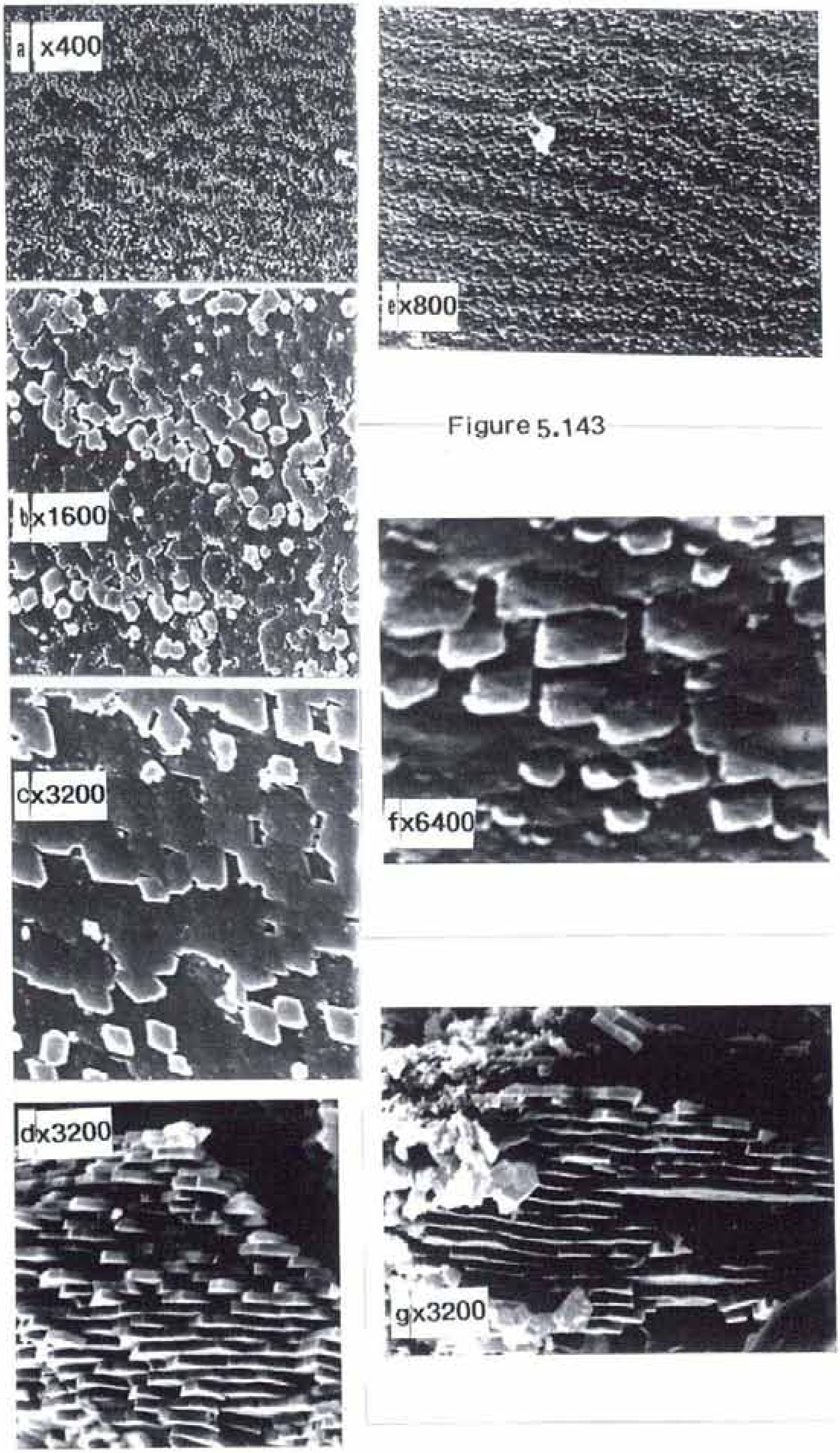


Figure 5.143

5.5.16. *PTERIA PENGUIN* (continued).

5.5.16.2. Prismatic Layer - Medial and Lateral Surfaces.

The Medial surface of the Prismatic Layer just distal to the peripheral Nacreo-prismatic Junction is devoid of an Inner Fibrous Sheath. (Figure 5.144. a). More distally the Inner Prismatic End Plates are covered by a fibrous Sheath of fibre periodicity about 0.3 µm (Figure 5.144. b).

The laterally exposed Outer Prismatic End Plates are covered by an Outer Fibrous Sheath of surface fibre periodicity of about 1.0 µm (Figure 5.144. c).

The Growth Processes of this species are distally finely pointed with recurved sides (Figure 5.144. d). and are covered by an Outer Fibrous Sheath of external fibre periodicity of about 0.7 µm. (Figure 5.144. e).

FIGURE 5.144. - *Pteria penguin* - S.E.M. Prismatic Layer, medial and lateral surfaces.

a. S.E.M. x 1600 Medial surface, bare Inner Prismatic End Plates just distal to the Nacreo-prismatic Junction.

b. .S.E.M. x 6100 Medial surface, Inner Fibrous Sheath covering distal Inner Prismatic End Plates.

c. S.E.M. x 1700 Outer Fibrous Sheath covering Lateral surface of Prismatic Layer.

d. S.E.M. x 27 Lateral surface. Four superimposed Growth Scales.

e. S.E.M. x 3200 Lateral surface. Outer Fibrous Sheath of Growth Scale.

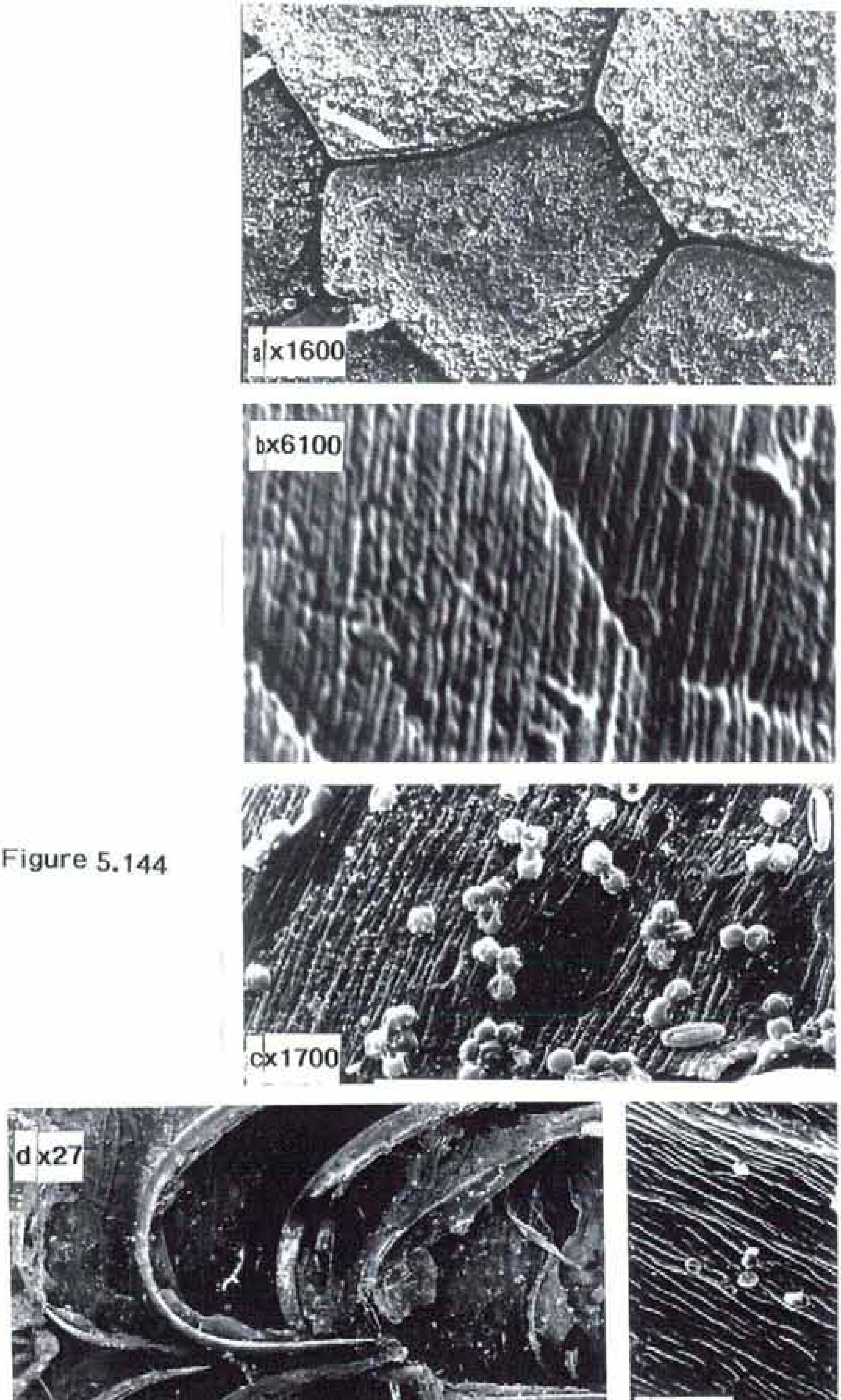


Figure 5.144

5.5.16. *PTERIA PENGUIN*

5.5.16.3. Prismatic Layer and Nacreo-prismatic Junction - Radial Broken Surface.

The Growth Scales of the Prismatic Layer of this species are unually clearly defined for the whole (or most) of their radial length by the Major Prisms of each Growth Scale possessing Outer and Inner Prismatic End Plates. These show as continuous lines in radial section of decalcified shell (peps in Figure 5.145. f). In S.E.M. of the radial broken surface the Inner Prismatic End Plates (ipep in Figure 5.145. b, c, d and e) appear to be edged by a sheet of material (s in Figure 5.145. c and d). This sheet appears to join laterally with both Linear Pattern Interprismatic Organic Matrix (lp in Figure 5.145. d), and Reteform Interprismatic Organic Matrix (r in Figure 5,145., c and d).

Transverse Linear Interprismatic Organic Matrix is the major residue of the Prismatic Layer following decalcification (TL Figure 5.145. f) and is sometimes seen with S.E.M. of radial broken Prismatic Layer (TL in Figure 5.145. c). Nacreous Organic Matrix residual after acid decalcification is finely fibrous radially and stains blue with azan, (nom inFigure 5.145, f). At the peripheral Nacreo-prismatic Junction free Nacre Tiles (NT Figure 5.145. e) lie in the grooves between the bare Inner Prismatic End Plates (IPEP Figure 5.145. e).

FIGURE 5.145. *Pteria Penguin* -S.E.M. Prismplpatic Layer and Nacreo -prismatic Junction - Radial broken surface. L.M. Radial Section decalcified Prismatic Layer and Nacreous Layers.

a. S.E.M. x 300 Five successive Growth Scales.

b. S.E.M. x 800 Inner Prismatic End Plates and Inter-prismatic Organic Matrices.

c. S.E.M. x 1600 Higher magnification of part of b above.

d. S.E.M. x 3200 Higher magnification of c above.

e. S.E.M. x 800 Ventral Nacreo-prismatic Junction.

f. L.M. x 42 Azan. Radial section of decalcified shell.

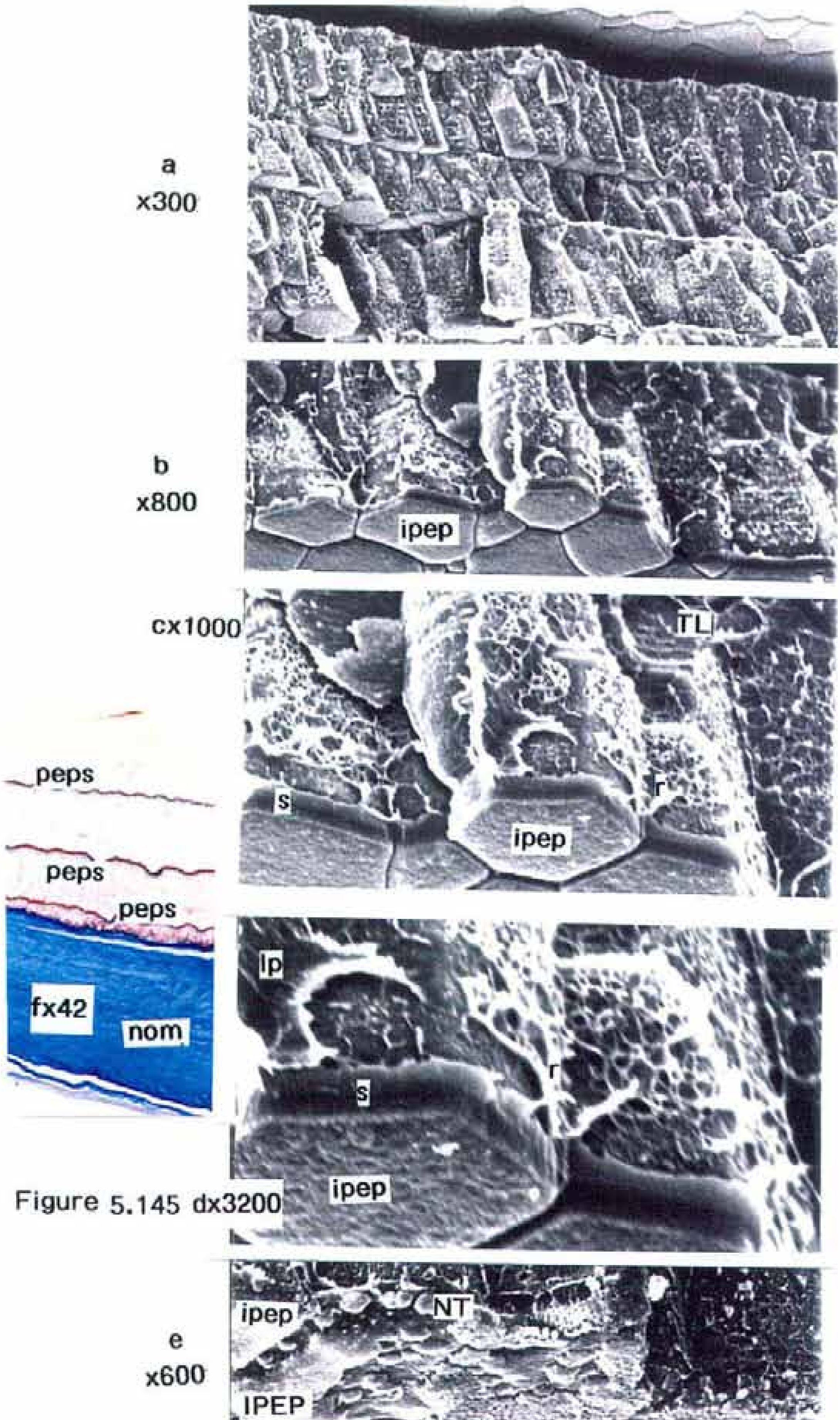

a
x300
b
x800
ipep
cx1000
TL
r
s
ipep
peps
peps
peps
fx42
nom
lp
r
s
ipep
dx3200
ipep
NT
e
x600
IPEP

Figure 5.145

5.5.16. *PTERIA PENGUIN*

5.5.16.4. External Mantle and LatF1. of Mantle Margin.

The Pallial Gland of this species has a strong fibrous connective tissue layer (ct in Figure 5.146. a) between the surface epithelium and the bodies of the Trabecular Turquoise Glands (tt in Figure 5.146. a). Deep to the latter are the saccular bodies of large Granular Cytoplasm Secretory Glands (gc in Figure 5.146. a). Although the secretory tissues are somewhat reduced in thickness and density, this spatial relationship of these two types of Glands is largely maintained throughout the Proximal, Middle and Distal Pallial Regions of this species (tt and gc in Figure 5.146. b, c and d).

In the Distal Folded Region there are very few Granular Cytoplasm Secretory Glands and the Trabecular Turquoise Glands are often intraepithelial (tt in Figure 5.146. e).

The Mantle Edge Gland (meg in Figure 5.146 f) is an elongate columnar epithelium all but devoid of unicellular secretory glands in the subepithelium.

FIGURE 5.146. *Pteria penguin* - L.M. of External Mantle and LatF1. of Mantle Margin.

a. L.M. x 310 Mallory's. Pallial Gland.
b. L.M. x 770 Mallory's. Proximal Pallial Region.
c. L.M. x 770 Mallory's. Middle Pallial Region.
d. L.M. x 770 Mallory's Distal Pallial Region.
e. L.M. x 770 Mallory's Distal Folded Region.
f. L.M. x 770 Mallory's Mantle Edge Gland.

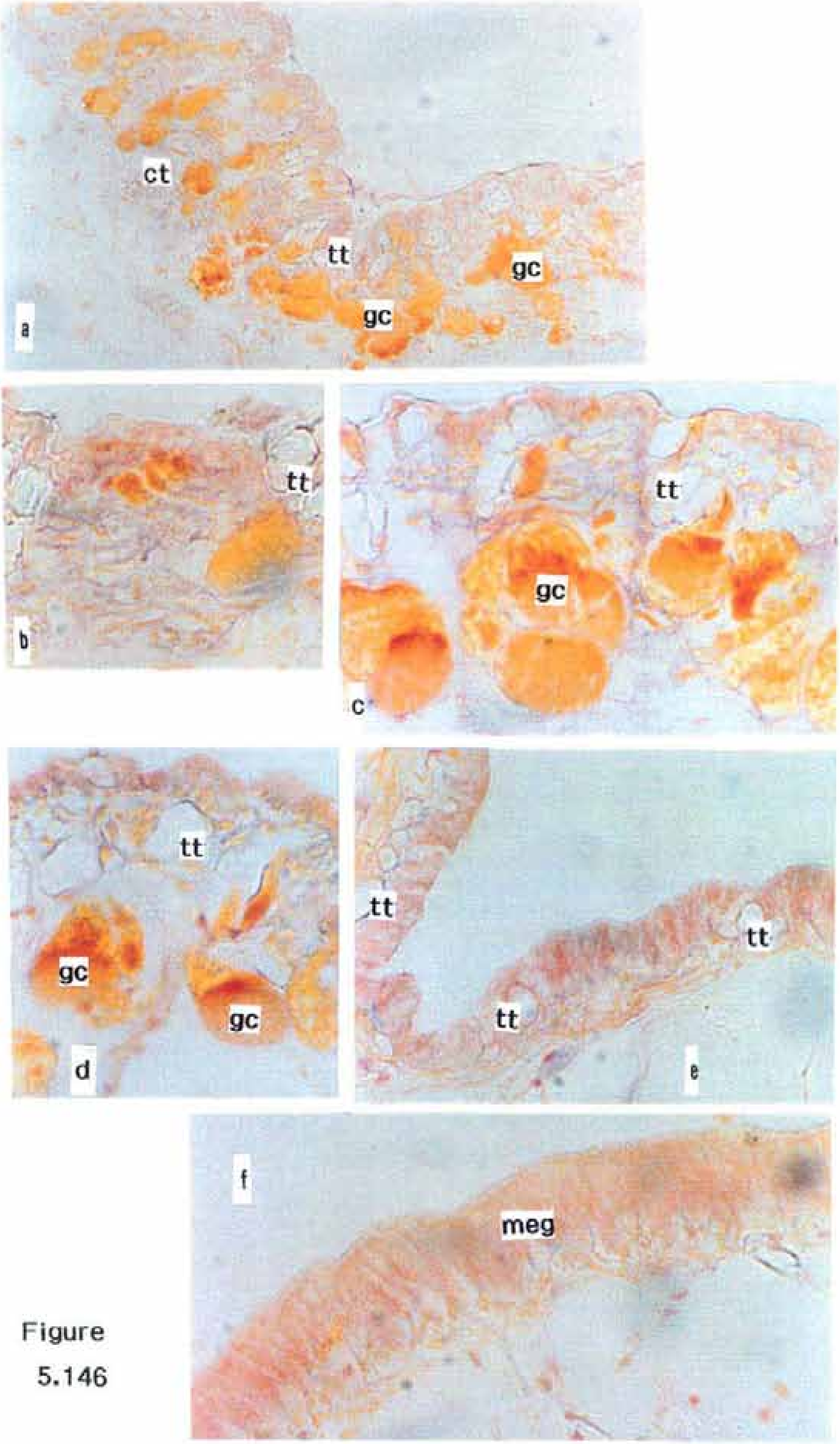


Figure
5.146

5.5.16. *PTERIA PENGUIN*

5.5.16.5. Mantle Margin Folds F1., Ancillary F1. and LatF2..

In Figure 5.147, L is lateral, M, medial, Fold F1. is F1, Fold Ancillary F1. is AF1, and Fold F2. is F2. The parenchyma of Terminal F1. contains Ovoid Blue Glands (ob in Figure 5.147.), and a variety of Granular Cytoplasm Secretory Glands (gc in Figure 5.147.). Tubular (t) and saccular (s) Granular Cytoplasm Secretory Glands continue in the subepithelium of Distal MedF1. (Figure 5.147.).

Large Saccular Granular Cytoplasm Secretory Glands which stain brownish purple and amber with Mallorys (mgc in Figure 5.147.) occur beneath the Middle MedF1. surface epithelium and fibrous connective tissue which in radial section shows transverse muscle fibres in fibrous tissue lacunae (f in Figure 5.147.).

Different species of Granular Cytoplasm Secretory Glands lie beneath the lateral surface of Groove F1. Ancillary F1.(agc in Figure 5.147). The surface epithelium of the Ancillary F1. fold is a columnar epithelium which increases in height going proximally (ce in Figure 5.147.).

Apical Groove F1F2. has an Apical Channel (ac in Figure 5.147.) from which issues the Pleated Secretion of Groove F1F2. (ps in Figure 5.147). The lateral side of the Apical Channel is formed by the cells of the Omega Gland and the medial side by Dactylocytes (o and d respectively in Figure 5.147.).

Proximal LatF2. is an elongate columnar epithelium with numerous apical cilia (pL2 in Figure 5.147.). It produces a secretion which contributes to the Vesiculate Secretion of Groove F1F2..

The subepithelium of Middle LatF2. (se mF2 in Figure 5.147.) is densely packed with Secretory Glands which secrete into Receptacle Glands in the surface epithelium.

FIGURE 5.147. *Pteria penguin* - L.M. Mantle Margin. MedF1., Ancillary F1., Apical Groove F1F2. and LatF2..

L.M. x 190 Mallory's.

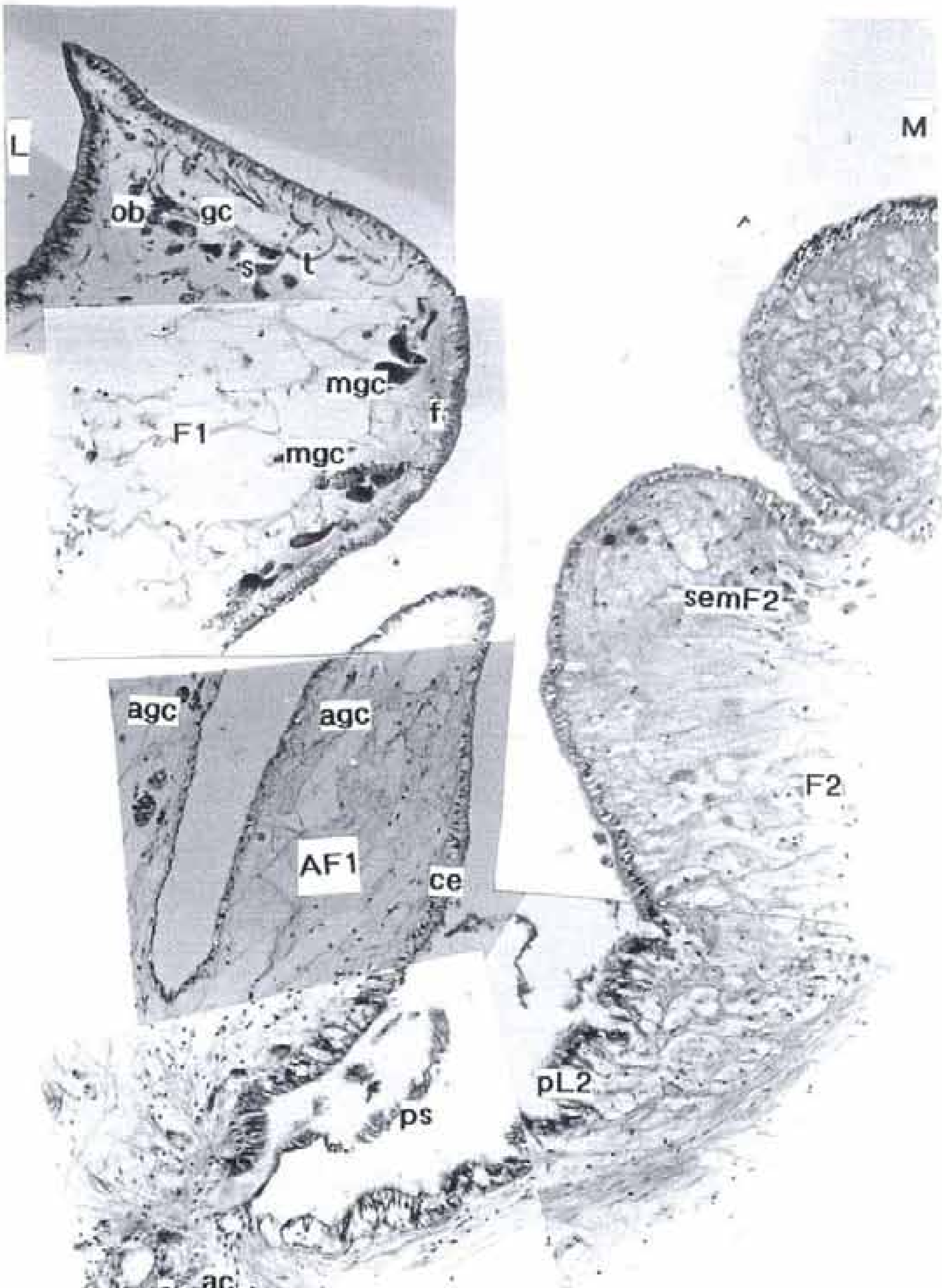


Figure 5.147

5.5.17. *PTERIA AVICULA.*

5.5.17.1. External Mantle. Pallial Nacreous Organic Matrices Formation. Mantle Margin.

The Trabecular Turquoise Glands of the Pallial Region (tt in Figure 5.148. a), are major contributors to the forming Inner Nacreous Layer Organic Matrices (inom in Figure 5.148. a). The Granular Cytoplasm Secretory Glands, many of which in this species stain yellow with A.B./M.S.B. also contribute their secretions to the forming Inner Nacreous Layer Organic Matrices (nom in Figure 5.148. b).

This species has an Omega Gland and Dactylocytes (o and d in Figure 5.148. c) bounding the lateral and medial sides of the Apical Channel (ac in Figure 5.148. c) at the proximal end of the Groove two Mantle Marginal Folds medial to the Mantle Edge Gland of LatF1.. These are therefore designated FoldF1. and Ancillary FoldF1. (F1. and AF1. in Figure 5.148. c). The Pleated Secretion of Groove F1F2. (PS in Figure 5.148. c) emanates from the Apical Channel. The Circumpallial Nerve lies just beneath the Apical fibrous Connective Tissue (cn and act respectively in Figure 5.148. c). The F1. and F2. Rami of the Circumpallial Nerve, (F1.n. and F2.n. respectively in Figure 5.148. c). innervate the secretory structures of MedF1. and LatF2. respectively. The secretory structures of Groove F1. Ancillary F1. produce a material with the same structure and staining affinities as the forming Nacreous Organic Matrices of the Pallial Region. (onom in Figure 5.148. c and d). (compare Figure 5.148. b, with Figure 5.149. a and b).

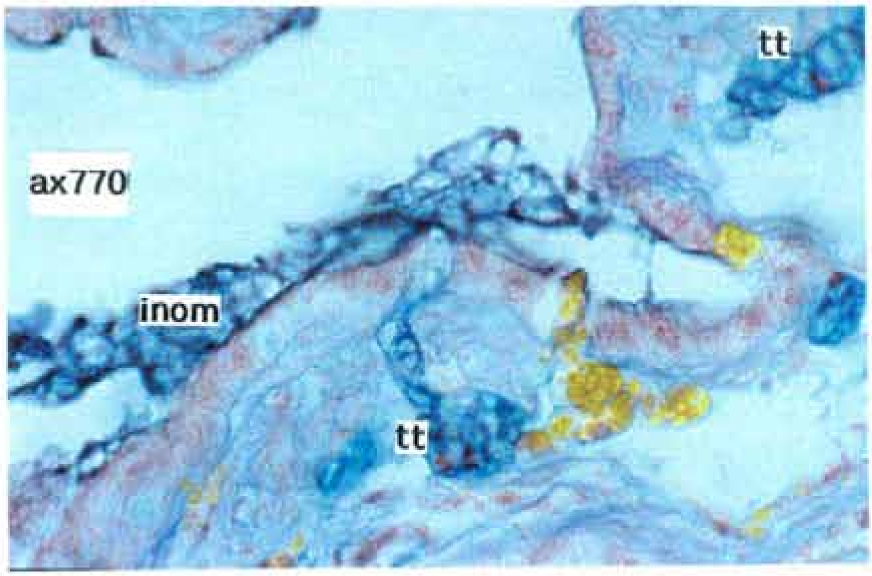
FIGURE 5.148. *Pteria avicula* Ext. Mantle, Mantle Margin and secretions.

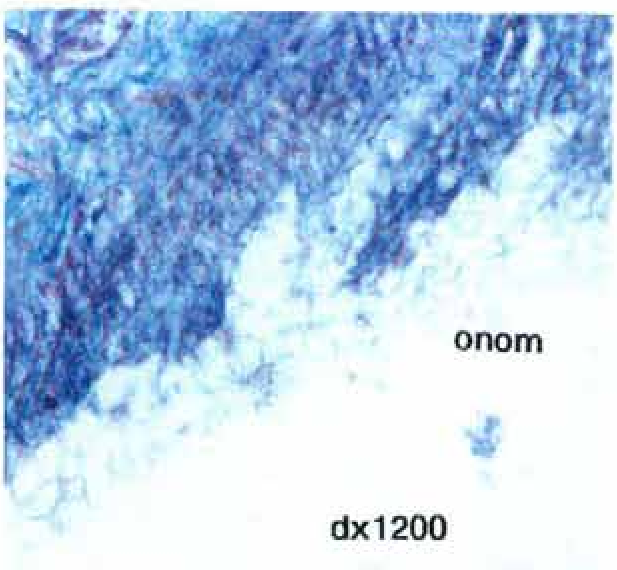
a. L.M. x 770 A.B./M.S.B. External Mantle, secretion of unicellular Glands.

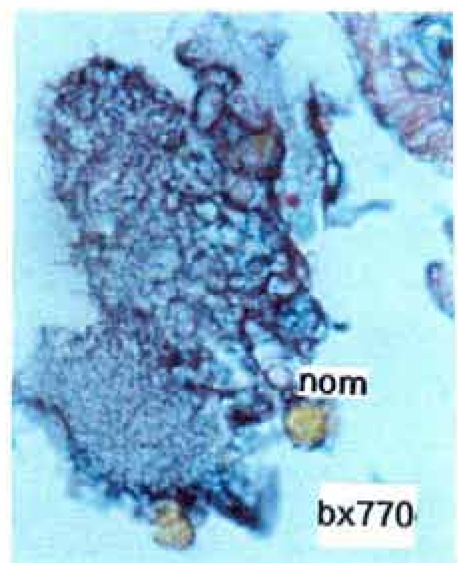
b. L.M. x 770 A.B./M.S.B. Pallial Region, Formation of Innwer Nacreous Layer Organic Matrices from unicellular gland secretions.

c. L.M. x 230 Steedman's. MedF1., Ancillary F1., Apical Groove F1F2. and LatF2..

d. L.M. x 1200 Steedman's. Groove F1.AncillaryF1..







d

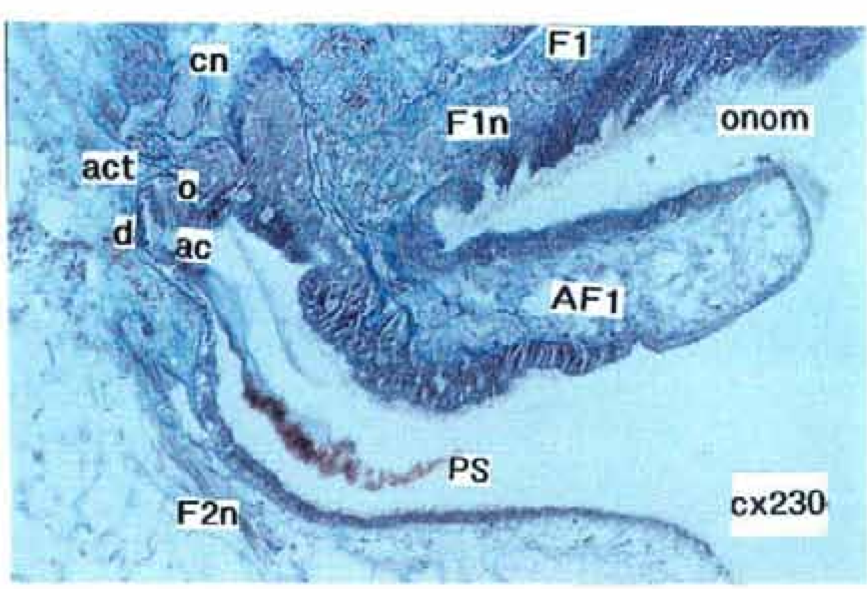


Figure
5.148

5.5.17. *PTERIA AVICULA.*

5.5.17.2. External Mantle - Secretion of Pallial Nacreous Organic Matrices.

Mantle Margin - Secretion of Prismatic Layer.

With Azan, the joined secretions of the Trabecular Turquoise Glands (which are refractory to this stain) and the Granular Cytoplasm Secretory Glands (most of which stain from Magenta to various shades of red and purple with this stain), (tt and gc respectively in Figure 5.149. a and b), stain the distinctive blue purple colour of Nacreous Organic Matrix. (nom in Figure 5.149. a and b).

Between the Pleated Secretion of Groove F1F2. (PS in Figure 5.149. c) and the Proximal LatF2. (PLF2. in Figure 5.149. c) the material secreted by this region can be seen forming into an Organic Matrix (PLOM in Figure 5.149. d) indistinguishable in structure and staining affinities from the material forming the proximal end of a Growth Scale between laminae of Nacreous Layer in *Isognomon ephippium* decalcified shell (Figure 5.152. b and c).

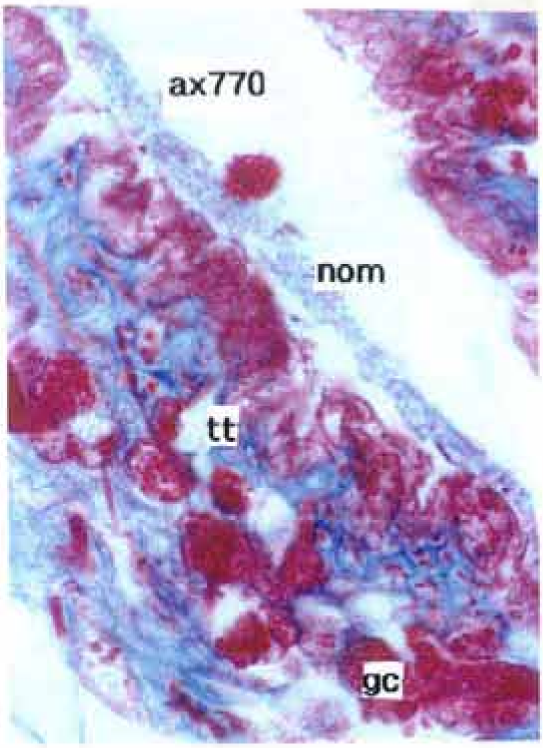
FIGURE 5.149. *Pteria avicula* - External Mantle - Secretion of Nacreous Organic Matrices; Mantle Margin - Secretion of Prismatic Layer Organic Matrices.

a. L.M. x 770 Azan. Pallial Region - Formation of Nacreous Organic Matrices.

b. L.M. x 1200 Azan As for a above at higher magnification.

c. L.M. x 120 Mallory's Groove F1F2. Pleated Secretion and Vesiculate Secretion.

d. L.M. x 1200 Mallory's. Organization of Vesiculate Secretion into Prismatic Layer Organic Matrices.



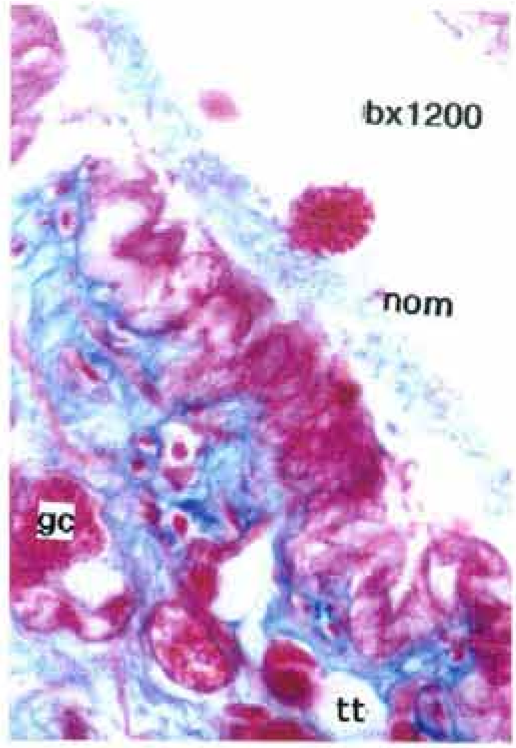
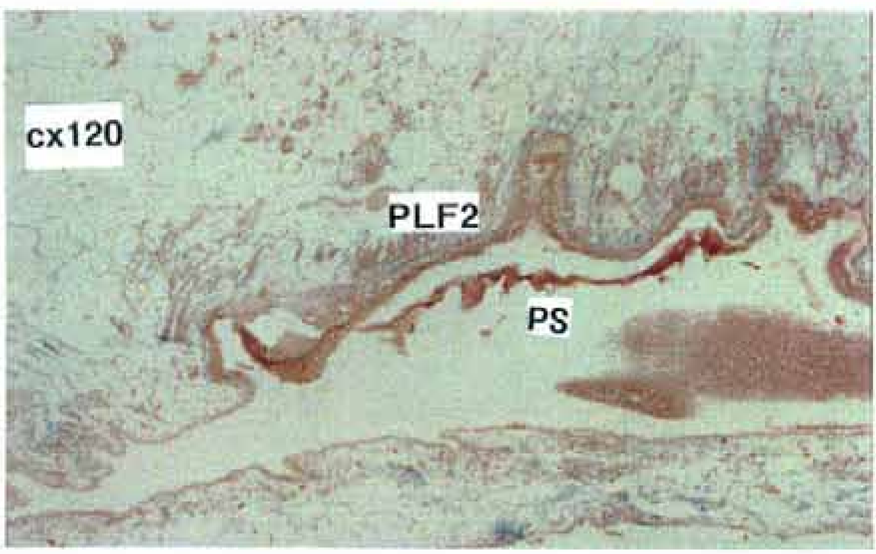
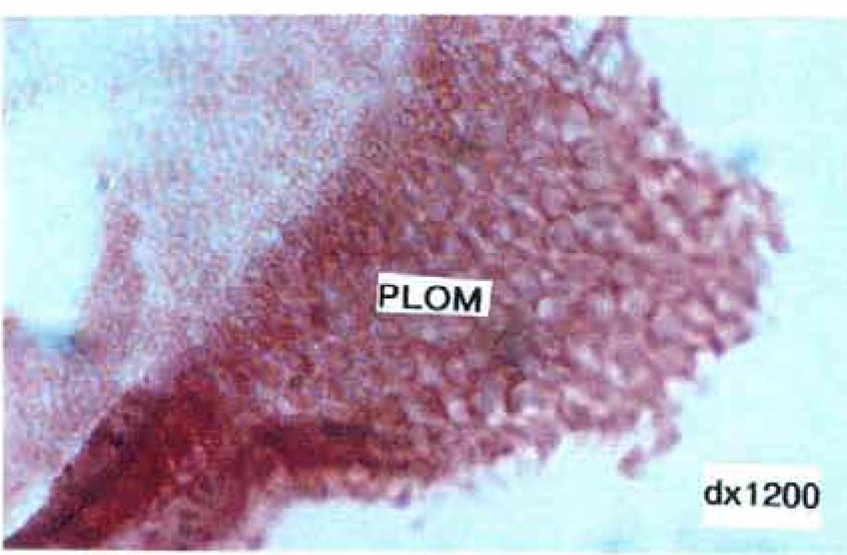


Figure
5.149





5.5.18. *ISOGNOMON ISOGNOMON.*

5.5.18.1. External Mantle - Outer Nacreous layer formation.
Mantle Margin - Inner Nacreous Layer Formation.

In Figure 5.150, L is lateral and M is medial.

With A.B./M.S.B. the turquoise coloured secretions of the Trabecular Turquoise Glands and the red-purple secretions of the Granular Cytoplasm Secretory Glands (tt and gc respectively in Figure 5.150. b, c and d) of both the External Mantle (Figure 5.150. b and d) and the F1. and Ancillary F1. of the Mantle Margin (Figure 5.150. b and c) produce the blue-turquoise forming Inner and Outer Nacreous Organic Matrices respectively (inom and onom respectively in Figure 5.150. b, c and d). Primitive eyes in this species occur on lateral terminal Ancillary F1. (l in Figure 5.150. a). The Pleated Secretion of Groove F1F2. divides the secreted material of F1. and Ancillary F1. lateral to it (F1. and AF1. in Figure 5.150. a and b) from that of F2. and LatF3. medial to it (F2. and LF3. in Figure 5.150. a and b).

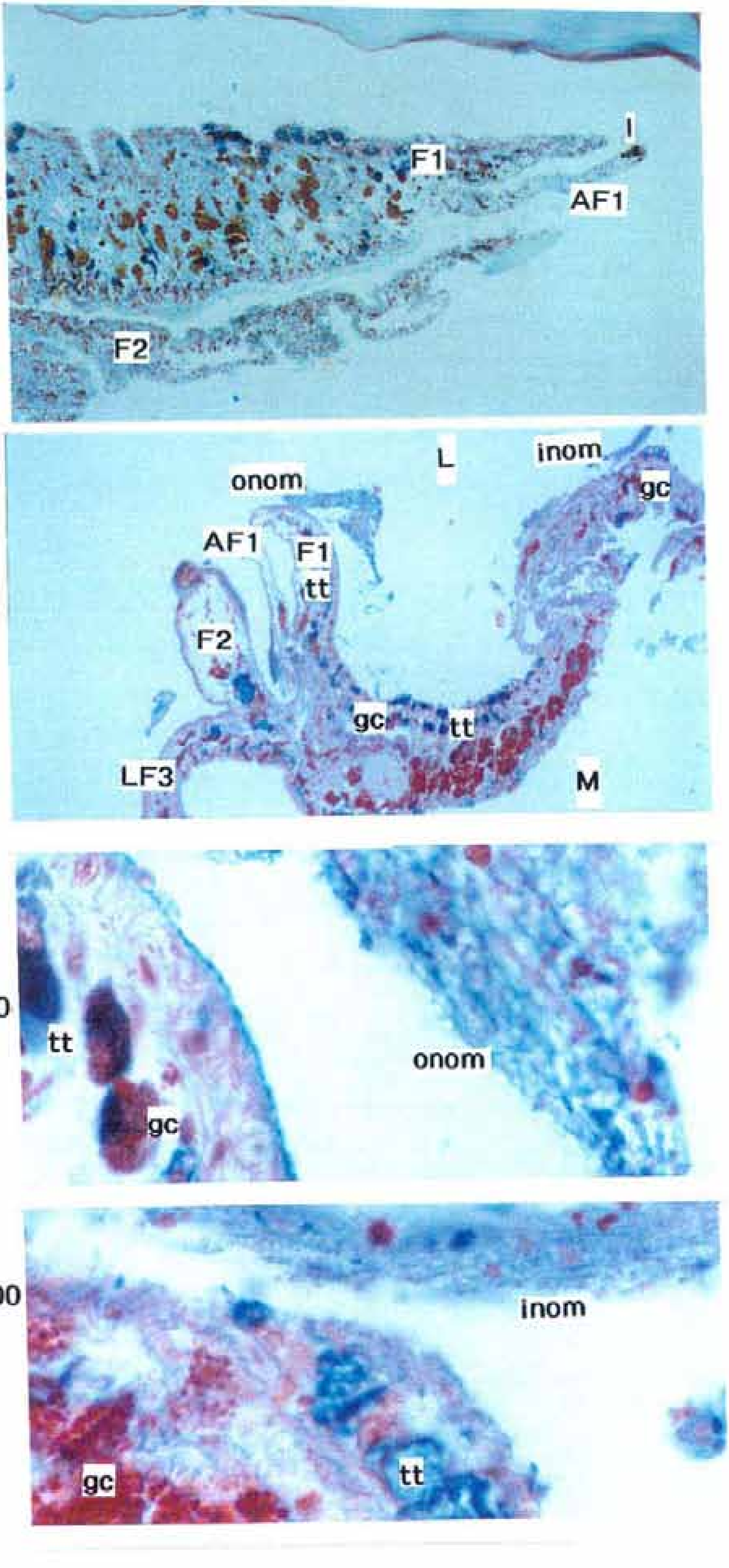
FIGURE 5.150. *Isognomon isognomon* - L.M. Nacreous Layer Organic Matrices
secretion by External Mantle and Mantle Margin F1..

a. L.M. x 150. A.B./M.S.B. Folds F1., Ancillary F1., F2. and F3..

b. L.M. x 77. A.B./M.S.B.

c. L.M. x 1200 A.B./M.S.B. LatF1. and Formation of Outer Nacreous Layer Organic Matrices.

d. L.M. x 1200 A.B./M.S.B. Proximal Pallial Mantle - Formation of Inner Nacreous Layer Organic Matrices.



ax150

bx77

cx1200

dx1200

Figure 5.150

5.5.19. *ISOGNOMON EPHIPPIUM.*

5.5.19.1. Inner and Outer Nacreous Layers - Medial Surfaces.

The Medial Surface of the Proximal Ventral Pallial Nacre displays a pattern of edges of Nacre Sheets of numerous anastomoses and short discontinuous edges with tight bends (Figure 5.151. a). The partly-bound and free Nacre Tiles extend over about half the distance between successive Nacre Sheets and tend to regular hexagons (Figure 5.151. b).

The Edges of the Nacre Sheets of the Inner Nacreous Layer at the Ventral Distal Pallial Region have a pattern tending to straight parallel lines with few anastomoses, (Figure 5.151. c). Here also the Inner Nacreous Layer Partly-bound and Free Nacre Tiles tend to regular Hexagons and extend for about half the distance between successive Nacre Sheets (Figure 5.151. d).

The Partly-bound and Free Nacre Tiles of the Outer Nacreous Layer near the Nacreo-prismatic Junction commonly have a truncated diamond shape with the long axis of the Nacre Tiles normal to both the edge of the adjacent Nacre Sheet and the Nacreo-prismatic Junction (pb in Figure 5.151. e and f).

FIGURE 5.151. *Isognomon ephippium* - Inner and Outer Nacreopus Layers.

a. S.E.M. x 98 Inner Nacreous Layer - Proximal Pallial Nacre.
b. S.E.M. x 3100 As for a above at higher magnification.
c. S.E.M. x 98 Inner Nacreous Layer Distal Pallial Nacre.
d. S.E.M. x 1700 As for c above at higher magnification.
e. S.E.M. x 820 Outer Nacreous Layer just proximal to Nacreo-prismatic Junction.
f. S.E.M. x 3300 As for e above at higher magnification.

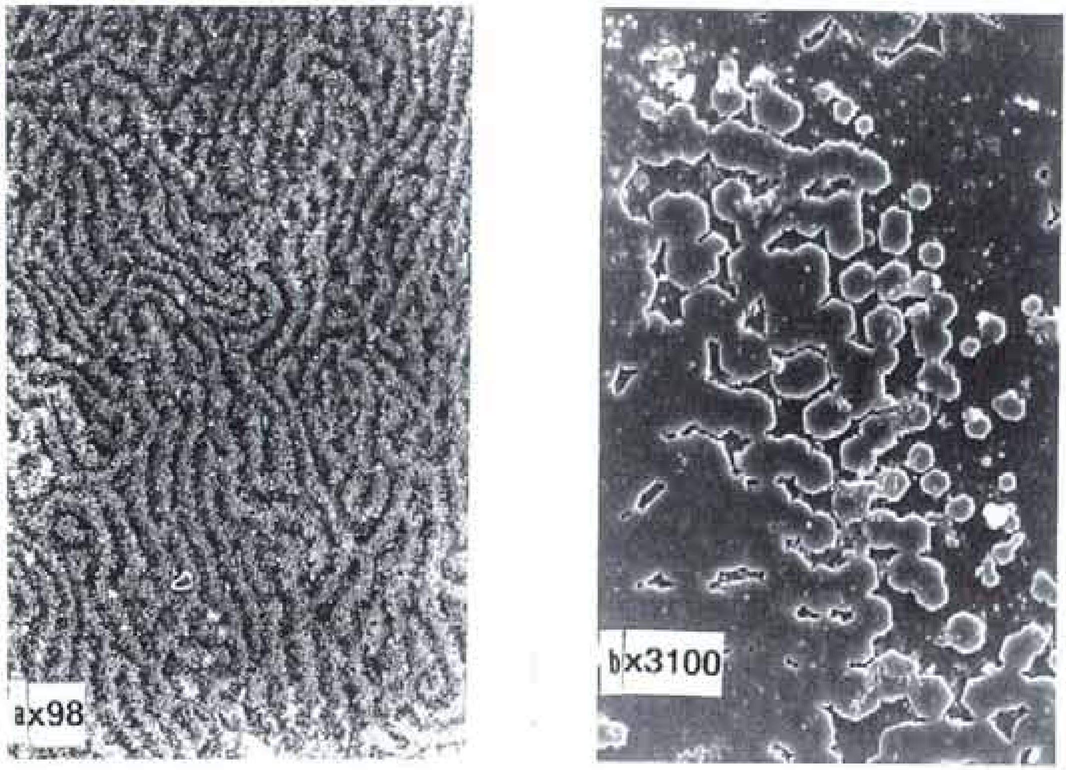
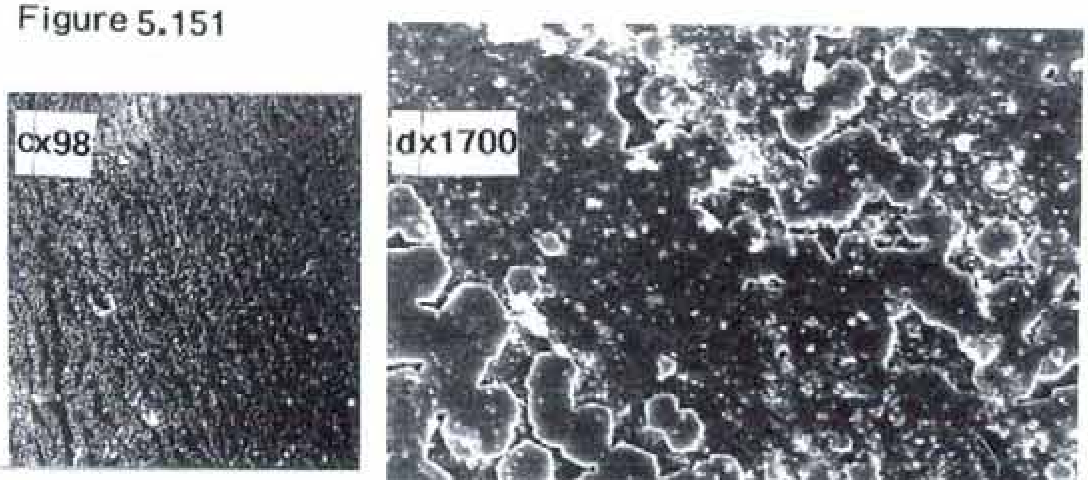
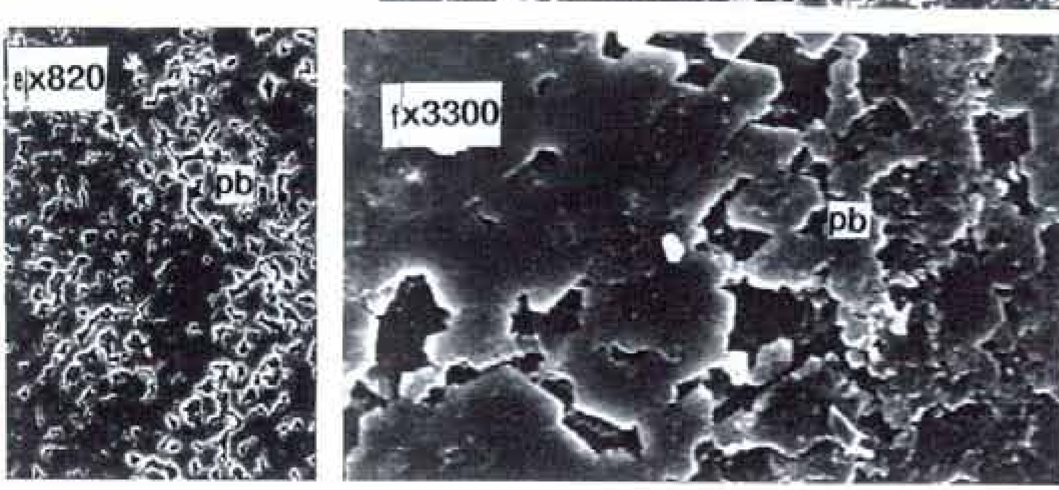


Figure 5.151





5.5.19. *ISOGNOMON EPHIPPIUM.*

5.5.19.2. Decalcified Shell Layers.

The Organic Matrices residual in decalcified Nacreous Layers stain blue with a brown-purple tinge with Mallorys (nom in Figure 5.152. a, b and c).

The proximal ends of the Prismatic Layer Growth Scales lie between laminae of Nacreous Layer (pgs in Figure 5.152 a, b and c). The microstructure and staining affinities (pgs in Figure 5.152. c) closely approximate the maturing secretions of Proximal LatF2. seen in *Pteria avicula* (Figure 5.149. c and d).

The Prismatic Layer Organic Matrices residual following acid decalcification show Major Prisms with Transverse Parallel Side Walls (mp and tp respectively in Figure 5.152. d). The Major Prisms have Outer Prismatic End Plates, Inner Lateral Structure and Inner Prismatic End Plates (opep, ILS and ipep in Figure 5.152. d).

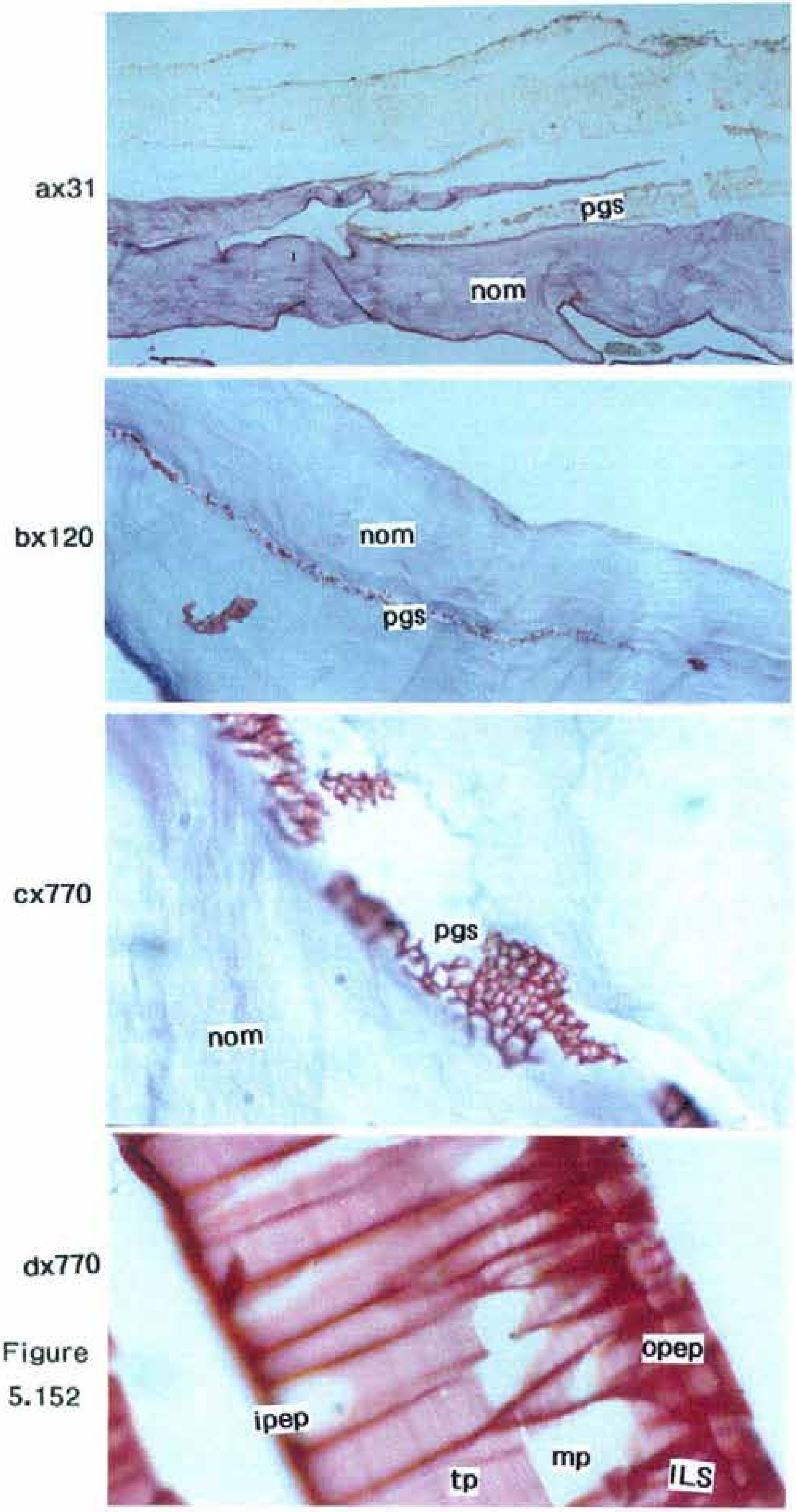
FIGURE 5.152. *Isognomon ephippium* - L.M. Decalcified shell Prismatic Layer and Nacreous Layers - Radial Section.

a. L.M. x 31. Mallory's. Nacre and Prismatic Layer.

b. L.M. x 120. Mallory's. Nacre and proximal part of Prismatic Layer Growth Scale.

c. L.M. x 770. Mallory's. As for b above at higher magnification.

d. L.M. x 770. Mallory's. Prismatic Layer.


ax31
pgs
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bx120
nom
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cx770
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nom
dx770
opep
ipep
mp
tp
ILS

Figure 5.152

5.5.19. *ISOGNOMON EPHIPPIUM.*

5.5.19.3. External mantle and Mantle Margin.

With A.B./M.S.B. the Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands of the External Mantle (tt and gc respectively in Figure 5.153. a), produce a turquoise staining Nacreous Organic Matrix (nom in Figure 5.153. a).

The Mantle Edge Gland (meg in Figure 5.153. b) somewhat unusual in that there are subepithelial Trabecular Turquoise Glands which secrete through it (tt in Figure 5.153. b) and the elongate columnar epithelial cells of the Gland bear basal pigment granules. (pg in Figure 5.153. b and c).

The Mantle Margin has F1. and Ancillary F1. Folds (F1. and AF1 in Figure 5.153. c and d) lateral to the Groove which bears the Pleated Secretion of Groove F1F2., (PS in Figure 5.153. c and d). Medial to Groove F1F2. are Folds F2. and F3. (F2 and F3 respectively in Figure 5.153. c and d).

The subepithelia beneath both the MedF1. and Lateral Ancillary F1. (MF1 and AF1. in Figure 5.153. c and d), bears a heavy concentration of both the External Mantle - F1. type of Trabecular Turquoise Gland and Granular Cytoplasm Secretory Glands (tt1 and gc in Figure 5.153. c and d).

The Pleated Secretion of Groove F1F2. rises in the Apical Channel between the Omega Gland laterally (o in Figure 5.153. c and d) and Dactylocytes medially.

The subepithelial Glands of Proximal LatF2. (pLF2. in Figure 5.153. c) discharge beneath the Pleated Secretion of Groove F1F2. via Receptacle Glands.

There is a dense population of the F2. F3. type of Trabecular Turquoise Gland just beneath the surface epithelium of MedF2. and LatF3. (tt2 in Figure 5.153. c). The surface epithelium of MedF2. and LatF3. is a columnar epithelium the cytoplasm of whose cells stain dark amber brown with Mallorys (MF2, LF3. respectively in Figure 5.153 d).

FIGURE 5.153. *Isognompon ephippium* – L.M. External Mantle and Mantle Margin.

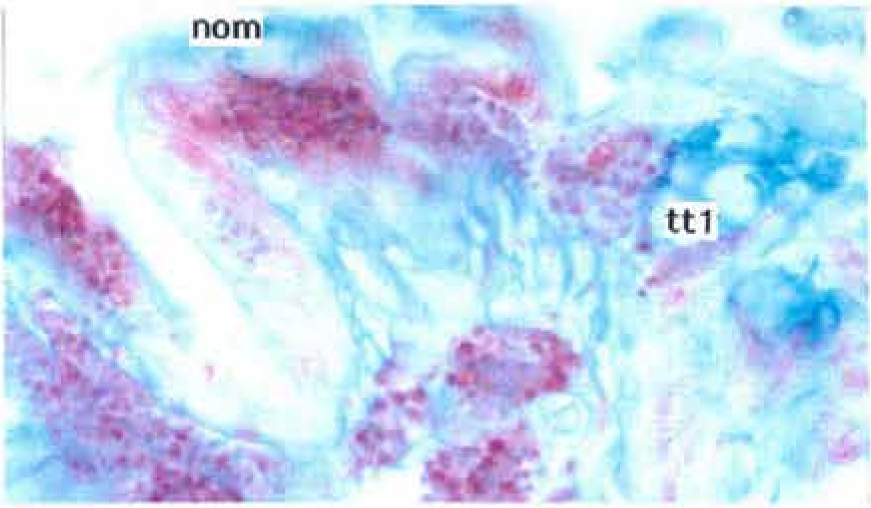
a. L.M. x 1200. A.B./M.S.B. Pallial Region.

b. L.M. x 1200. A.B./M.S.B. Mantle Edge Gland.

c. L.M. x 120 A.B./M.S.B. Mantle Margin.

d. L.M. x 77. Mallory's. Mantle Margin.

ax1200



bx1200

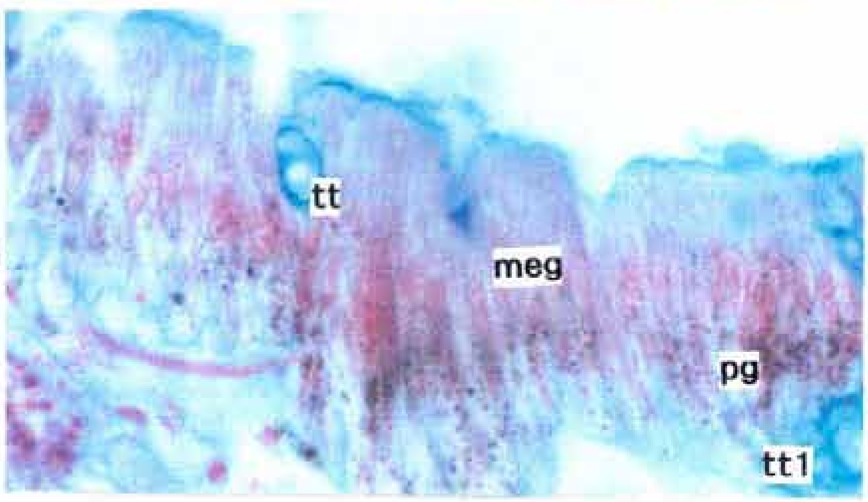
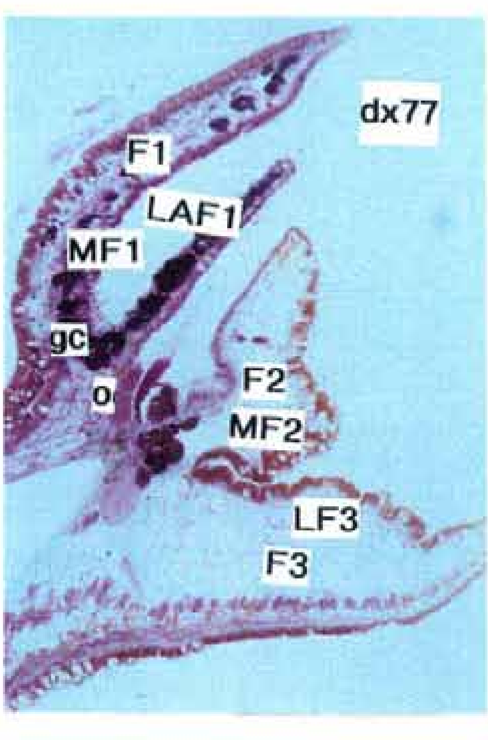
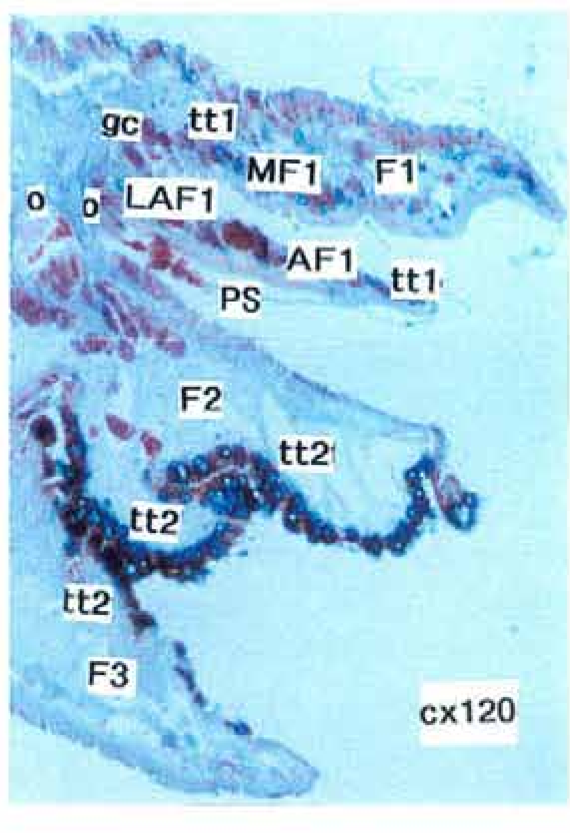


Figure 5.153





5.5.20. *MALLEUS ALBA.*

5.5.20.1. External mantle and Mantle Margin.

With A.B./M.S.B. the fused secretions of the Trabecular Turquoise Glands and the Granular Cytoplasm Secretory Glands of the External Mantle (tt and gc respectively in Figure 5.154. a) form a blue-turquoise staining Inner Nacreous Layer Organic Matrix zz(inom in Figure 5.154. a).

This species has an unusually large number of conspicuous Ovoid Blue Glands in Terminal F1. (ob and tF1 respectively in Figure 5.154. b). With A.B./M.S.B. the Distal Diffuse Glands in this species stain bright amber (dd in Figure 5.154. b).

The Pleated Secretion of Groove F1F2. (PS in Figure 5.154. b) issues from the Apical Channel.

There are type F2. F3. Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands which latter stain red and amber with A.B./M.S.B. in the subepithelium of F2. and LatF3. (tt2 and gc respectively in Figure 5.154. b).

FIGURE 5.154. *Malleus alba* - L.M. External Mantle and Mantle Margin.

a. L.M. x 230. A.B./M.S.B. External Mantle.

b. L.M. x 770. A.B./M.S.B. Mantle Margin.

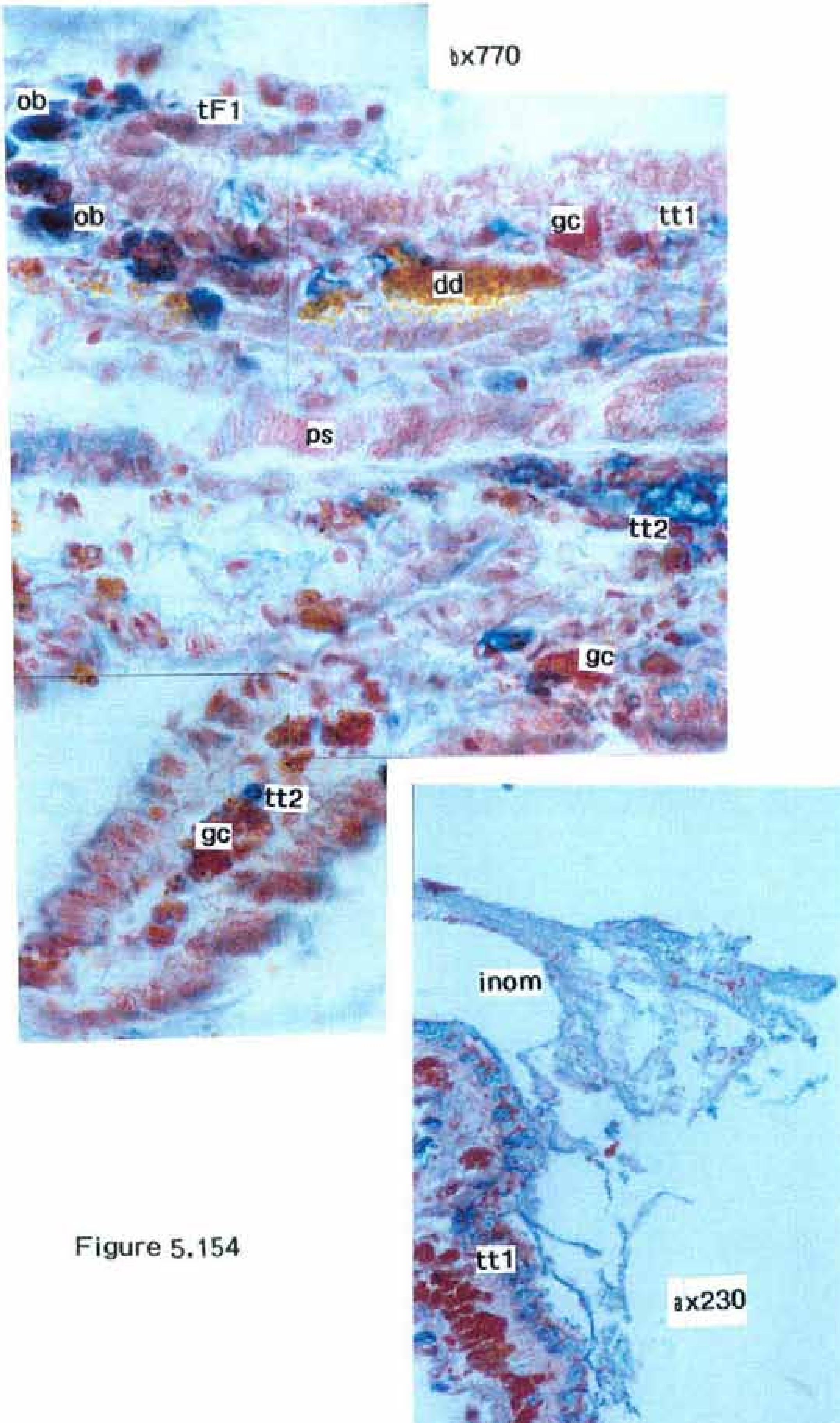


Figure 5.154

5.6. OSTREOIDEA.

5.6.1. *HYOTISSA HYOTIS.*

5.6.1.1. Valves.

The valves of this oyster are composed of tubular hollow prisms whose long axes are about normal to the lateral and medial surface of the valve, closed at each end by a sheet- like Shell Layer.

The walls of the prisms have transverse parallel structures.

Thus there are at least two Shell Layers which alternate throughout the thickness of the valve.

Since the lateral surface of the valve is devoid of uncovered ends of prisms, growth must proceed by the laying down of a sheet like Shell Layer then the construction on this of the Prismatic Shell Layer. Except for (probably aberrant) small areas near the periphery, the Prismatic Layer is then covered medially by a further sheet like Shell Layer. It is not known whether the sheet like Shell Layers are a single lamina or are bilaminate and to pursue this point is outside the scope of this investigation.

The relationship between complexity of shell structure and degree of Tissue differentiation of the Mantle Margin is illustrated by the species of the Ostreoidea. The degree of tissue differentiation medial to the Pleated Secretion of Groove F1F2. in *Hyotissa hyotis*, *Saccostra Echinata* and *Saccostrea cucullata* are to be compared with that in the *Pinctada* and contrasted against that in *Ostrea sp* etc.

FIGURE 5.155 *Hyotissa hyotis* Valves.

a. x 0.85 Medial surface - Left valve.

b. x 0.85 Lateral surface - Right valve.

a

b

Figure 5.155

5.6.1.2. The Ventral Mantle Margin.

There are three Marginal Mantle Folds, F1., F2. and F3. going latero-medially (F1, F2 and F3 in Figure 5.156. a.).

The Lat F1. is dominated by an extensive Mantle Edge Gland (meg in Figure 5.156 a and b).

MedF1. has three regions, Distal MedF1., Middle MedF1. and Proximal MedF1..

The subepithelium of Proximal MedF1. hosts a large secretory structure (P in Figure 5.156. a and b).

There is a well developed Omega Gland (o in Figure 5.156. b, and Figure 5.157.), which occupies the lateral aspect of the apex of Groove F1F2.. The Pleated Secretion of Groove F1F2., (PS in Figure 5.156. b) issues from the Apical Channel between the Omega Gland and a specialised epithelium lining the opposing Proximal LatF2..

A large Circum-pallial Nerve lies immediately beneath Apical Groove F1F2. (n in Figure 5.156 a).

The large F2. Ramis of the Circum-pallial Nerve innervates the secretory structures of Proximal latF2. (f2n in Figure 5.156. b).

The MedF2. is divided into histologically distinct Distal and Proximal Regions. The Proximal MedF2. subepithelium is occupied by an extensive and distinctive secretory gland (PM2 in Figure 5.156. a and Figure 5.157.).

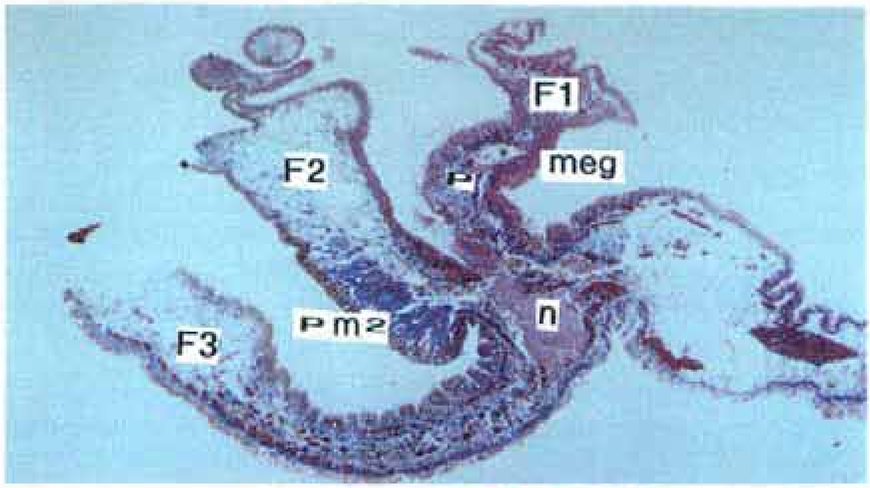
LatF3. is similarly divided histologically into Proximal LatF3. and Distal LatF3..

FIGURE 5.156. *Hyotissa Hyotis* - Mantle Margin.

a. L.M. x 77 Azan Mantle Margin.

b. L.M. x 770 Azan Proximal MedF1. and Apical Groove F1f2..

a
x 77



b
x 770

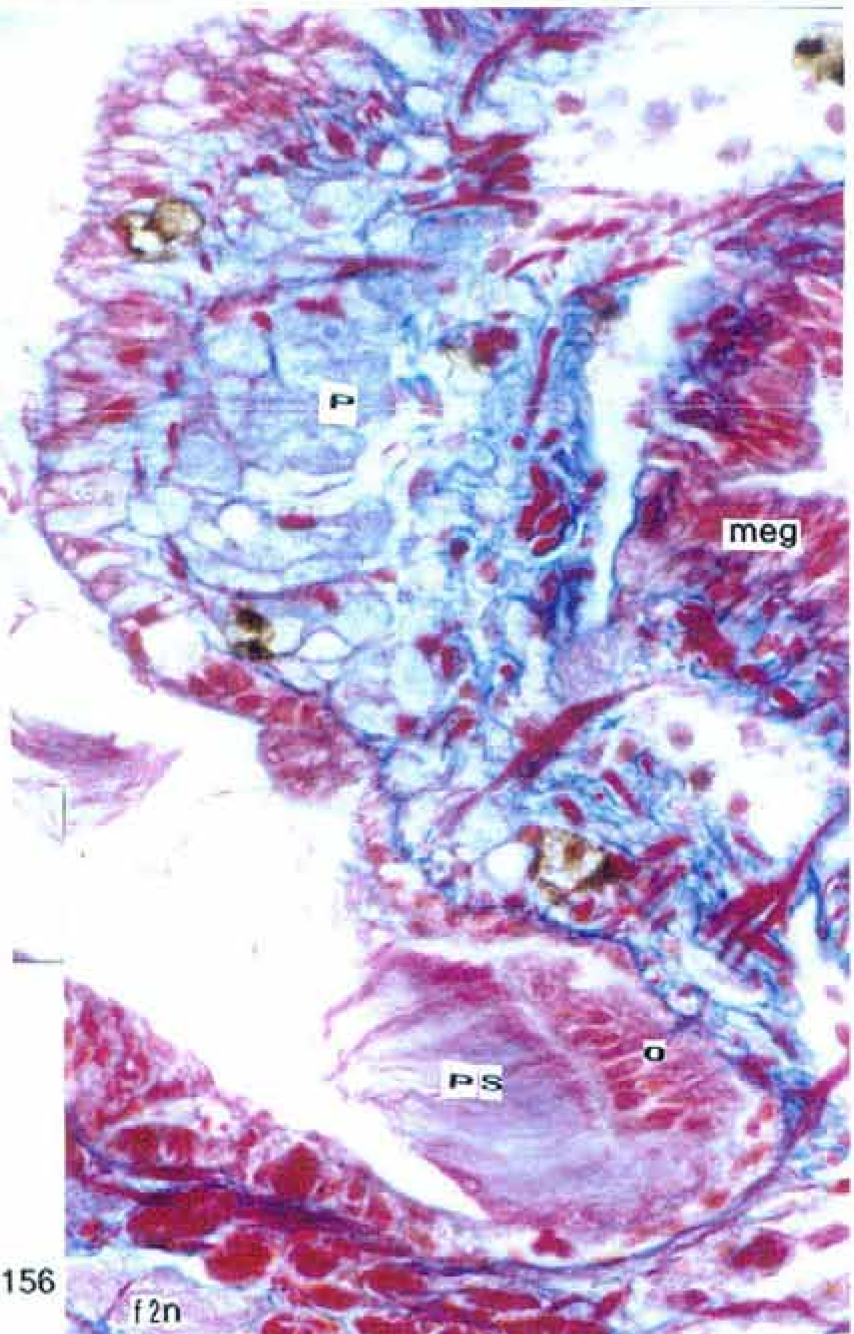


Figure5.156

5.6.1.3. Apical Groove F1F2. and Subepithelial Secretory Gland of Proximal MedF2..

The unicellular secretory glands of the glandular structure in the Proximal medF2. (pm2 in Figure 5.157.) subepithelium are extremely elongate being up to 10 µm in length.

FIGURE 5. 157 *Hyotissa Hyotis* - Apical GrooveF1F2. and Proximal MedF2..
L.M. x 770 Azan.

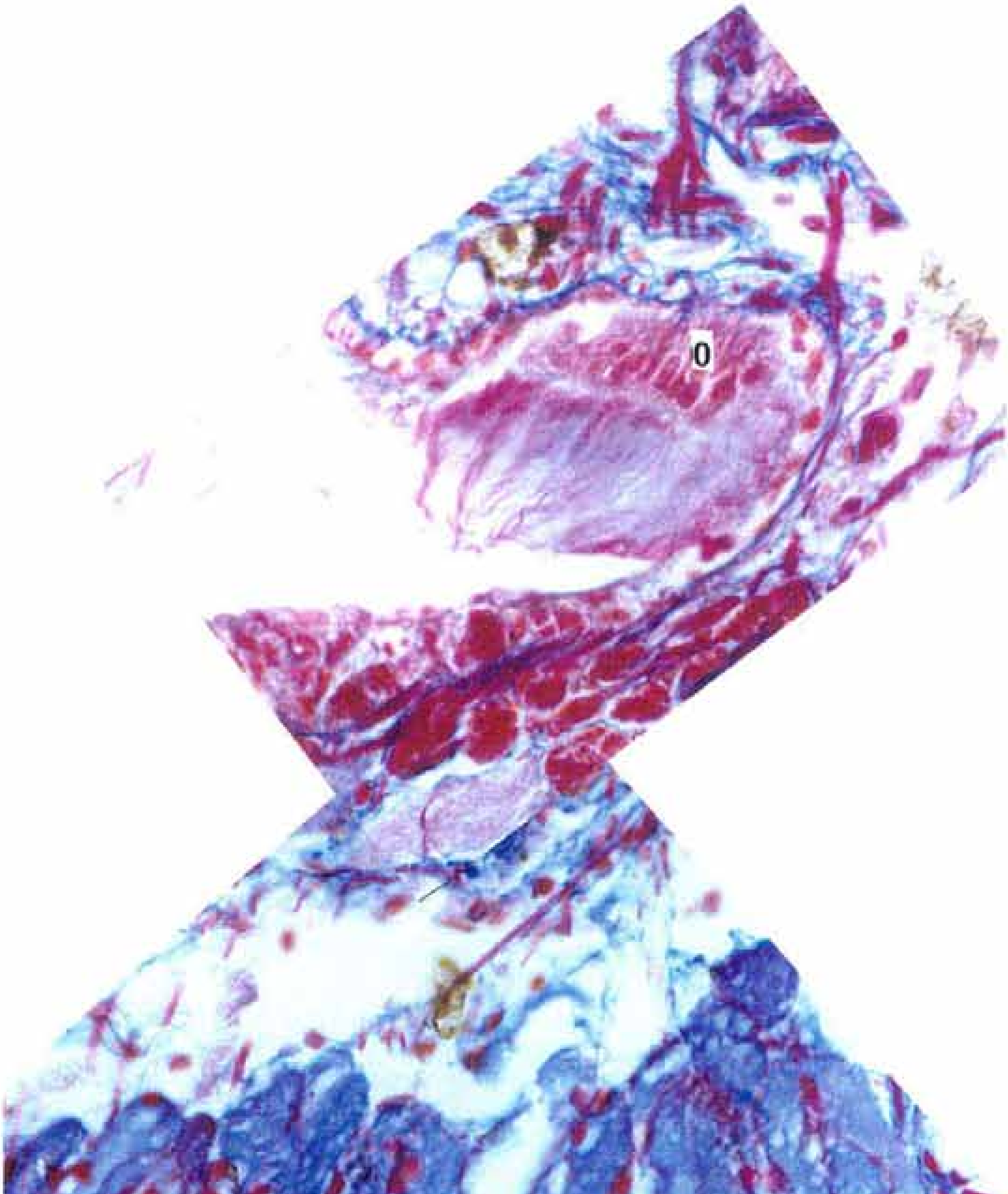


Figure5.157
x 770

5.6.2. *OSTREA SP.*

5.6.2.1. Valves and Mantle Margin.

The Upper (right) Valve of *Ostrea sp.* has two Shell Layers - an outer Prismatic Layer and an inner crossed lamellar Shell Layer.

The lower (left) Valve of this species has a structure of crossed lamellar structure alternating with a different Shell Layer which is different from the outer Prismatic Layer of the upper Valve (Figure 5.158. a).

The Mantle Margin has three Marginal Mantle Folds, F1., F2. and F3.. There is an Omega Gland (o in Figure 5.158. b) on the lateral side of Apical Groove F1F2. medial to which issues a Pleated Secretion (PS in Figure 5.158. b). Proximal to Apical Groove F1F2. are a Circum-pallial Nerve and a Curcum-pallial sinus (n and s respectively in Figure 5.158. b).

FIGURE 5.158. *Ostrea sp.* L.M. Valves and Upper Mantle Margin.

a. x 1

b. L.M. x 77 A.B./M.S.B.

a

x1

b x77

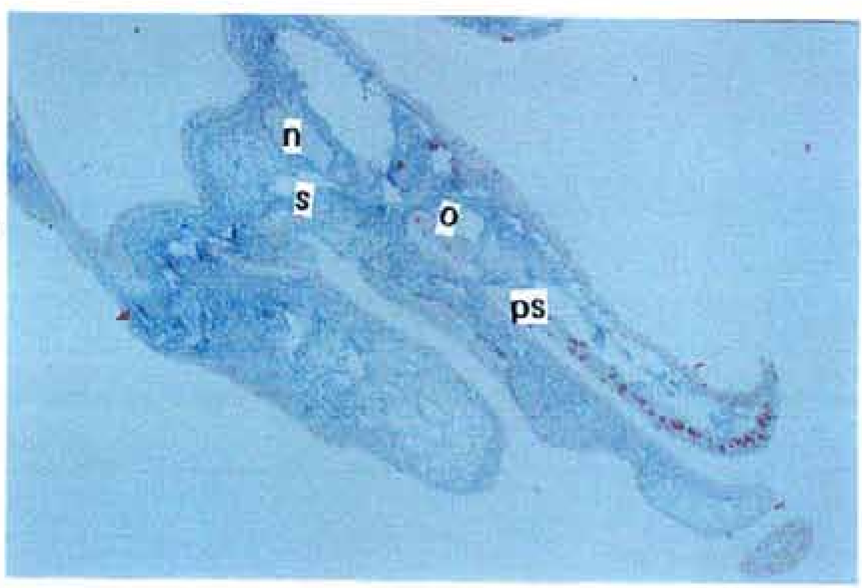


Figure 5.158

5.6.3. *SACCOSTREA ECHINATA.*

5.6.3.1. S.E.M. of Shell Layers.

The upper (right) valve of *Saccostrea echinata* has both a surface Prismatic Layer (pl Figure 5.159. a and b) which may be bilaminate (Figure 5.159. a). The prisms are irregularly polygonal in cross-section (pcs Figure 5.159. b). The greater part of the valve is composed of Crossed Lamellar Structure (cls Figure 5.159. c). However Prismatic Layers may be apparently irregulary embedded in the latter.

The Lower Valve is usually cemented to a surface but again consists of Crossed Lamellar Structure Shell Layers (cls in Figure 5.159. d), interspersed between Prismatic Layers.

FIGURE 5.159. *Saccostrea echinata.* Shell Layers in the Upper and Lower Valves.

a. S.E.M. x 350 Upper Valve.

b. S.E.M. x 700 Upper Valve.

c. S.E.M. x 2800 Upper Valve.

d. S.E.M. x 1400 Lower Valve.

ax350

bx700

cx2800

dx1400

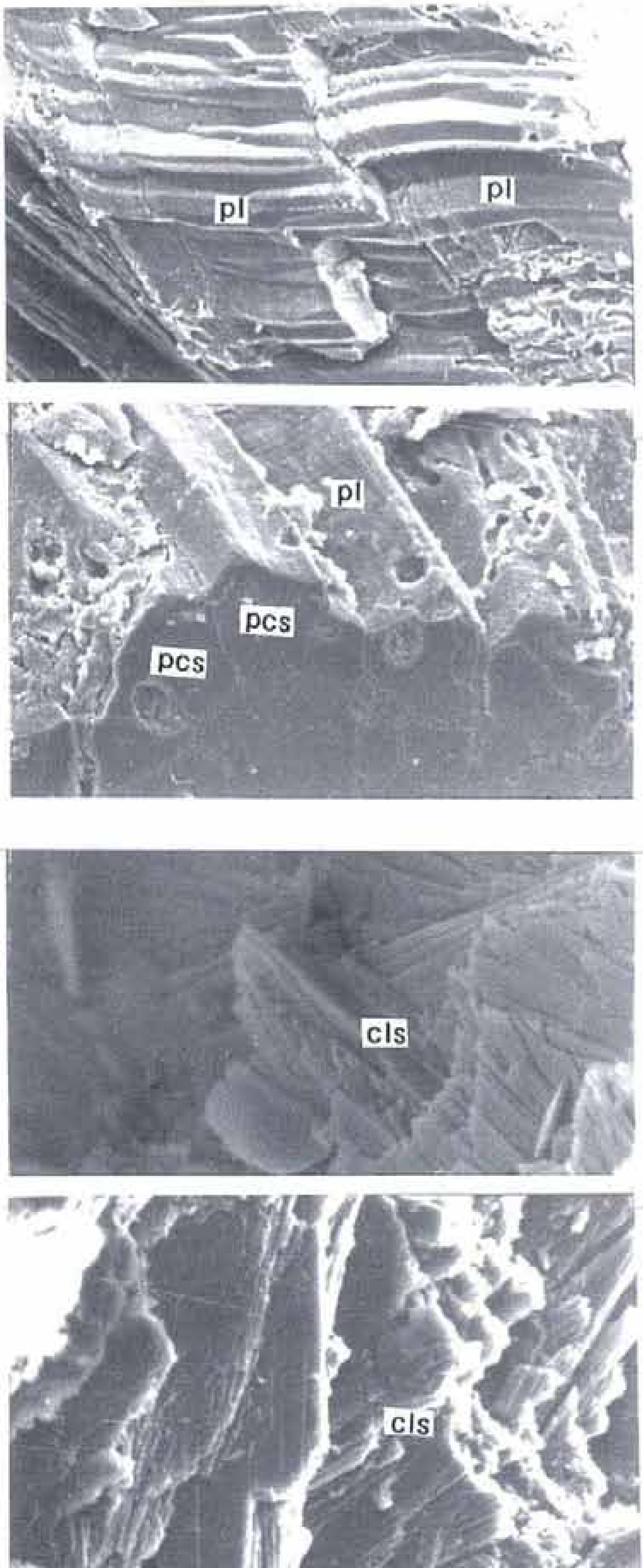


Figure 5.159

5.6.3.2. The Upper External Mantle and Mantle Margin.

The upper External Mantle has a Pallial Gland distal to the Adductor Muscle set in dense fibrous connective tissue (pg in Figure 5.160. a and b). The unicellular glands of the Pallial Gland are Trabecular Turquoise Glands and a variety of Granular Cytoplasm Secretory Glands (g in Figure 5.160. b). The Pallial Mantle, (pm, Fig. 5.160. a and c) similarly has a considerable population of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands which latter are generally lighter staining than those of the Pallial Gland (g in Figure 5.160. c).

The Mantle Margin has three Marginal mantle Folds with a Mantle Edge Gland on LatF1., (meg in Figure 5.160. d), Ovoid Blue Glands at Terminal F1. (obg in Figure 5.160. d), and a dense mass of Granular Cytoplasm Secretory Glands in the subepithelium of MedF1. of at least two different kinds - staining purple and reddish with Azan (pgs and rgs respectively in Figure 5.160 d).

The Apical Groove F1F2. has an Omega Gland on its lateral aspect (o in Figure 5.160. a). From between this and the apical surface of Proximal LatF2. issues the Pleated Secretion of Groove F1F2. (ps in Figure 5.160 a and d). Sub-apically is a Circum-pallial nerve and a Circum-pallial sinus with haemocytes (cpn and cps respectively in Figure 5.160. a).

Middle LatF2. is crenular and ciliated and Distal LatF2. is ciliated (ml2 and dl2 in Figure 5.160. d). MedF2. has a pigmented epithelium and subepithelial. Proximal MedF2. has a population of Turquoise and Granular Cytoplasm Secretory Glands as does Proximal LatF3. (pm2 and pl3 respectively in Figure 5.160 d).

FIGURE 5.160. *Succostrea echinata* - L.M. Upper External Mantle and Mantle Margin.

a. L.M. x 54 Azan.

b. L.M. x 220 Azan.

c. L.M. x 220 Azan.

d. L.M. x 220 Azan.

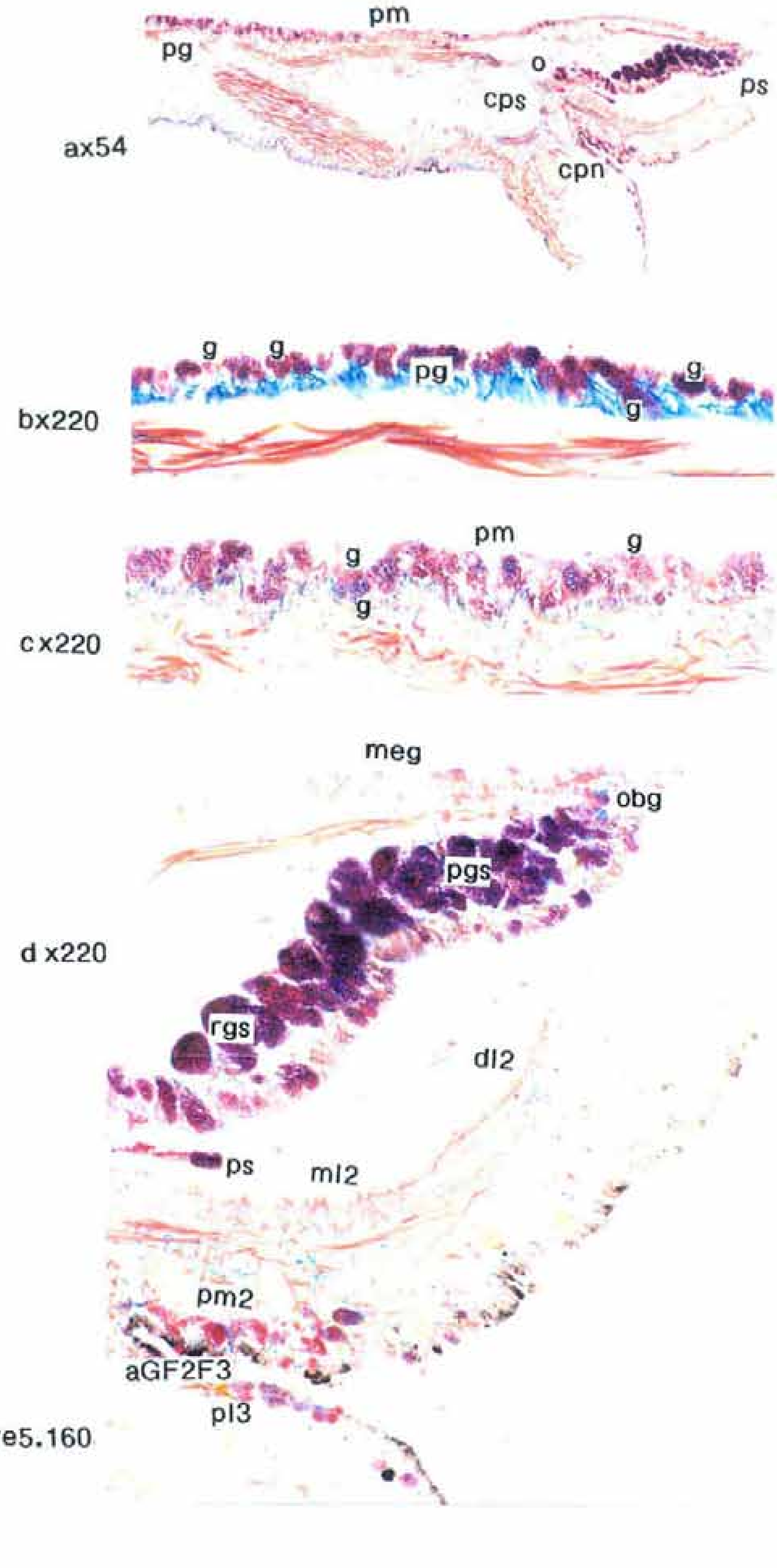

pm
pg
o
ps
cps
ax54
cpn
g
g
pg
g
bx220
g
pm
g
g
g
cx220
meg
obg
pgs
d x220
rgs
dl2
ps
ml2
pm2
aGF2F3
pl3

Figure5.160

5.6.3.3. Lower External Mantle and Mantle Margin.

The most obvious histological differences between the upper Mantle Margin and the Lower Margin in this species are:
1., that the secretory Glands of MedF1. which are again divided into purple staining distally and red staining proximally with Azan. (pg and rg in Figure 5.161. b) are more prominent in the lower Mantle Margin: and
2. The array of Granular Cytoplasm Secretory Glands in Apical Groove F2F3. (aGF2F3 in Figure 5.161. c) is more prominent in the lower Mantle Margin. and there is a ridge of secretory tissue here overlying subepithelial glands which stain yellow with Azan (y in Figure 5.161. c).

FIGURE 5.161. *Saccostrea echinata* - Lower Mantle Margin.

a. L.M. x 77 Azan External Mantle and Mantle Margin.

b. L.M. x 310 Azan Secretory Glands of MedF1..

c. L.M. x 310 Azan Secretory STructures of Apical Groove F2F3..

a
x77

b
x310

c
x310

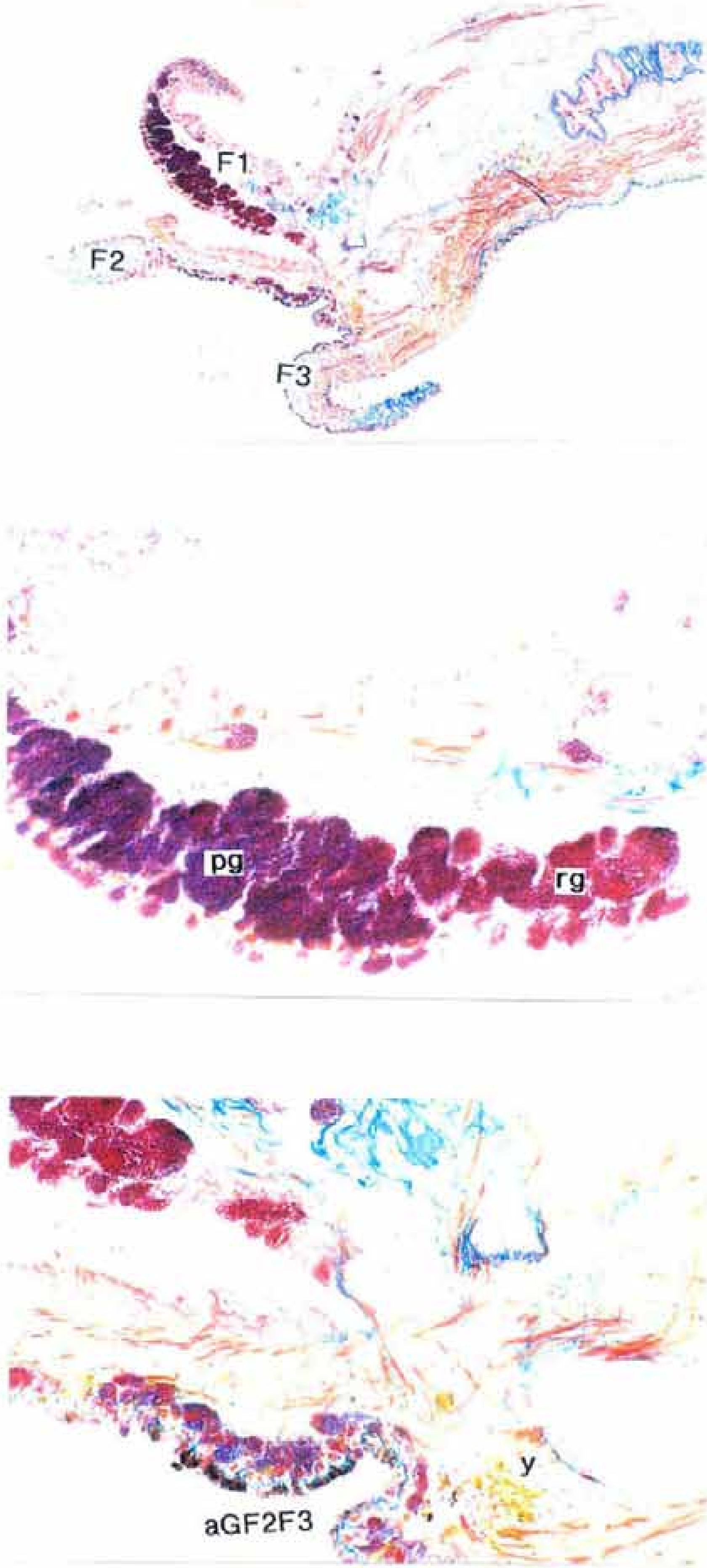


Figure
5.161

5.6.4. *SACCOSTREA CUCCULLATA.*

5.6.4.1. Upper and Lower Mantle Margins.

The lower Valve of this oyster has an anterior periphery without obvious internal Growth Processes of Prismatic Layer, and a posterior periphery which rises normal to the attached surface with external Growth Processes, as has the upper valve.

The lower Anterior Mantle Margin (Figure 5.162. a) has a very different array of secretory structures from either the lower posterior Mantle Margin (Figure 5.162. b and c) or the upper Mantle Margin (Figure 5.162. d).

In all Mantle Margins there are three Marginal Mantle Folds (F1, F2 and F3. in Figure 5.162. a, b, c and d). The apex of Groove F1F2. in each case has an Omega Gland (o in Figure 5.162. a, b c and d). A pleated secretion is in each case generated medial to the Omega Gland (PS in Figure 5.162. a, b, c and d).

Ovoid Blue Glands are more extensive and extend further proximally in the anterior lower Mantle Margin than the other Mantle Margins (obg in Figure 5.162. a, b, c and d). The Granular Cytoplasm Secretory Glands of MedF1. mostly stain pinkish-red with A.B./M.S.B. in the lower Mantle Margin (g in Figure 5.162. a, b and c), but are far more prominent posteriorly than anteriorly. In the Upper Mantle Margin the equivalent glands stain orange-red with A.B./M.S.B. (g in Figure 5.162. d).

A gland which stains green with A.B./M.S.B. forms almost a tissue beneath MedF2. in the anterior lower Mantle Margin (gr in Figure 5.162. a) but is absent or nearly so from the other Mantle Margins.

FIGURE 5.162. *Saccostrea cucculata.* L.M. Upper and Lower Mantle Margins.

a.x 77 A.B./M.S.B. Anterior lower Mantle Margin.

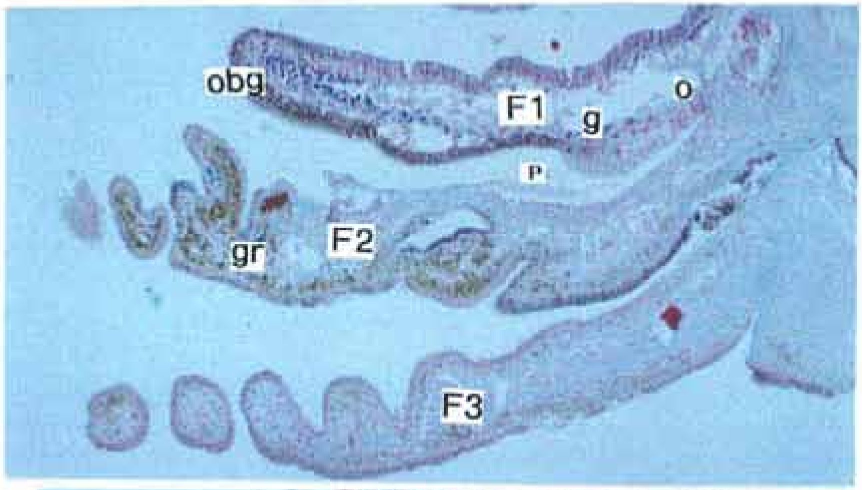
b.x 150 A.B./M.S.B. Posterior lower Mantle Margin.

c.x 310 A.B./M.S.B. Posterior lower Mantle Margin.

d.x 77 A.B./M.S.B. Two Upper Mantle Margins

a

x77



b

x150

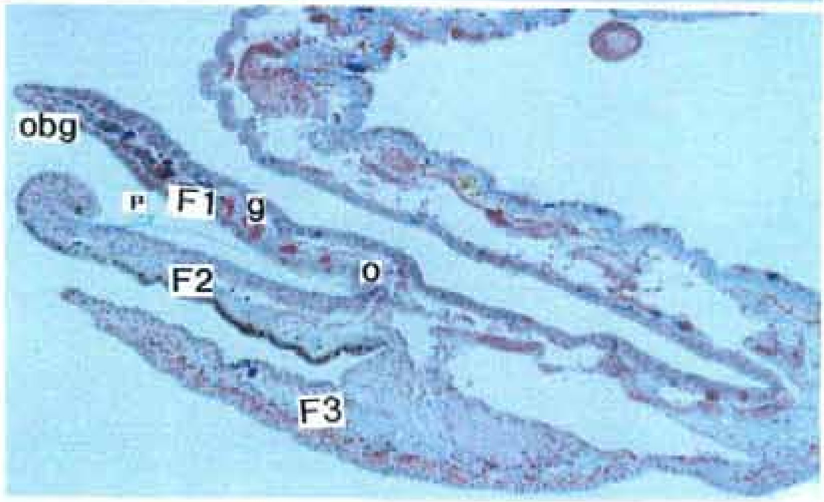
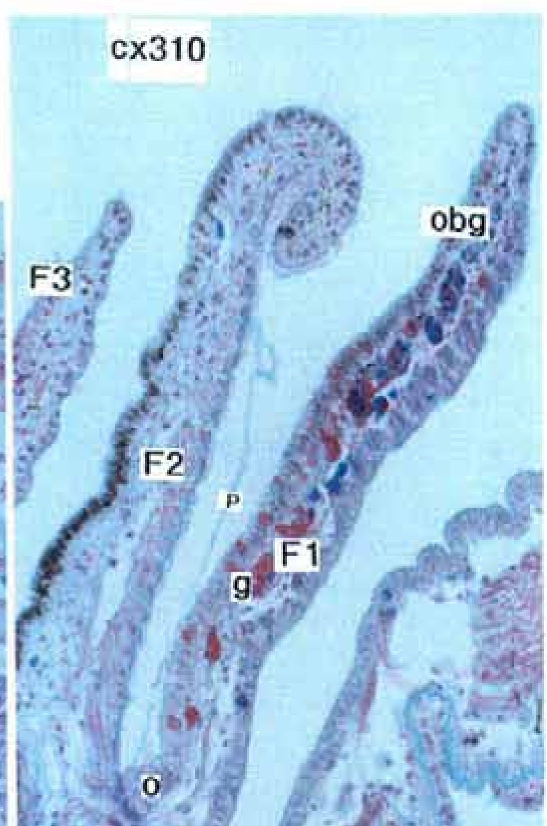
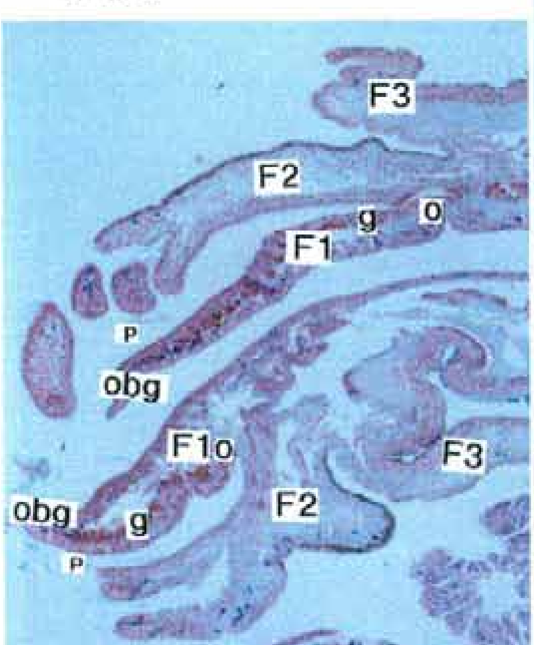


Figure 5.162



d x77



5.7. ANOMIOIDEA.

5.7.1. *PLACUNA PLACENTA.*

5.7.1.1. Upper and Lower Mantle Margins.

Both upper and lower valves of this species have a corrugated fairly opaque outer Shell Layer and an inner Shell Layer composed of non-corrugated translucent sheets.

The upper External Mantle has numerous intra-epithelial Trabecular Turquoise Glands (TT in Figure 5.163. c).

On both Upper and Lower Mantle Margins there are three Mantle Margin Folds (F1, F2 and F3 in Figure 5.163. a, b, c and d).

F2. is trilobate on both the Upper and Lower Mantle Margins (l1, l2 and l3 in Figure 5.163. a and c).

The outer two lobes of both Upper and Lower Mantle Margins are lined by secretory epithelia (e in Figure 5.163. c).

The medial lobe of the upper Mantle Margin is lined by squamous epithelium and relatively acellular (l3 in Figure 5.163. b).

An Omega Gland at Apical Groove F1F2. generates a Pleated Secretion of Groove F1F2. (o and P respectively of Figure 5.163. c).

The LatF3. surface epithelium (e in Figure 5.163. d) appears in radial section to be raised into tufts about 5 cells across with prominent basal nuclei and clear apical cytoplasm (with Azan).

Despite very different shells, the histologies of the External Mantles and Mantle Margins of *Placuna placenta, Spondylus lamarcki* and *Amusium pleuronectes* are remarkably similar and very different from those of the oysters and pearl oysters.

FIGURE 5.163. *Placuna placenta* – Upper and Lower Mantle Margins.

a. L.M. x 77 Azan Lower Ventral Mantle Margin, Radial Section.

b. L.M. x 77 Azan Upper Ventral Mantle Margin, Radial Section.

c. L.M. x 310 Azan Upper Ventral Mantle Margin F1. and F2..

d. L.M. x 310 Azan Upper Ventral Mantle Margin LatF3.

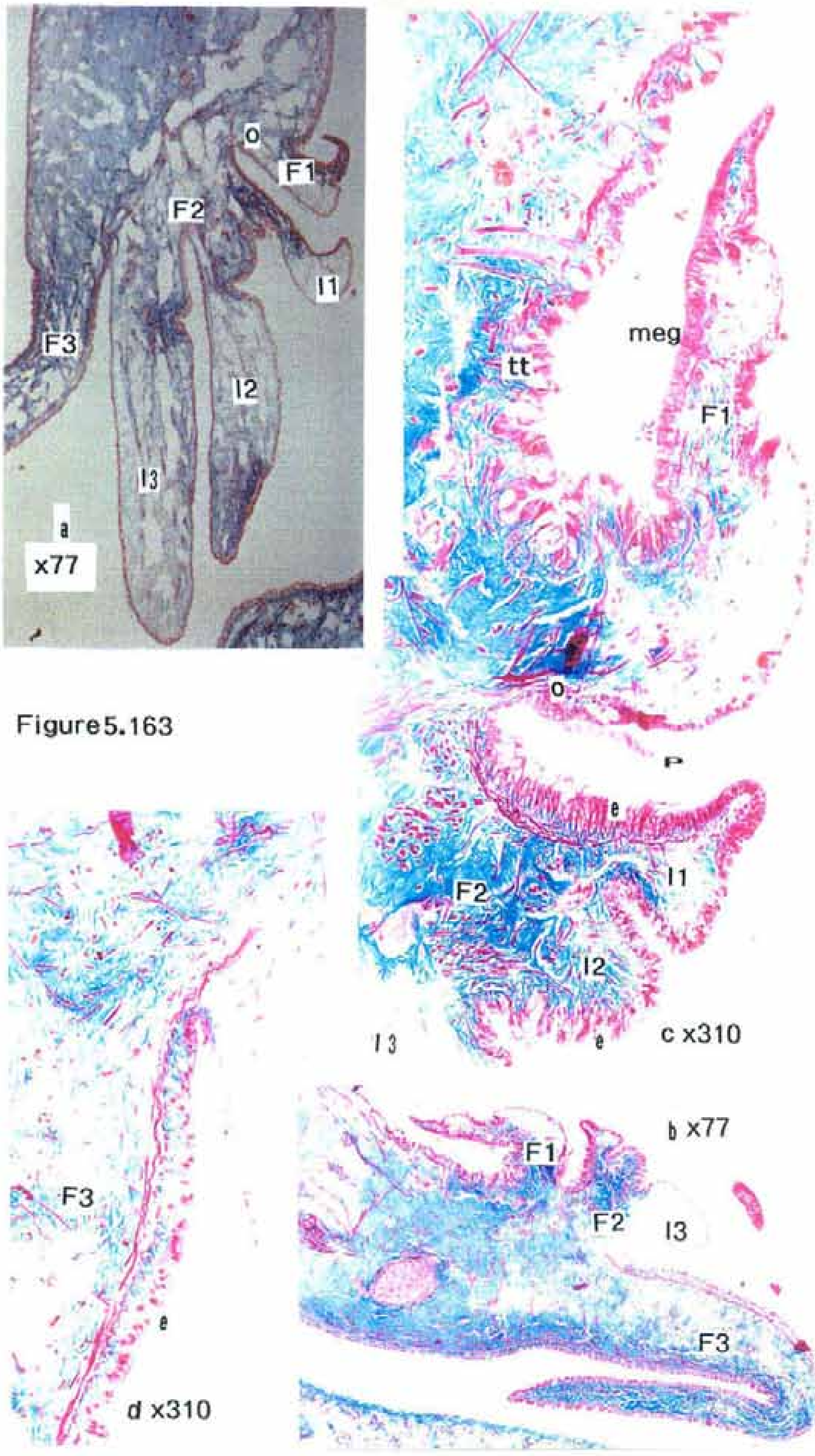

o
F1
F2
I1
F3
I2
I3
a
x77
meg
tt
F1
o
P
e
I1
F2
I2
I3
e
c x310
F3
e
d x310
b x77
F1
F2
I3
F3

Figure5.163

5.8. PECTINOIDEA.

5.8.1 *SPONDYLUS LAMARCKI.*

5.8.2. *AMUSIUM PLEURONECTES.*

5.8.1.1. – 5.8.2.1. Upper and Lower Mantle Margins.

The Upper and Lower Mantle Margins of the above species are sufficiently similar for them to be described together. The major difference between the Mantle Margin of the two species is the comparatively large size of F3. in *Amusium pleuronectes.*

There are three Marginal Mantle Folds on both the upper and lower Mantle Margins called F1., F2. and F3. from lateral to medial (F1, F2 and F3 in Figure 5.164. a, b, c, d, e and f).

Both upper and lower LatF1. have a Mantle Edge Gland of columnar epithelium (m in Figure 5.164. a, b, c, e and f).

On both LatF1. and MedF1. there are populations of Trabecular Turquoise Glands. They are subepithelial on LatF1. and intraepithelial on MedF1. (TT in Figure 5.164. d).

Apical Groove F1F2. on both mantles has a laterally placed tuft of secretory epithelium – an Omega Gland (o in Figure 5.164. a, b, c, d, e and f) – which generates a secretion which stains turquoise with A.B./M.S.B.

F2. bears an array of sensory structures including eyes (e in Figure 5.164. b and e) and is usually multilobate. There is a pronounced secretory epithelium in Apical Groove F2F3. (s in Figure 5.164. a and f).

There is a tufted secretory epithelium on LatF3. (t in Figure 5.164. a and c which stains turquoise with A.B./M.S.B.).

The Circum-pallial Nerve (n in Figure 5.164. a) is not immediately physically related to Apical Groove F1F2..

FIGURE 5.164. *Spondylus lamarcki* and *Amusium pleuronectes.*
Mantle Margins.

a. L.M. x 40 Mallorys *Spondylus lamarcki* Upper Mantle Margin.

b. L.M. x 40 A.B./M.S.B. *Spondylus lamarcki* Upper Mantle Margin.

c. L.M. x 40 A.B./M.S.B. *Spondylus lamarcki* Lower Mantle Margin.

d. L.M. x 310 Mallorys *Spondylus lamarcki* Upper Mantle Margin
Apical Groove F1..

e. L.M. x 40 A.B./M.S.B. *Amusium pleuronectes* Upper Mantle.

f. L.M. x 40 M.S.B. *Amusium pleuronectes* Lower Mantle.

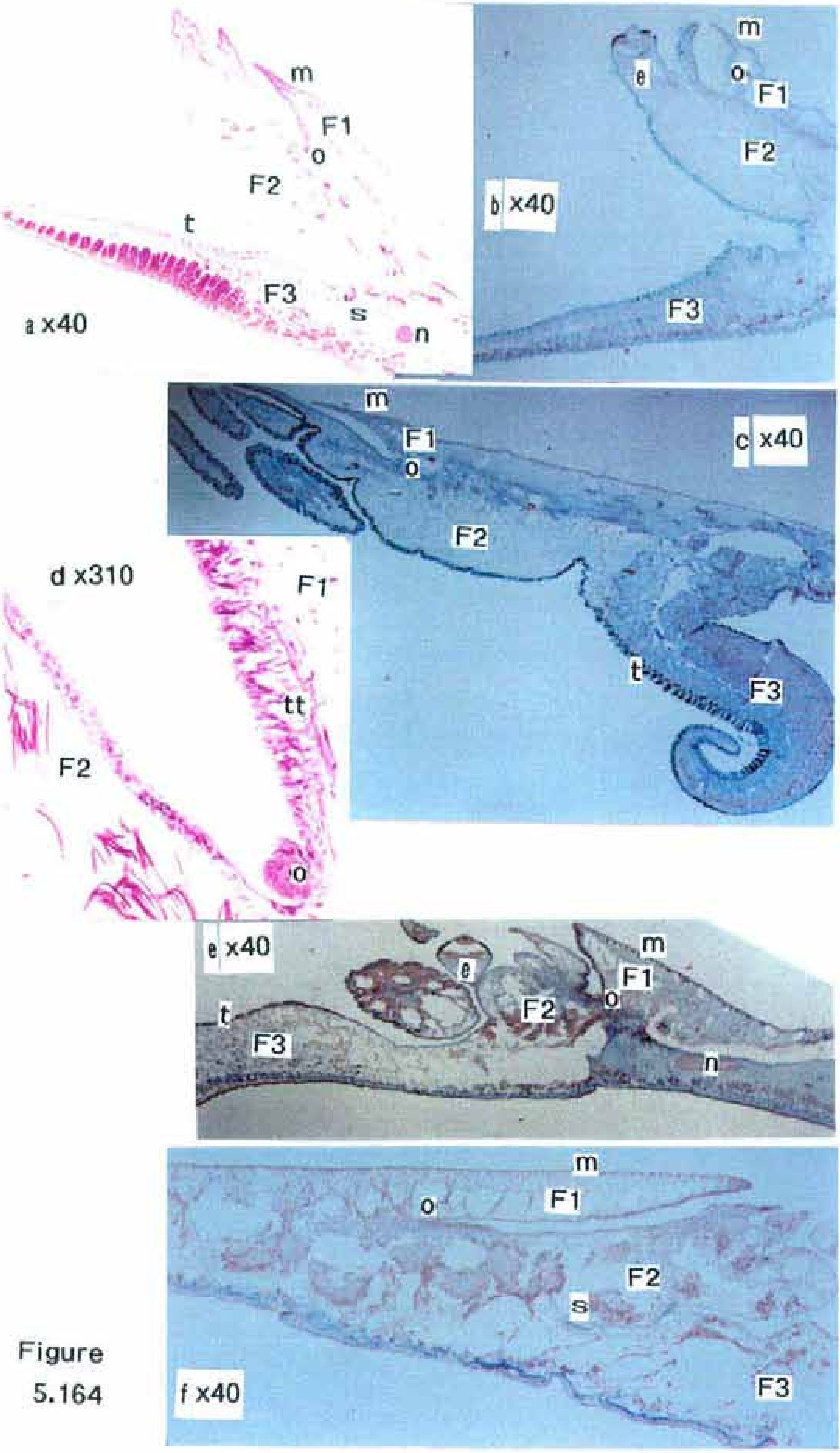

a x40
m
F1
o
F2
t
F3
s
n
b x40
e
m
o
F1
F2
F3
c x40
m
F1
o
F2
t
F3
d x310
F1
tt
F2
o
e x40
e
m
F1
o
F2
t
F3
n
f x40
m
o
F1
F2
s
F3

Figure 5.164

5.9. LIMOPSOIDEA.

5.9.1. *MELAXINAEA VITREA.*

5.9.1.1. External Mantle and Proximal part of Mantle Margin.

The Pallial Mantle is almost entirely composed of the Beeswax Glands (b in Figure 5.165. a). These constitute a glandular tissue of Granular Cytoplasm Secretory Glands of at least four species (labelled 1, 2, 3 and 4 in Figure 5.165 a).

Opposed to the Beeswax Glands across the Pallial Space (ps in Figure 5.165 a) is the Inner Shell Layer, the decalcified Organic Matrix of which is shown in Figure 5.165. (isl).

Outside the Inner Shell Layer is the Outer Calcarous Shell Layer (osl in Figure 5.165. b, c and d) which is produced by the secretory structures of LatFl., proximal to the Mantle Edge Gland (m in Figure 5.166. a and b). These consist of at least three different species of Granular Cytoplasm Secretory Gland marked 1, 2 and 3 in Figure 5.165. c) and Trabecular Turquoise Glands (tt in Figure 5.165 d).

The Secretory Glands (S in Figure 5.165. c) where with Mallorys they stain predominantly amber but also red and purple are medial to the glands which produce the middle lamina of the trilaminate outer non-calcareous Shell Layer (q.v. rg in Figure 5.166. a and b), and thus may be the source of the Outer Lamina of the outer Shell Layer but this is uncertain as the outer lamina is refractory to staining and difficult to section in situ.

FIGURE 5.165. *Melaxinaea vitrea* L.M. External Mantle and Mantle Margin.

a.	L.M. x	Mallorys	Pallial Mantle.
b.	L.M. x	Mallorys	Mantle Margin.
c.	L.M. x	Mallorys	Proximal part of Extra-pallial Region.
d.	L.M. x	A.B./M.S.B.	Proximal part of Extra-pallial Region.

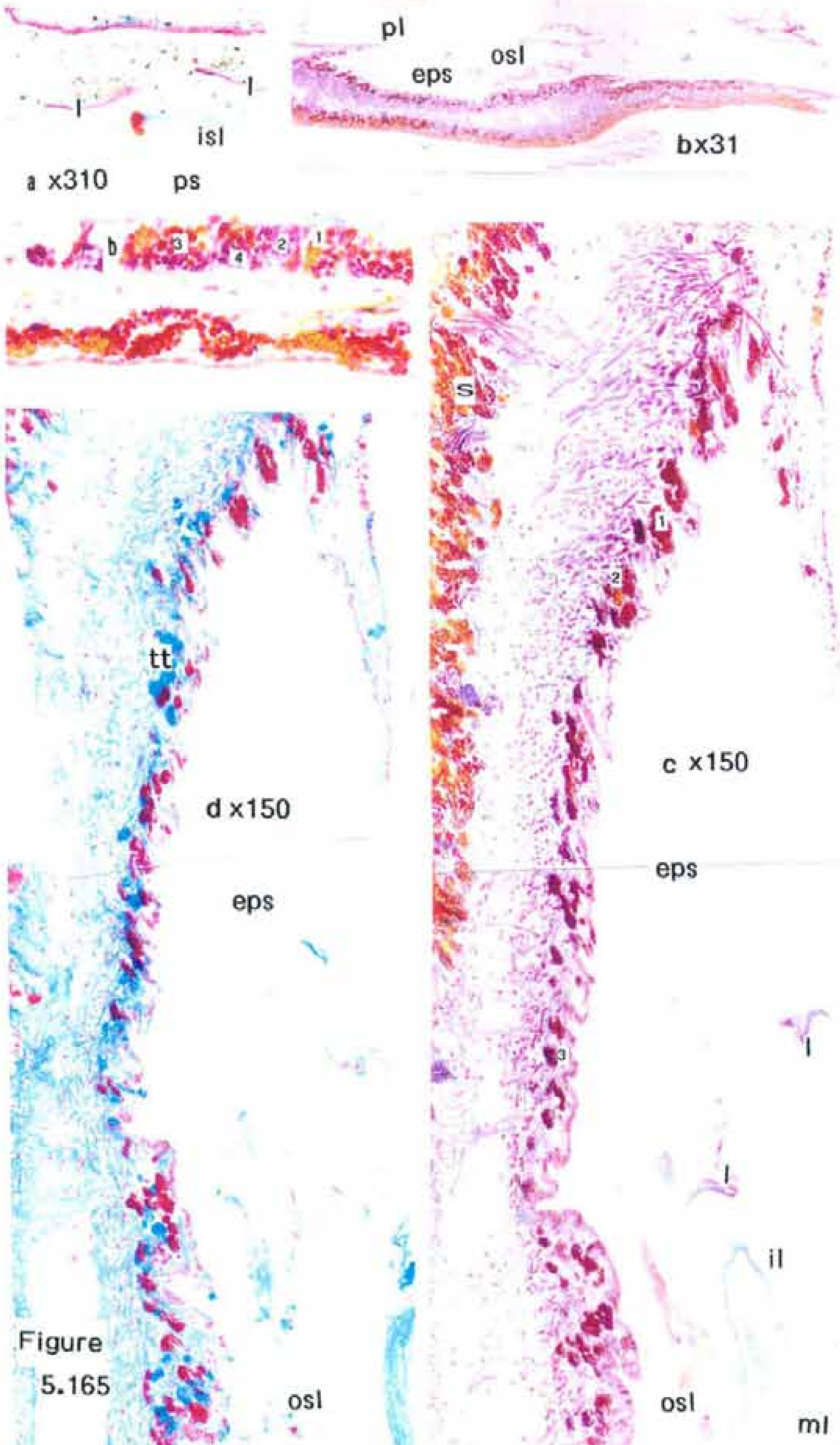

pl
osl
eps
l
l
isl
bx31
a x310
ps
b
3
4
2
1
S
1
2
tt
c x150
d x150
eps
eps
l
3
l
il
osl
osl
ml

Figure 5.165

5.9.1. *MELAXINAEA VITREA.*

5.9.1.2. Mantle Edge Gland and Folds F1F2..

The Mantle Edge Gland (m in Figure 5.166. a and b) in this species is relatively much reduced consisting of a radially restricted area of elongate columnar epithelial cells. Immediately distal to this is a deep notch (n in Figure 5.166. a and b) into which numerous subepithelial Trabecular Turquoise Glands secrete (tt in Figure 5.166. b).

This secretion may be part of the origin of the laminae which stain magenta with Mallorys associated with the Outer Shell Layer (l in Figure 5.165. a and c Figure 5.166. a).

The epithelium of the Distal LatF1. beyond the notch is pigmented (p in Figure 5.165. b) and the subepithelium appears devoid of Trabecular Turquoise glands and has what appears to be only one type of Granular Cytoplasm Secretory Gland. (g in Figure 5.166. a and b).

What appear to be primitive light sensory organs are located on terminal F1. (e in Figure 5.166. b).

On the lateral surface of Apical F1F2. is the Omega Gland (o in Figure 5.166. a and b) which elaborates the inner layer of the outer non-calcerous Shell Layer (il in Figure 5.166. a and b). On Proximal LatF2. are Receptacle Glands which stain amber with Mallorys and yellow with A.B./M.S.B. (rg in Figure 5.166. a and b) and their associated subepithelial Glands. They appear to be the origin of the middle lamina of the Outer non-calcareous Shell Layer (ml in Figure 5.166. a and b).

The overall patterns in External Mantle and Mantle Margin tissue differentiation in Limopsoidea and Arcoidea are very similar throughout these groups and, despite the fact that their Shell Layer assemblages are radically different, also very similar to that seen in the oysters and pearl oysters They bear very little resemblance to those members of the Heterodonta which have similar Shell Layers.

FIGURE 5.166. *Melaxinaea vitrea* Mantle Edge Gland and Folds F1F2..

a. L.M. x 150 Mallorys.

b. L.M. x 150 A.B./M.S.B.

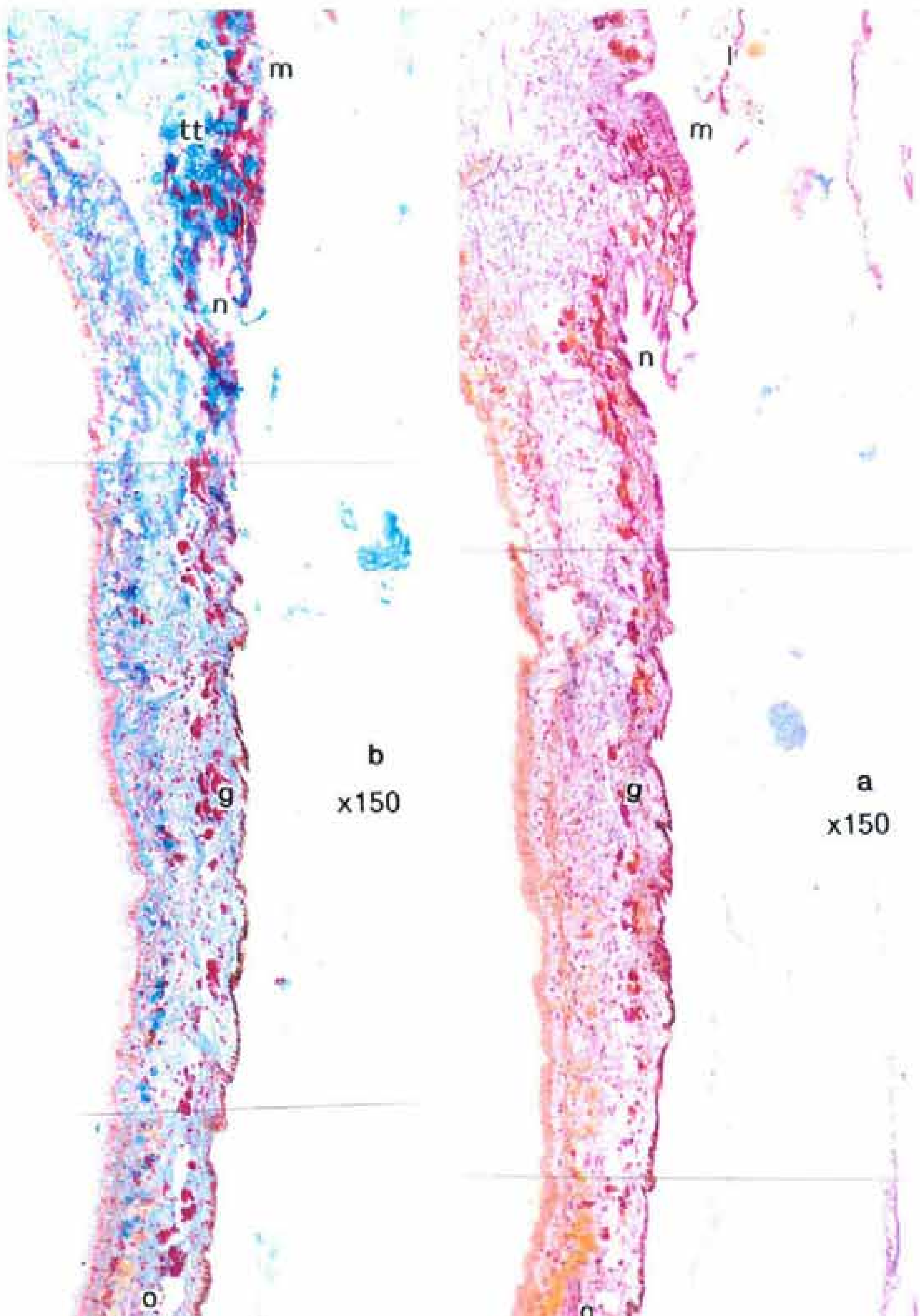


Figure 5.166

5.10. ARCOIDEA.

5.10.1. *ANADARA ANTIQUATA.*

5.10.1.1. External Mantle and Mantle Margin of *Anadara antiquata.*

In tangential section the External Mantle of this species is raised into ridges (r in Figure 5.167 a) of tissue whose periodicity roughly corresponds with that of the fluting of the Valve. The External Mantle epithelium bears Beeswax Glands (b in Figure 5.167. a).

Distal to the Pallial Line, the sequence of tissues and Glands is shown in Figure 5.167. b, - on the medial surface of the Extrapallial Space (eps in Figure 5.167. b) is the Proximal LatF1., the columnar epithelium of the Mantle Edge Gland, and a notch with subepithelial Secretory Glands (pl1, meg, n and s respectively in Figure 5.167. b). Terminal F1. is notched and is in part, the source of the Inner Lamina (il in Figure 5.167. b and c) of the trilaminate outer non-calcareous Shell Layer. The Middle Lamina (ml in Figure 5.167. b and c) is produced by secretory structures on LatF2. The Outer Lamina which is difficult to section and refractory to staining is necessarily produced by secretory structures medial to F2.

5.10.2. *ANADARA PILULA.*

5.10.2.1. External Mantle and Mantle Margin of *Anadara pilula.*

The External Mantle of this species has a prominent Beeswax Gland (Figure 5.167. d). There are at least three different unicellular glands constituting the Beeswax Gland. (1, 2 and 3 in Figure 5.167. d).

The tissues, glands and secretions of the Mantle Margin of this species are the same as in *Anadara antiquata* and in Figure 5.167. e are similarly labelled as for that species.

FIGURE 5.167 External Mantle and Mantle Margins of *Anadara antiquata* and *Anadara pilula*.

a. L.M. x 77 Azan Tangential section of External Mantle of *A. antiquata*.

b. L.M. x 31 M.S.B. Mantle Margin of *A. antiquata*.

c. L.M. x 230 M.S.B. Inner and Middle lamina of outer non-calcareous Shell Layer of *A. antiquata*.

d. L.M. x 310 Azan *Anadara pilula*. Beeswax Glands.

e. L.M. x 77 Mallorys *Anadars pilula*. Mantle Margin.

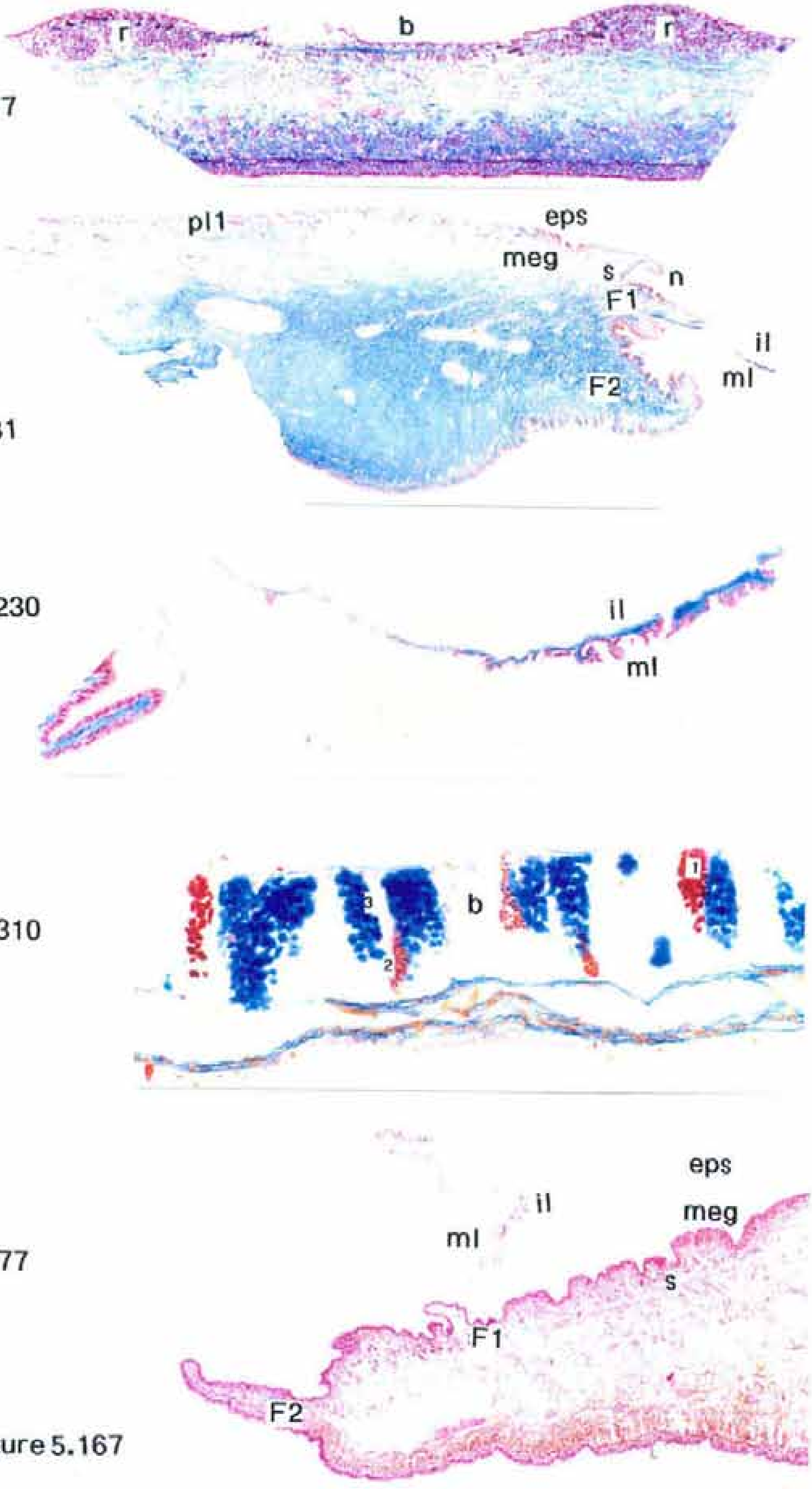

r
b
r
a x77
pl1
eps
meg
s
n
F1
il
ml
F2
b x31
c x230
il
ml
d x310
b
1
2
3
eps
il
meg
ml
e x77
s
F1
F2

Figure 5.167

5.10.3. *ARCA ALADDIN.*

5.10.3.1. External Mantle and Mantle Margin.

The External Mantle of *Arca aladdin* has a Beeswax Gland (b in Figure 5.168 a and b) with at least three species of unicellular secretory gland (1, 2 and 3 in Figure 5.168. a). The Beeswax Gland is the medial lining of the Pallial Space (ps in Figure 5.168. a, b and c).

The Pallial Space is divided from the Extra-pallial Space (eps in Figure 5.168. b, c and d) by the Pallial Line Junction (pl in Figure 5.168. b).

The medial lining of the Extrapallial Space is occuped proximally by Proximal LatF1. (pL1 in Figure 5.168. b, c and d) with its distinctively staining subepithelial Glands and distally by the highly differentiated elongate columnar cells of the Mantle Edge Gland (meg in Figure 5.168. b and d). Terminal F1. is host to a specialised Unicellular Granular Cytoplasm Secretory Gland and also Ovoid Blue Glands (t and ob in Figure 5.168. d and e).

MedF1. hosts elongate glands which stain orange-brown with M.S.B. (mfl in Figure 5.168. e). The Omega Gland is on the lateral side of Apical Groove F1F2. (o in Figure 5.168. b and e) which secretes the Inner Lamina of the trilaminate outer non-calcareous Shell Layer (blue staining with Azan) (il in Figure 5.168. b and e).

The Receptacle Glands (r in Figure 5.168. b and e) are on Proximal LatF2. and secrete the middle lamina of the trilaminate outer non-calcareous Shell Layer - red staining with Azan (ml in Figure 5.168. b and e).

FIGURE 5.168. *Arca aladdin* - External Mantle and Mantle Margin.

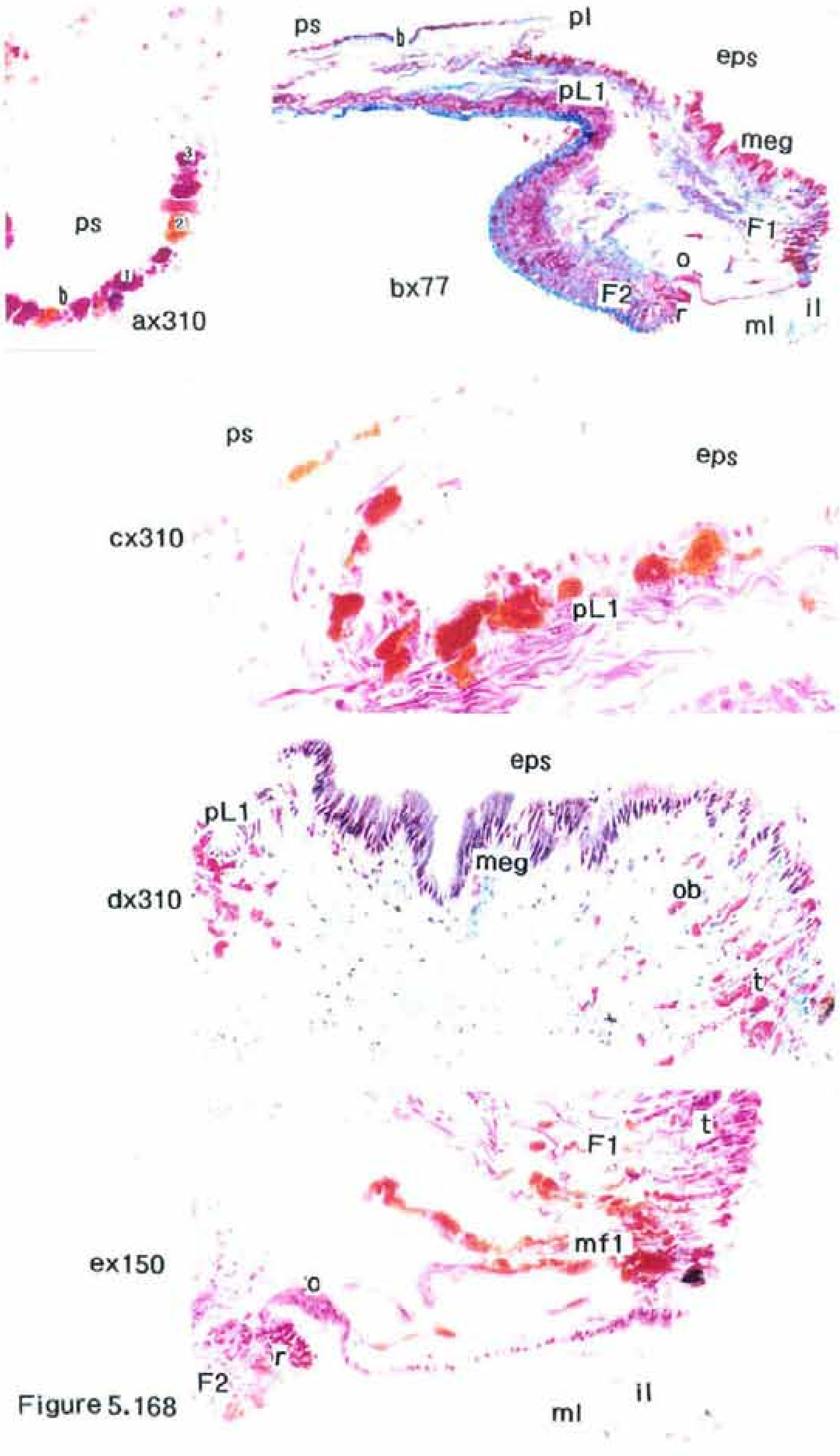
a. L.M. x 310 Mallory's External Mantle, Beeswax Glands.

b. L.M. x 77 Azan External Mantle and Mantle Margin.

c. L.M. x 310 Mallory's Pallial Line Junction and Proximal Lat F1..

d. L.M. x 310 M.S.B. Mantle Edge Gland and Glands of Terminal F1..

e. L.M. x 150 M.S.B. MedF1. and LatF2..


ps
b
pl
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pL1
meg
3
2
1
ps
F1
o
b
bx77
F2
ax310
r
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il
ps
eps
cx310
pL1
eps
pL1
meg
dx310
ob
t
t
F1
mf1
ex150
o
r
F2
ml
il

Figure 5.168

5.10.4. *BARBATIUM AMYGDALUMTOSTUM*

5.10.4.1. External Mantle and Mantle Margin.

The External Mantle of this species bears Beeswax Glands (b in Figure 5.169 a) forming the medial surface of the Pallial Space (ps in Figure 5.169. a). The Mantle Margin has three folds F1., F2. and F3. from lateral to medial (F1, F2 and F3 respectively in Figure 5.169. b and d).

LatF1. is relatively elongate and the Mantle Edge Gland (meg in Figure 5.169. b, c and d) is situated towards the middle of it. There is thus an unusually extensive Terminal Epithelium (t in Figure 5.169. d). Some parts of the Mantle Edge Gland subepithelium are devoid of glands which are abundant in other parts (g in Figure 5.169. c and d).

Ovoid Blue Glands (ob in Figure 5.169. d) occur in terminal F1..

The lateral side of Apical Groove F1F2. bears an Omega Gland which generates the Inner Lamina (il in Figure 5.169. b and d) of the outer trilaminate non-calcareous Shell Layer. This stains blue with Azan. The Middle Lamina of the outer Shell Layer, which stains red with Azan, (ml in Figure 5.169. b and d), is largely secreted by the Receptacle Glands (r in Figure 5.169. b and d) of Proximal LatF2.. These recieve the secretions of the underlying Granular Cytoplasm Secretory Glands). (s in Figure 5.169. d).

LatF3. (lF3. in Figure 5.169. d) has conspicuous secretory glands in the subepithelium.

FIGURE 5.169. *Barbatium amygdalumtostum* - External Mantle and Mantle Margin.

a. L.M. x 230 Mallorys. Beeswax Glands of the Pallial Region.

b. L.M. x 46 M.S.B. Mantle Margin.

c. L.M. x 310 Azan. Mantle Edge Gland.

d. L.M. x 150 Azan. Mantle Margin Folds F1., F2. and F3..

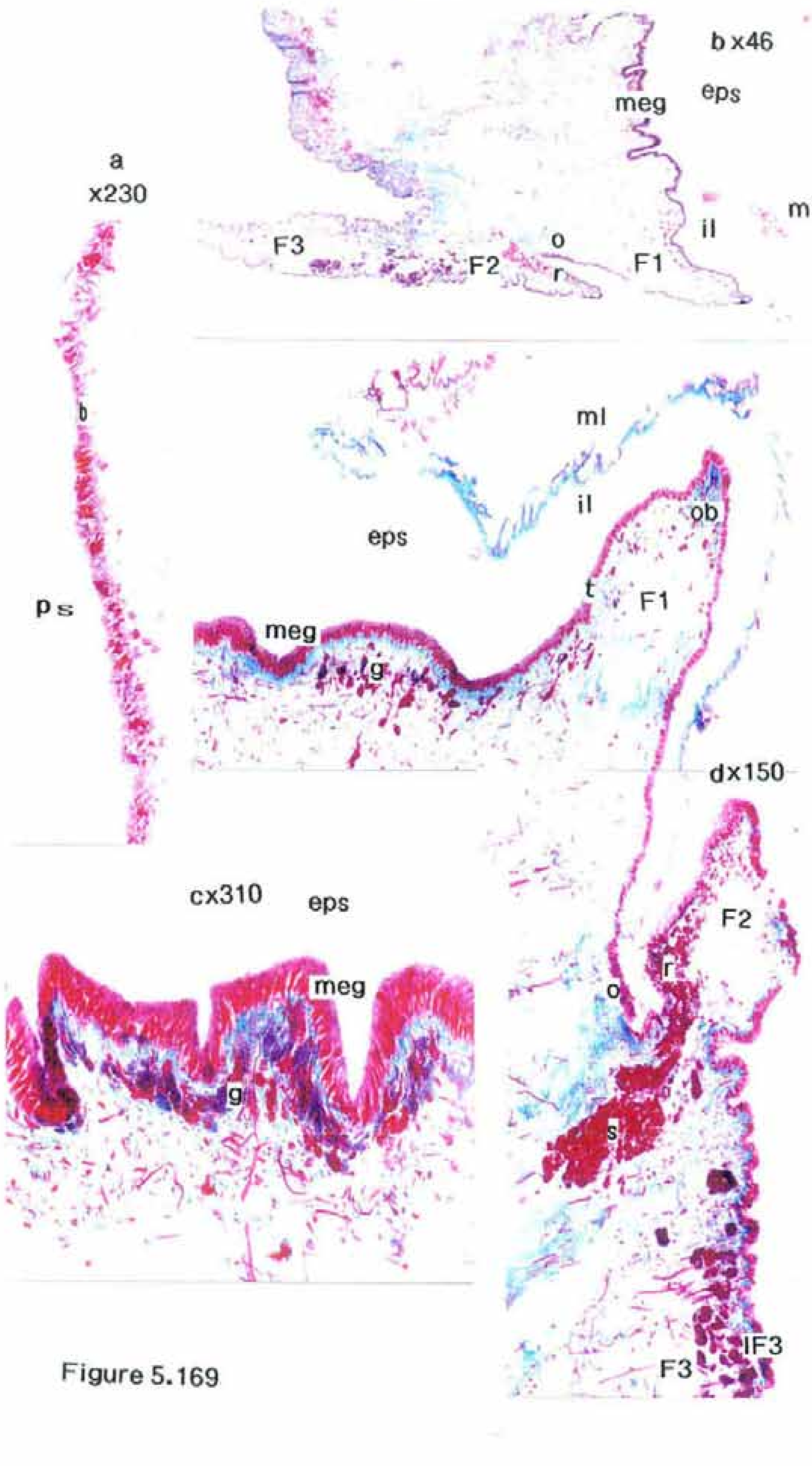

b x46
eps
meg
ml
il
F3
o
F2
r
F1
a
x230
b
ps
ml
il
ob
eps
t
F1
meg
g
dx150
F2
r
o
s
IF3
F3
cx310
eps
meg
g

Figure 5.169

5.10.5. *MESOCIBOTA LUANA.*

5.10.5.1. External Mantle and Mantle Margin.

The External Mantle has Beeswax Glands on the medial surface of the Pallial Space (b and ps in Figure 5.170. a and b).

The Mantle and Valve are joined at the Pallial Line (pl in Figure 5.170. b).

The Extra-pallial Space (eps in Figure 5.170) is bounded medially, from proximal to distal, by the Proximal LatF1., the Mantle Edge Gland and Terminal Epithelium (pL1, meg and t respectively in Figure 5.170. b and c).

MedF1. (mF1 in Figure 5.170. b and d) is deeply folded, has columnar surface epithelium, and subepithelial secretory glands (sg in Figure 5.170. b and d).

The Omega Gland (o in Figure 5.170. d) is on the lateral side of Apical Groove F1F2. There are Receptacle Glands and their associated subepithelial Secretory Glands in the Proximal LatF2. Region (r and s respectively in Figure 5.170. d). The latter are of at least two species staining amber and magenta with Mallorys.

LatF3. has subepithelial secretory glands (lF3. in Figure 5.170. d).

FIGURE 5.170. *Mesocibota luana* External Mantle and Mantle Margin.

a. L.M. x 310 Mallorys External Mantle.

b. L.M. x 77 Mallorys Mantle Margins.

c. L.M. x 310 M.S.B. Mantle Edge Gland.

d. L.M. x 150 Mallorys MedF1., Apical Groove F1F2. and LatF3..

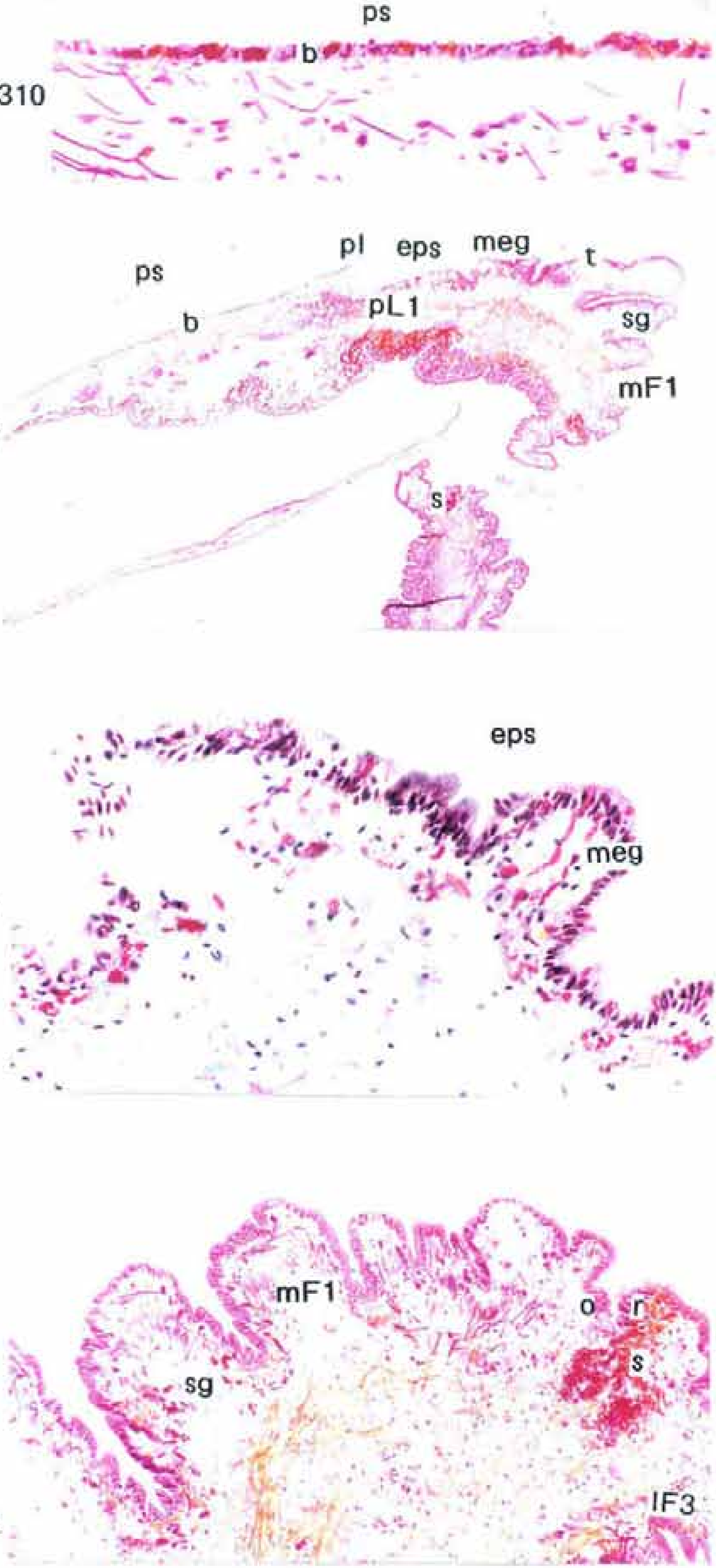

ps
b
ax310
ps
pl
eps
meg
t
b
pL1
sg
mF1
bx77
s
eps
meg
cx310
mF1
o
r
s
sg
dx150
lF3

Figure5.170

5.10.6. *TRISIDOS TORTUOSA.*

5.10.6.1. Mantle Margin.

Distal to the junction of Mantle and Valve at the Pallial Line the Extra-pallial Space (eps in Figure 5.171. a, b and c) is bordered medially by the Proximal LatF1., the Mantle Edge Gland and the Terminal Epithelium, (pL1, meg and t respectively in Figure 5.171. a, b and c). Proximal LatF1. has a dense population of subepithelial secretory glands.

The elongate columnar epithelium of the Mantle Edge Gland (meg in Figure 5.171. b), overlies a subepithelium which also hosts a number of secretory glands (sg in Figure 5.171. b).

MedF1. is divided into Distal MedF1. (dmf1 in Figure 5.171a and c) and Proximal MedF1. by a deep notch (n in Figure 5.171 a and c). The notch divides a subepithelium with numberous large Granular Cytoplasm Secretory Glands distally (g in Figure 5.171. c) from one proximally, which is devoid of those glands.

The Omega Gland on the lateral side of Apical Groove F1F2. (o in Figure 5.171. d), is the source of a secretion which stains blue with Azan.

The medial surface of Apical Groove F1F2. is occupied by a very thin tongue of tissue (t in Figure 5.171. d).

The entire LatF2. is occuped by a unique structure which constitutes about half the volume of F2. and stains yellow with Azan (lF2 in Figure 5.171. d). Within this tissue are two types of unicellular secretory gland. Proximally they stain bright purple with Azan and more distally blue with this stain. (p and b respectively in Figure 5.171. d).

FIGURE 5.171. *Trisidos tortuosa* - LM External Mantle and Mantle Margin.

a.	L.M. x 31	Azan	Mantle Margin.
b.	L.M. x 310	Azan	Mantle Edge Gland.
c.	L.M. x 310	Azan	Terminal Lat F1. and MedF1..
d.	L.M. x 310	Azan	Apical Groove F1F2. and Proximal LatF2..

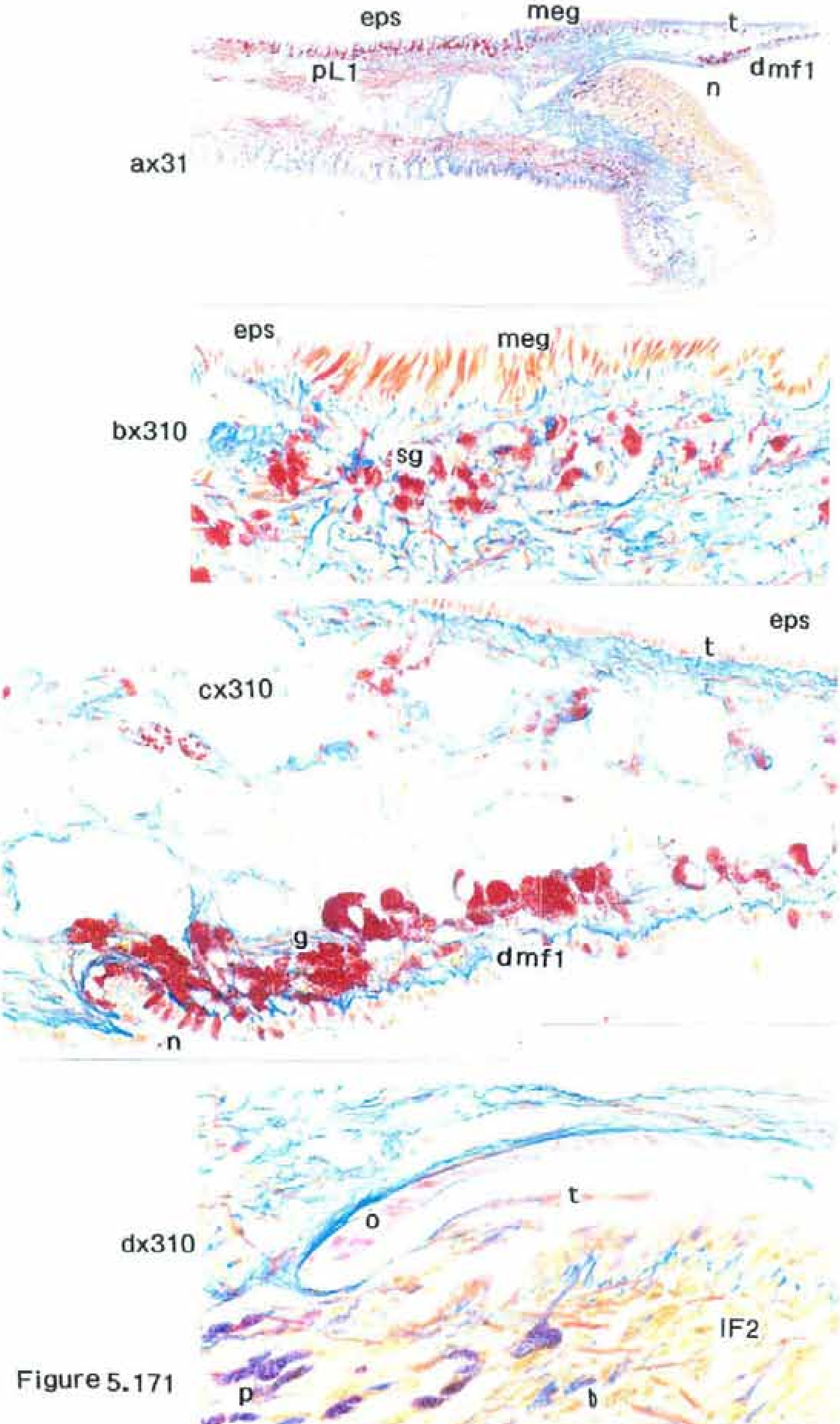

eps
meg
t
pL1
dmf1
n
ax31
eps
meg
bx310
sg
eps
t
cx310
g
dmf1
n
t
o
dx310
IF2
p
b

Figure 5.171

5.11 TELLINOIDEA

5.11.1. *Phylloda foliacea.*

5.11.2. *Tellina sp.*

Both these species have a single Marginal Mantle Groove (g in Figure 5.172. a, b, c and d) and a junction of External Mantle to the medial surface of the Valve at the Pallial Line (j in Figure 5.172. a). The outer non-calcareous Shell Layer which stains amber with Mallorys, pink with Steedmans, red with Azan and bright yellow with A.B./M.S.B. (ol in Figure 5.172. a, b, c and d) is generaged at the apex of the single Marginal Mantle Groove (a in Figure 5.172. b, c and d).

Two physically different secretions which stain similarly with the stains used originate in MedF1. and the distal part of LatF1. (the first), and the proximal part of LatF1. (the second). Both these species have a Proximal LatF1. which consists of two large distinctive outfoldings which parallel the Mantle periphery, (PLF1. in Figure 5.172. a, b, c and d).

In the Tellinoidea, where the Outer Shell Layer is generated in Groove F1F2, the medial surface of Fold F2. is histologically simple and there is no FoldF3. . The variety of External Mantle and Mantle Margin tissue differentiation and its relationship to site of origin and complexity of Shell layer assemblages exhibited by the various species of the Heterodonta is indicated in Figs. 5.172– 5.183.

FIGURE 5.172. L.M. *Phylloda foliacea* and *Tellina sp.* – External Mantle and Mantle Margin.

a. L.M. x 46 Mallorys Mantle and Mantle Margin *Phylloda foliacea.*

b. L.M. x 150 Steedmans Mantle Margin *Phylloda foliacea.*

c. L.M. x 460 Azan Mantle Margin *Phylloda foliacea.*

d. L.M. x 230 A.B./M.S.B. Mantle Margin *Tellina sp.*

a x46

b
x150

c
x460

d
x230

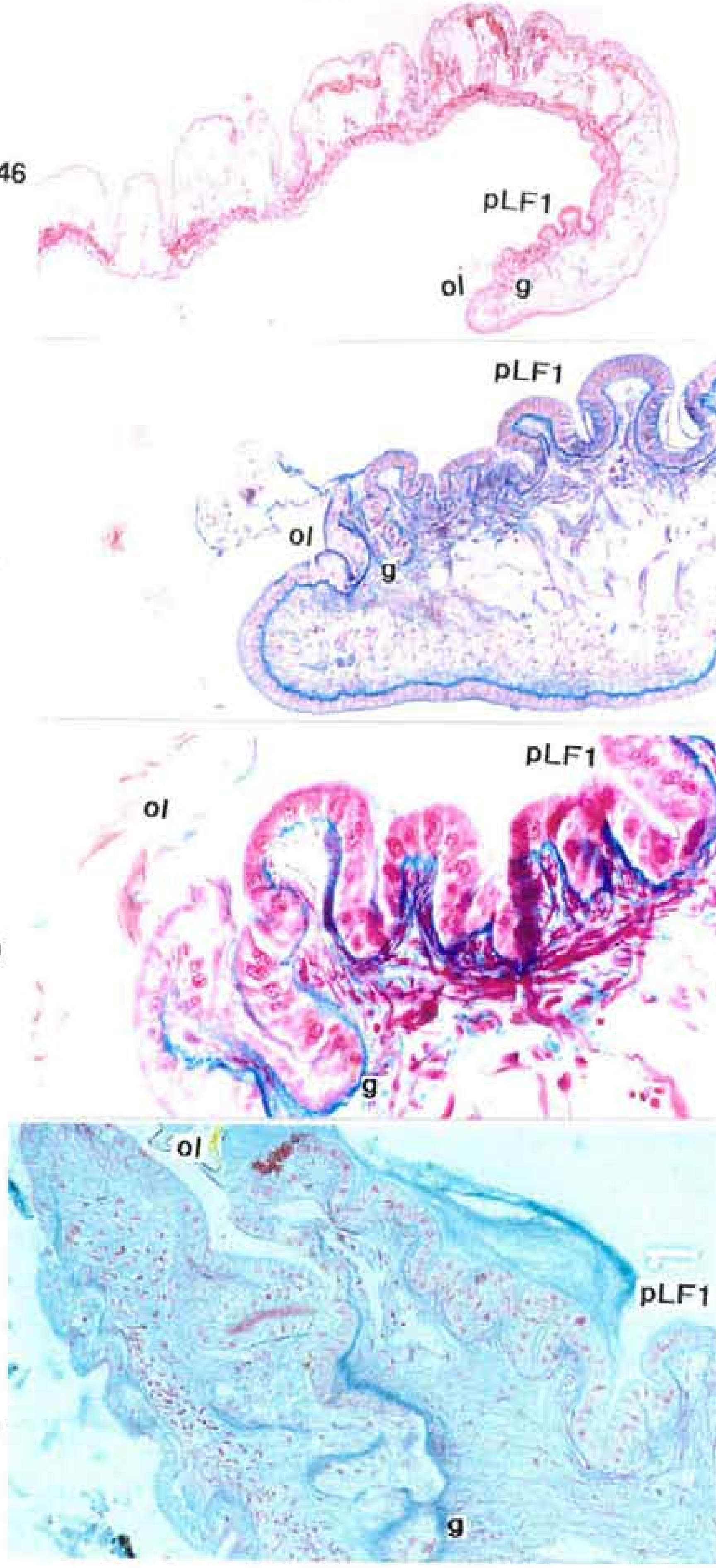


Figure
5.172

5.12. VENEROIDEA.

5.12.1. *Dosinia juvenilis.*

The External Mantle and the medial surface of the Shell of this species are joined at the Pallial Line Junction (j in Figure 5.173. a). There are three Marginal Mantle Folds (F1. F2 and F3 in Figure 5.173). A bilaminate outer non-calcareous Shell Layer issues from Groove F1F2. and recurves around terminal F1. to form the outer lateral layer of the shell. Proximal LatF1. (pLF1. in Figure 5.173. a) is a large vacuolar outfolding.

Distal LatF1. carries the elongate columnar cells of the Mantle Edge Gland (meg in Figure 5.173). and beyond that a less elongate columnar epithelium.

Of the two laminae of the secretion from Groove F1F2. the more lateral in the Groove (i.e., the more medial on the Shell Layer) (lg in Figure 5.173. b and c) is produced by the secretory strucutres of MedF1. including the large secretory glands (sg in Figure 5.173. a. b. c and d). The more medial in the groove (i.e.. the more lateral in the Shell Layer). (ms in Figure 5.173. b. c and d) stains amber with Mallorys. yellow with M.S.B. and red with Azan.

There is a dense mass of unicellular glands in the proximal subepithelium of Groove F2F3. which stain blue with Steedman's and Azan but are refractory to staining with Mallorys or M.S.B. (bg in Figure 5.173. a and d).

FIGURE 5.173. Mantle Margin of *Dosinia juvenilis*

a. L.M. x 120 Steedmans

b. L.M. x 460 Mallorys

c. L.M. x 460 M.S.B.

d. L.M. x 190 Azan.

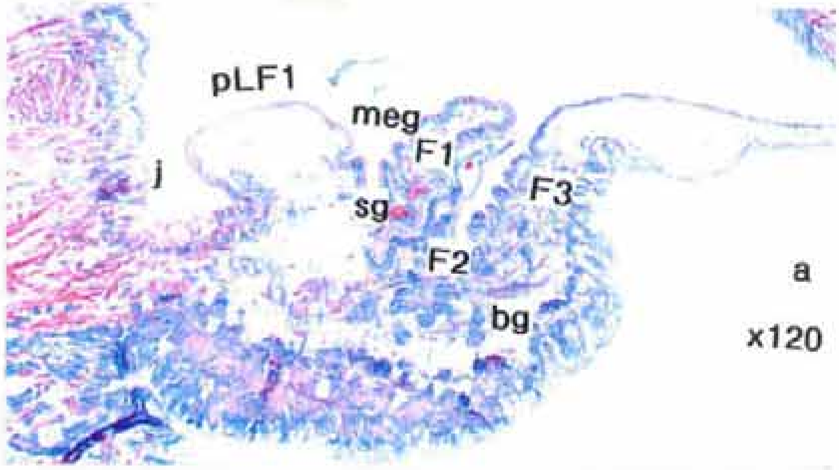
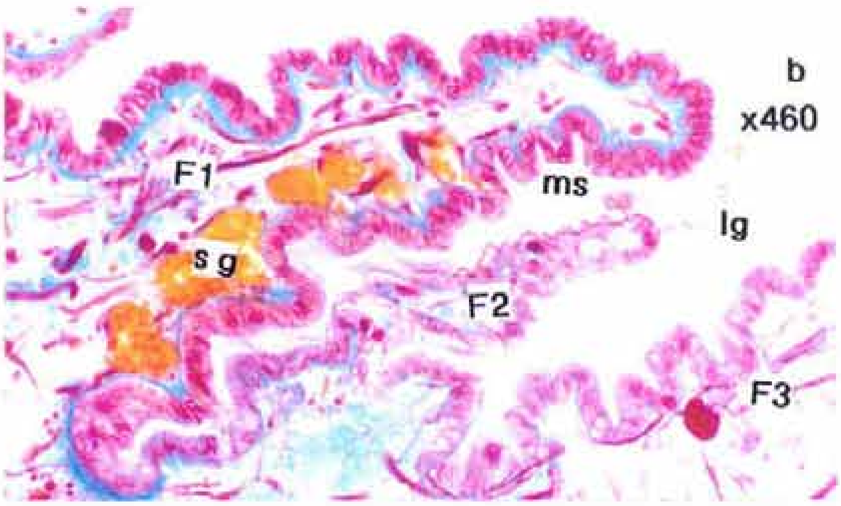
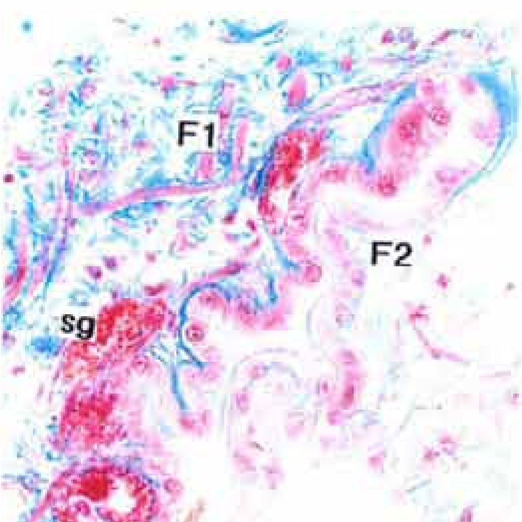
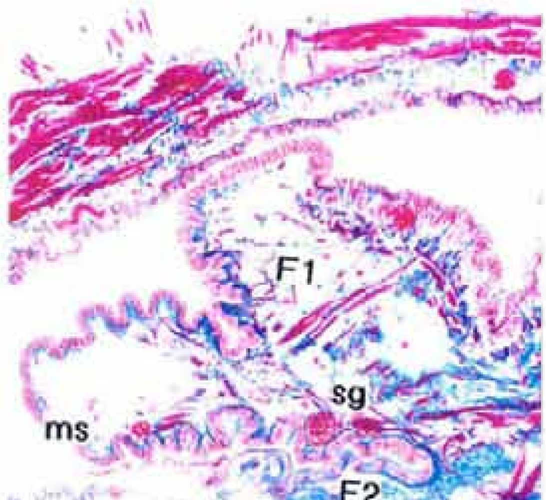


Figure5.173



c x460





5.12. VENEROIDEA (Continued....)

5.12.2 *Circe trigona.*

The External Mantle is joined to the medial surface of the Valve at the Pallial Line Junction (j in Figure 5.174. a). There are three Marginal Mantle Folds (F1., F2. and F3. in Figure 5.174. a, b, c and d). A bi-laminate secretion issues from Groove F1F2. and recurves around the terminus of FoldF1. to form part of the outer non-calcareous Shell Layer. The lateral lamina in the Groove (and therefore the medial lamina of the Shell Layer) is the more copious and stains blue colours with all stains used (ls in Figure 5.174. a and b). The more medial lamina in the Groove (ms in Figure 5.174. a and b), and therefore the more lateral on the Shell Layer, stains yellow with M.S.B. and A.B./M.S.B., amber with Mallorys and red with Azan.

There is a massive concentration of acid mucopolynaccharide secretory glands in the subepithelium of MedF2. and LatF3. (tt in Figure 5.174. a and d).

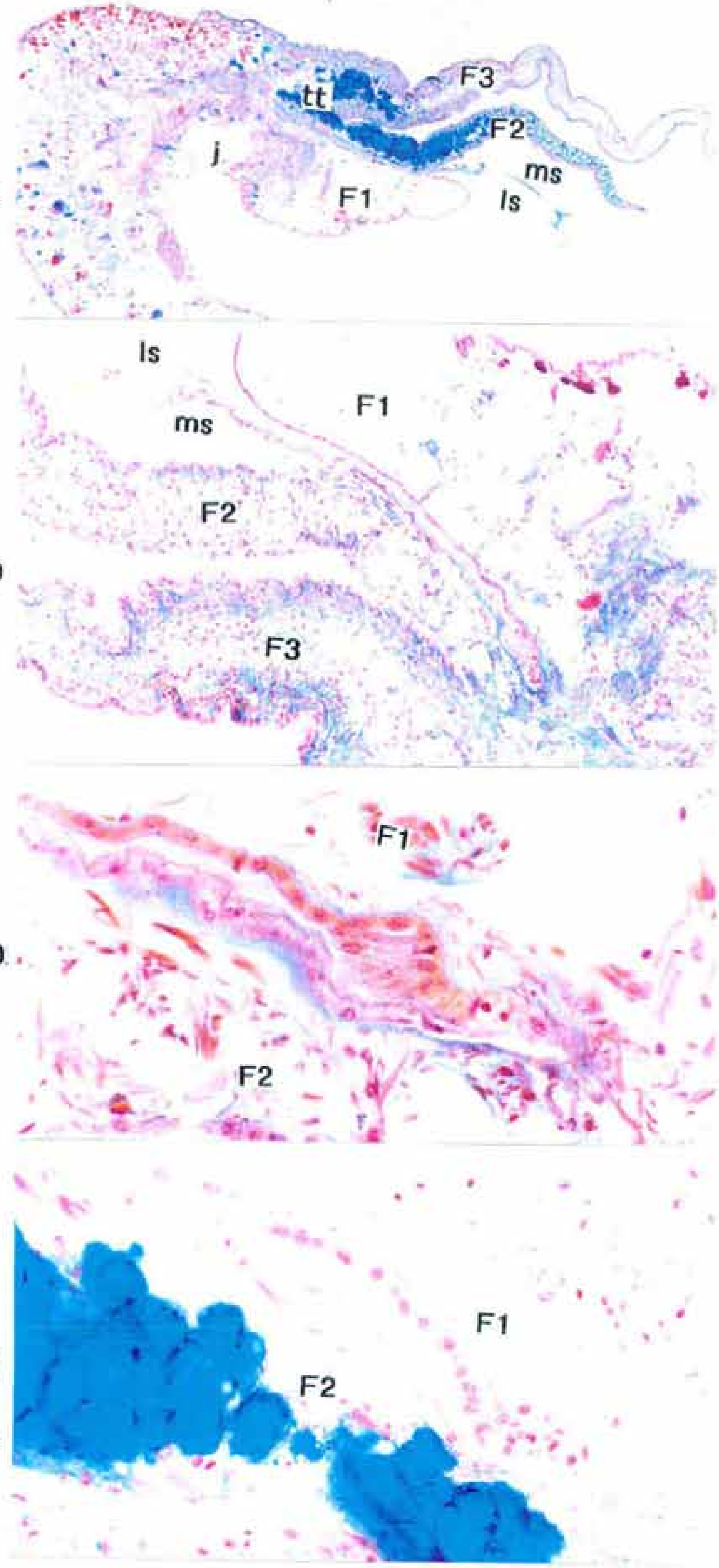
FIGURE 5.174. *Circe trigona* External Mantle and Mantle Margin.

a. L.M. x 38 A.B./M.S.B. External Mantle and Mantle Margin.

b. L.M. x 120 M.S.B. Mantle Margin.

c. L.M. x 460 Mallorys. Apical Groove F1F2.

d. L.M. x 390 A.B./M.S.B. Apical Groove F1F2.



a
x38

b
x120

c
x460

d
x390

Figure
5.174

5. VENEROIDEA.

5.12.3. *Gafrarium divaricatum.*

5.12.4. *Gafrarium tumidum.*

In these species the External Mantle is joined to the medial surface of the valve at the Pallial Line Junction (j in Figure 5.175.). There are four Marginal Mantle Folds. A bilaminate secretion issues from Groove F1F2.. The more medial lamina in the Groove, which becomes the more lateral in the Shell Layer, after recurving around Terminal F1, stains amber with Mallory's, yellow with A.B./M.S.B. and red with Azan. (ms in Figure 5.175. a, b and d). The more lateral in the Groove (and hence more medial in the Shell Layer stains purple with Mallorys and turquoise with A.B./M.S.B. (ls in Figure 5.175. a and b).

There are numerous large glands which stain turquoise with A.B./M.S.B. in the subepithelium beneath the second, third and fourth Marginal Mantle Folds, (tt in Figure 5.175).

FIGURE 5.175.

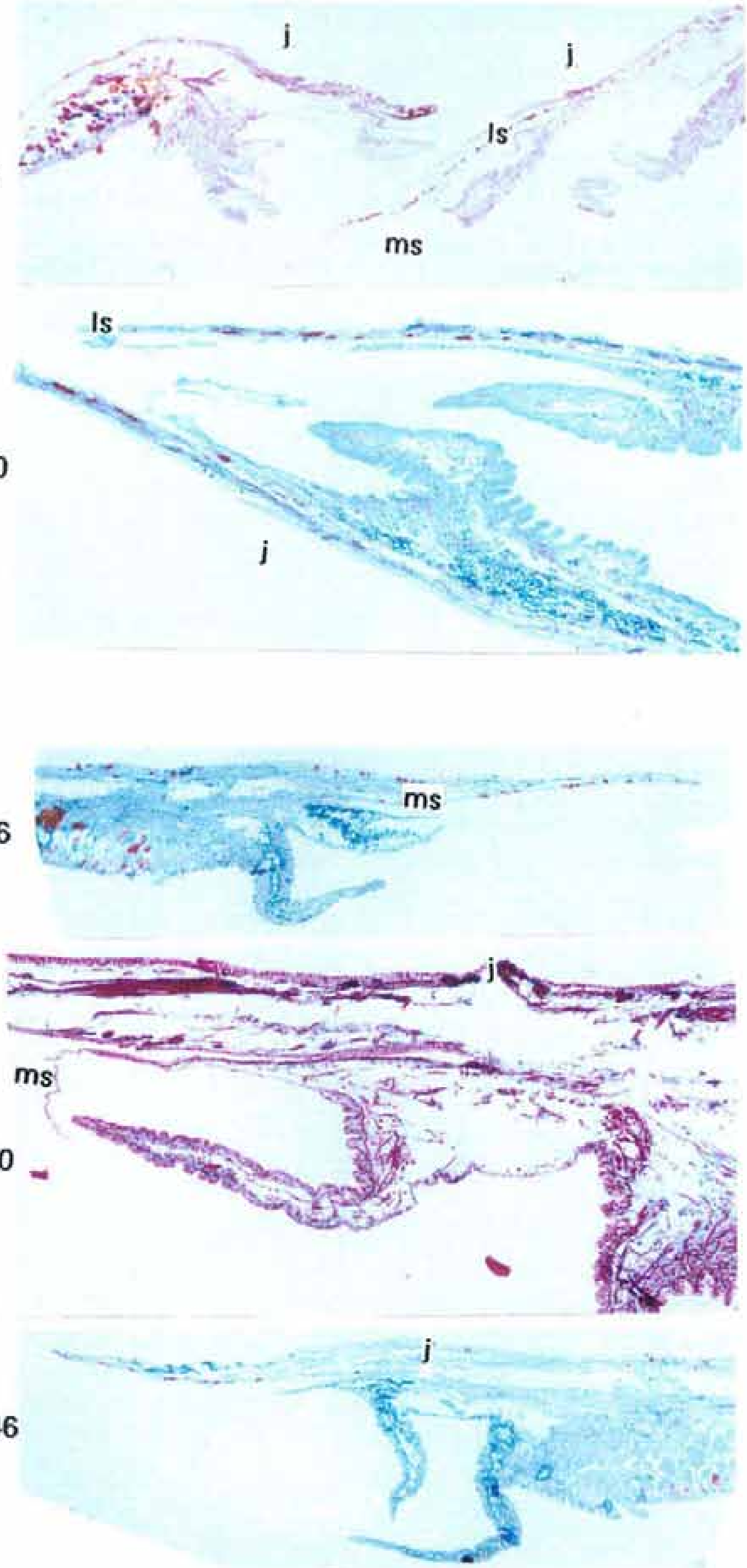
a. L.M. x 46 Mallorys Mantle Margins *Gafrarium divaricatum.*

b. L.M. x 120 A.B./M.S.B. Mantle Margins *Gafrarium divaricatum.*

c. L.M. x 46 A.B./M.S.B. Mantle Margin *Gafrarium timidum.*

d. L.M. x 120 Azan Mantle Margin *Gafrarium timidum.*

e. L.M. x 46 A.B./M.S.B. Mantle Margin *Gafrarium timidum.*



ax46

bx120

cx46

dx120

Figure
5.175

ex46

5.12.5. GAFRARIUM ?SP.

This species has a junction of External Mantle and medial surface of the valve at the pallial Line (j in Figure 5.176. a). There are what appear to be four Marginal Mantle Folds. Discharging into the Groove between the more lateral two apparent folds from the subepithelium are Granular Cytoplasm Secretory Glands which appear very similar to those on MedF1. of *Gafrarium durivcatum* (Figure 5.175. a, b, c and d), and *G. tumidum* and other Veneroidea (e.g., *Dosina juvenilis*(Figure 5.173. a, b, c and d)). Between what appear to be the second and third most lateral Marginal Folds a sheet of secreted material (sm in Figure 5.176. e) which stains amber with Mallory's, yellow with M.S.B. and red with Azan issues and recurves around the termini of the Marginal Mantle Folds lateral to it to become the deeply folded outer non-calcareous Shell Layer (OSL in Figure 5.176. a, b, c and d). The method of secretion of the precursor of the unilaminate outer non-calcareous Shell Layer is very similar to the structures which have the same function in *Gafrarium divaricatum* and *G. tumidum* (Figure 5.175. a, b, c, d and e).

There are three calcareous Shell Layers medial to the outer non-calcarous Shell Layer. The most medial of these (4SL in Figure 5.176. a, b, c and d), is coextensive with the Pallial Space (ps in Figure 5.176. a, b and c). The growing surfaces of the other two Shell Layers (2SL and 3SL in Figure 5.176. a, b, c and d) are enclosed in the Extra-pallial Space (eps in Figure 5.176. a, b, c and d).

FIGURE 5.176. *Gafrarium ?sp.*

a. LM x 150 M.S.B. Four Shell Layers after decalcification.

b. LM x 230 Mallory's As for a above.

c LM x 770 Mallory's Higher magnification of b above.

d LM x 230 Azan Mantle Margin and secretion of outer Shell Layer.

e LM x 460 Azan Four Shell Layers.

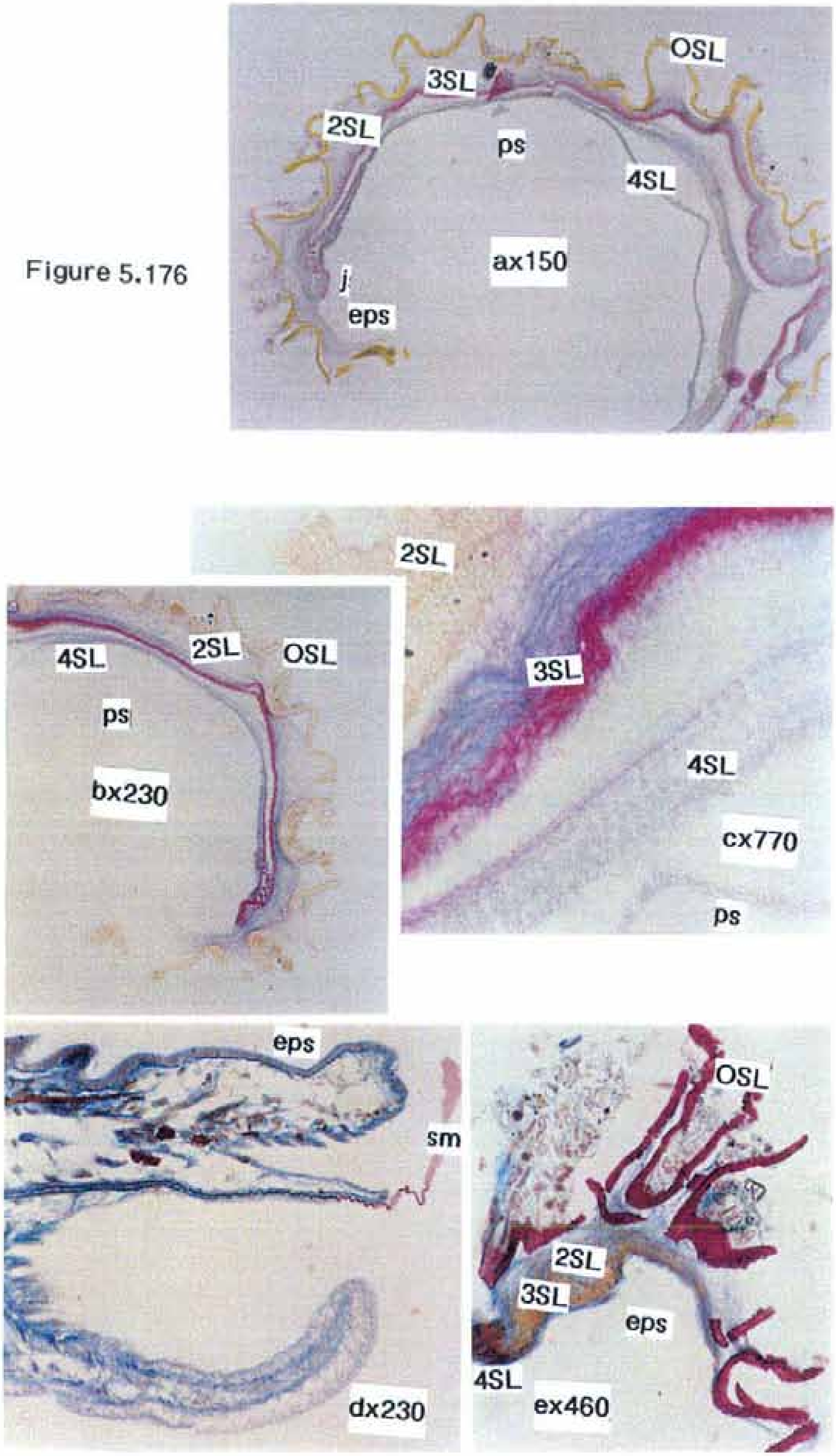

OSL
3SL
2SL
ps
4SL
ax150
j
eps
2SL
4SL
2SL
OSL
ps
bx230
3SL
4SL
cx770
ps
eps
sm
dx230
OSL
2SL
3SL
eps
4SL
ex460

Figure 5.176

5.12. VENEROIDEA.

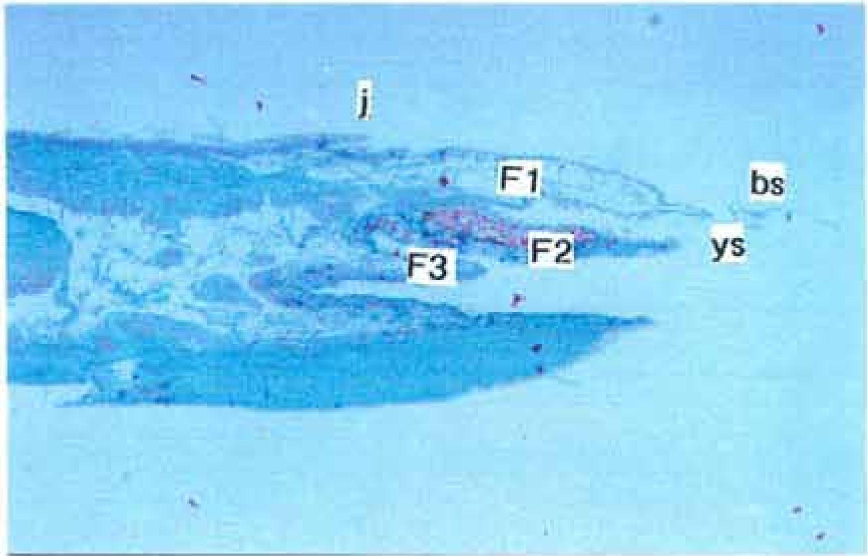
5.12.6. *TAPES SP.*

The external Mantle joins the medial surface of the valve at the Pallial Line Junction (j in Figure 5.177. a).This species has three Marginal Mantle Folds (F1. F2 and F3 in Figure 5.177. a andb) and produces a bilaminate secreted sheet from Groove F1F2. The more medial of the two sheets of secreted material in the Groove F1F2. stains yellow with A.B./M.S.B. (ys in Figure 5.177 a). and the more lateral stains blue with this stain. (bs in Figure 5.177 a). In this species F2. appears to act as a metabolic store (F2 in Figure 5.177 a and b).

FIGURE 5.177. *Tapes sp.* Mantle Margin

a. L.M. x 46 A.B./M.S.B. Folds F1., F2. and F3..

b. L.M. x 230 A.B./M.S.B. Secretory structures of Groove F1F2. and stored metabolites of F2..



ax46

bx230

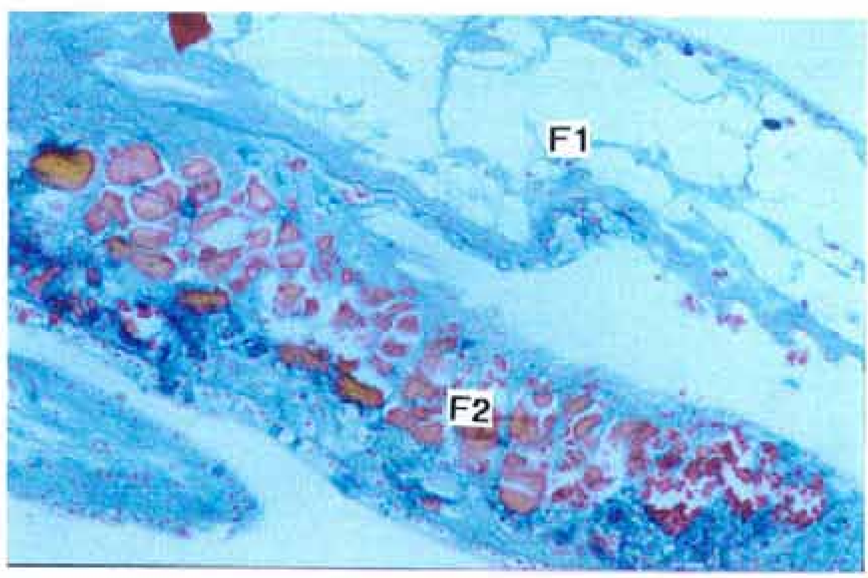


Figure 5.177

5.12. VENEROIDEA.

5.12.7. *PLACAMEN CALOPHYLLUM.*

5.12.8. *GLOBIVENUS EMBRITHES.*

Placament calophyllum has a junction of External Mantle to the medial surface of the Valve at the Pallial Line (j in Figure 5.178. a).

The Mantle Margin has only two Marginal Mantle Folds, (F1. and F2. in Figure 5.178. a). In this respect it resembles the *Tellinacea*. The apical region of Groove F1F2. also closely resembles the *Tellinacea* (Figure 5.172.), and produces a sheet of secreted material which issues from the Groove and becomes the outer non-calcified Shell Layer. It stains yellow with A.B./M.S.B.

In *Glolenvinus embrithes* the External Mantle joins the medial surface of the Valve at the Pallial Line Junction (j in Figure 5.178. c).

There are three Marginal Mantle Folds, (F1., F2. and F3. in Figure 5.177. c). A sheet of secreted material which stains yellow with A.B./M.S.B. is formed in Groove F1F2.

There are Granular Cytoplasm Secretory Glands in the subepithelium of MedF1. (gc in Figure 5.178. c). Trabecular Turquoise Glands occur in subepithelial LatF2., LatF3., and in a Glandular Structure on MedF3. (tt in Figure 5.178. c).

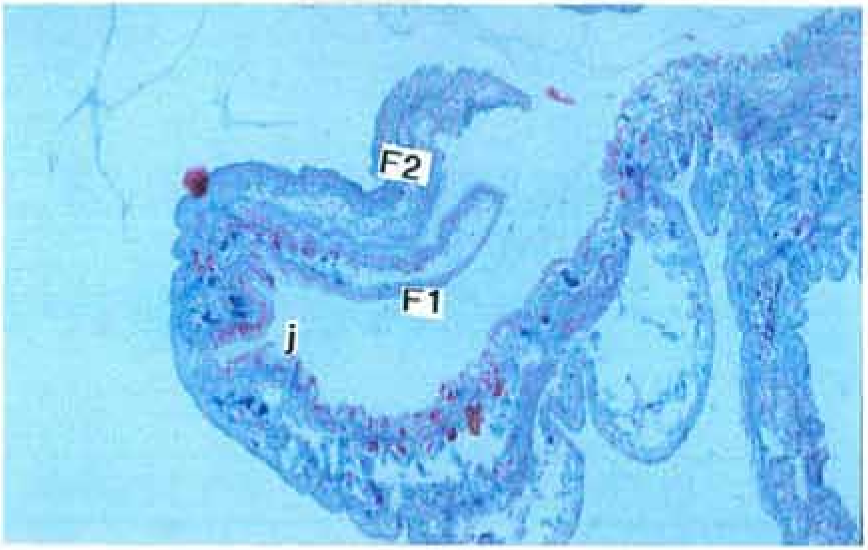
FIGURE 5.178. External Mantles and Mantle Margins of *Placamen calophyllum* and *Globivenus embrithes.*

a. L.M. x 120 A.B./M.S.B. *Placamen calophyllum* - Mantle Margin.

b. L.M. x 1200 A.B./M.S.B. *Placamen calophyllum* - Mantle Margin.

c. L.M. x 45 A.B./M.S.B. *Globivenus embrithes* - Mantle Margin.

ax120



bx1200

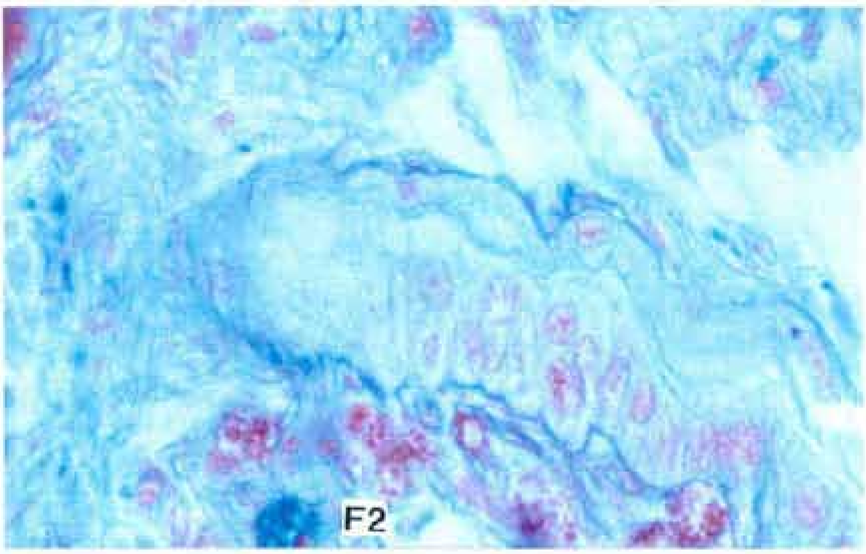
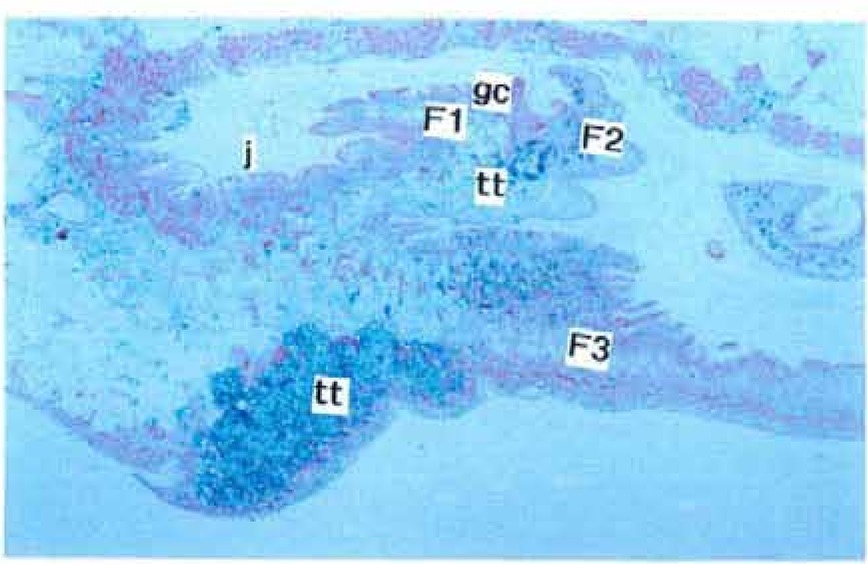


Figure 5.178

cx45



5.13. SOLENOIDEA.

5.13.1. *SOLEN VAGINA.*

5.13.2. *SOLEN GRANDIS.*

The Mantle Margins and Shell Layers of these species are very similar under L.M. and will be described together.

The mantles in this Superfamily are fused. On either side of the protrusion of tissue at the point of fusion are two folds and thus two grooves. On the lateral surface of the more lateral of these two folds is the Mantle Edge Gland, (meg in Figure 5.179. a) so this Fold can be termed F1. (Fl in Figure 5.179. a). The apex of the Groove immediately lateral to the protrusion at the site of fusion, however is similar structurally and histologically to Apical Groove F1F2. in other heterodont Superfamilies. If it is truly analagous to this feature it should be termed apical Groove F1F2., (aF1F2. in Figure 5.179. b), and the Fold lateral to it would then be Ancillary Fold F1., (AF1 in Figure 5.179. a). This is the terminology used here.

The Outer Lamina of the trilaminate outer non-calcareous layer (ol in Figure 5.179. a and c), is a thin sheet of dense material produced by Apical Groove F1F2. (a F1F2. in Figure 5.179. b).

The Inner Lamina (il in Figure 5.179. a and c) is produced by Groove F1. Ancillary F1. (g in Figure 5.179. a). Thus necessarily the comparatively thick middle lamina of the outer non-calcareous Shell Layer (ml in Figure 5.179. c) must be produced by Med Ancillary F1..

FIGURE 5.179. *Solen vagina* and *Solen Grandis* - Mantle Margin and Outer Shell Layer.

a. L.M. x 46 A.B./M.S.B. *Solen vagina* - Mantle Margins.

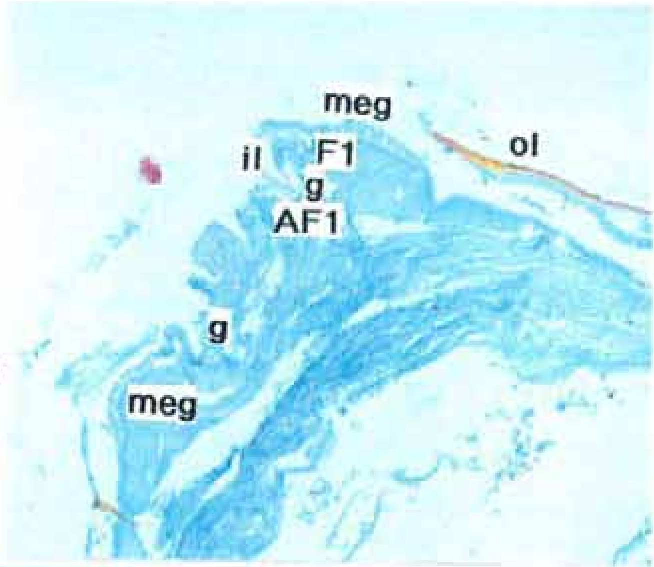
b. L.M. x 1200 A.B./M.S.B. *Solen vagina* - Fusion of Mantles.

c. L.M. x 1200 Steedmans *Solen Grandis* - Outer Shell Layer.

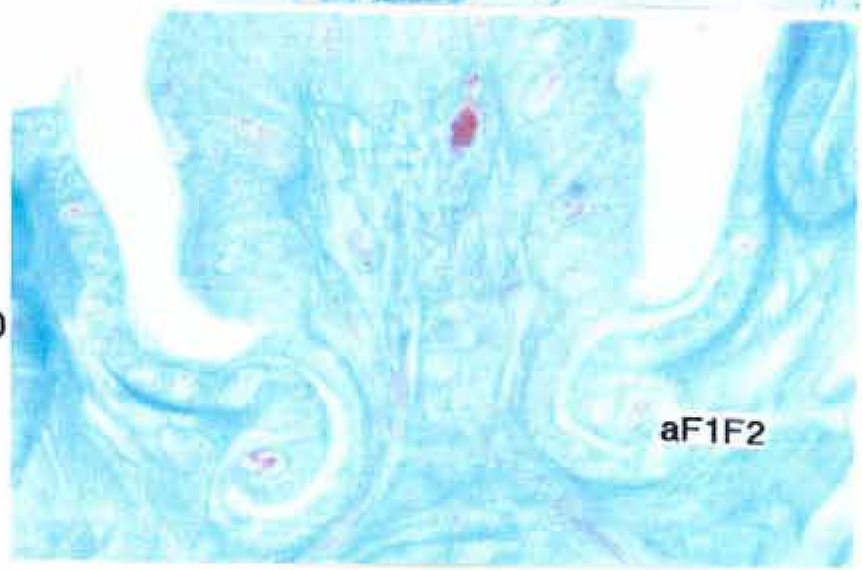
Figure

5.179

ax46



bx1200



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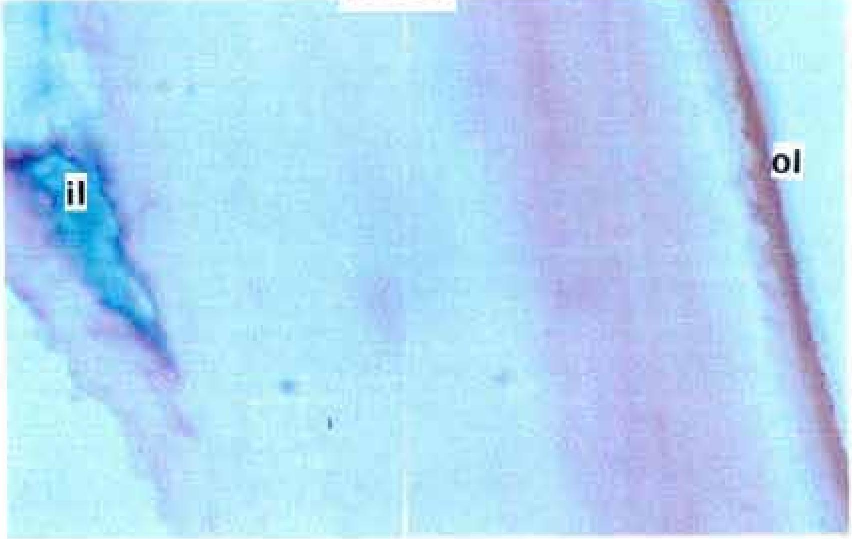


5.14. CARDITOIDEA.

5.14.1. *CARDITA VARIEGATA.*

The External Mantle is joined to the medial surface of the Valve at the Pallial Line.

The Mantle Margin has three Folds (F1., F2. and F3. in Figure 5.180. a and b).

A bilaminate sheet which becomes the outer non-calcareous Shell Layer is produced by the Mantle Margin.

A secretion which stains blue with Mallory's and turquoise with A.B./M.S.B. is produced in Groove F1F2., (il in Figure 5.180. a and b). A secreted sheet which stains amber with Mallory's and yellow with M.S.B. is produced in Groove F2.F3., (ol in Figure 5.180. a and b).

On MedF2. and LatF3. there are subepithelial Glands which stain yellow with A.B./M.S.B. (y in Figure 5.180. b).

LatF3. has Granular Cytoplasm Secretory Glands in the subepithelium which stain magenta with A.B./M.S.B. (gc in Figure 5.180. b).

FIGURE 5.180 *Cardita variegata.*

a. L.M. x 230 Mallorys. Mantle Margin.

b. L.M. x 230 A.B./M.S.B. Mantle Margin.

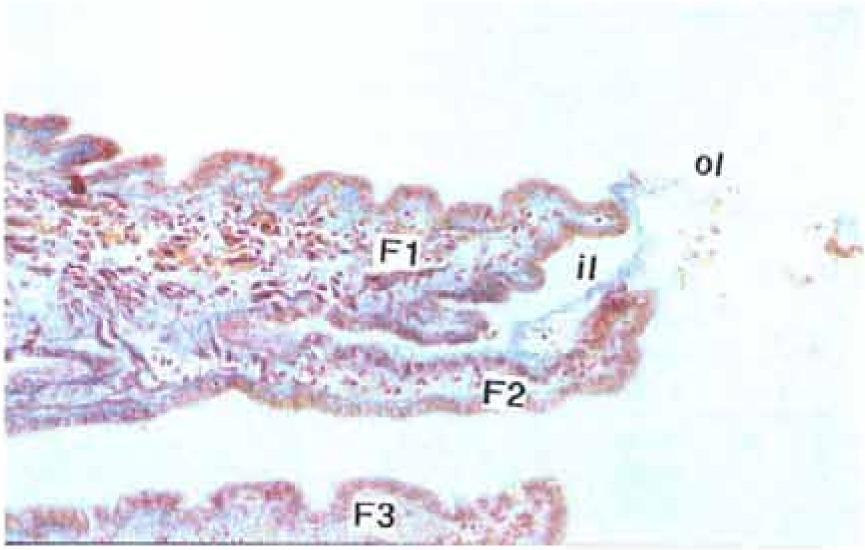
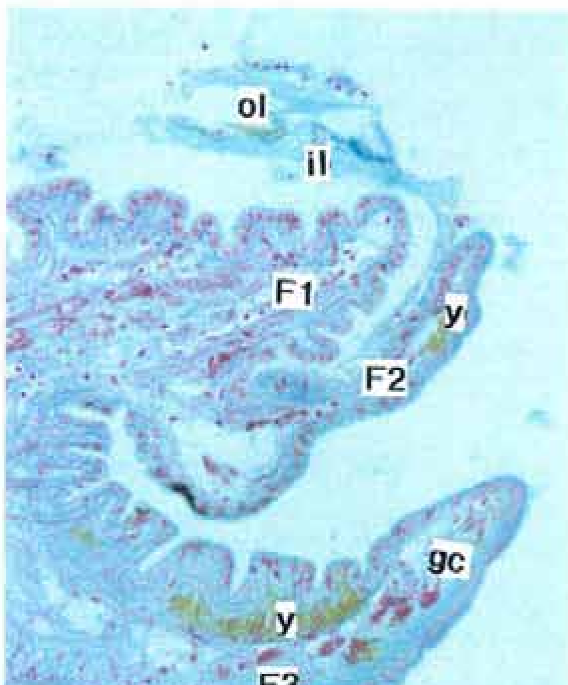


Figure 5.180



5.15 MACTROIDEA

5.15.1 *MACTRA ABBREVIATA.*

The Mantle Margin consists of Folds F1. and F2. (F1 and F2 in Figure 5.181, a). Apical Groove F1F2. produces the outer lamina of a bilaminate outer non-calcareous Shell Layer which stains bright yellow wtih A.B./M.S.B. (ol in Figure 5.181, a), amber with Mallorys and red with Azan.

The inner lamina of the bilaminate outer shell layer stains turquoise with A.B./M.S.B. and appears to be secreted by distal MedF1. and perhaps Terminal F1. (il in Figure 5.181 a).

The organic matrices of the outer calcareous Shell Layer are secreted by LatF1. between Terminal F1. and the Pallial Line and stain similarly to the Inner Lamina of the outer non-calcareous Shell Layer, (om in Figure 5.181, a).

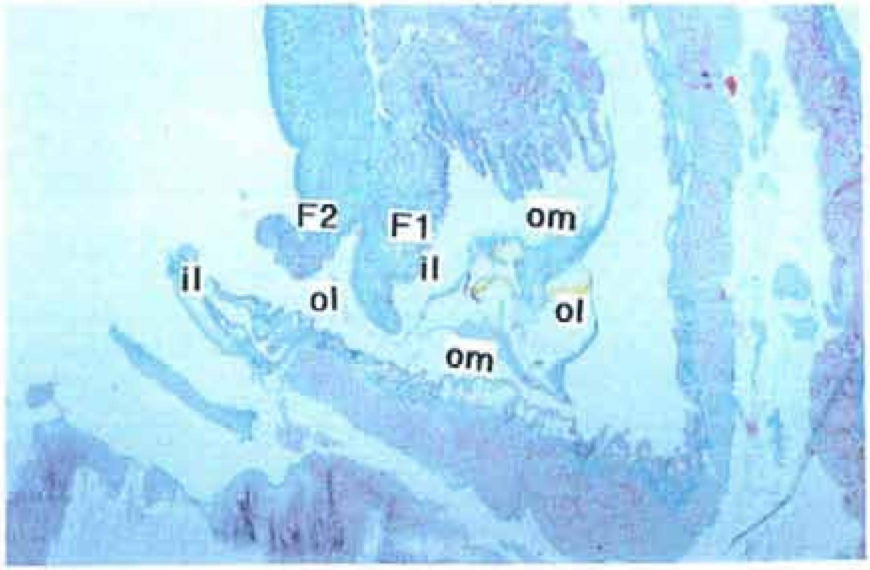
5.15.2. *Mactra dissimilis*

This species has a bilaminate outer non-calcareous Shell Layer the outer lamina of which is secreted by Groove F1F2., (F1F2 in Figure 5.181, b), and stains red with Azan (ol Figure 5.181, b), yellow with M.S.B. and A.B./M.S.B. The inner lamina of the outer non-calcareous Shell Layer stains blue with Azan and appears to be secreted by LatF1. (il Figure 5.181, b).

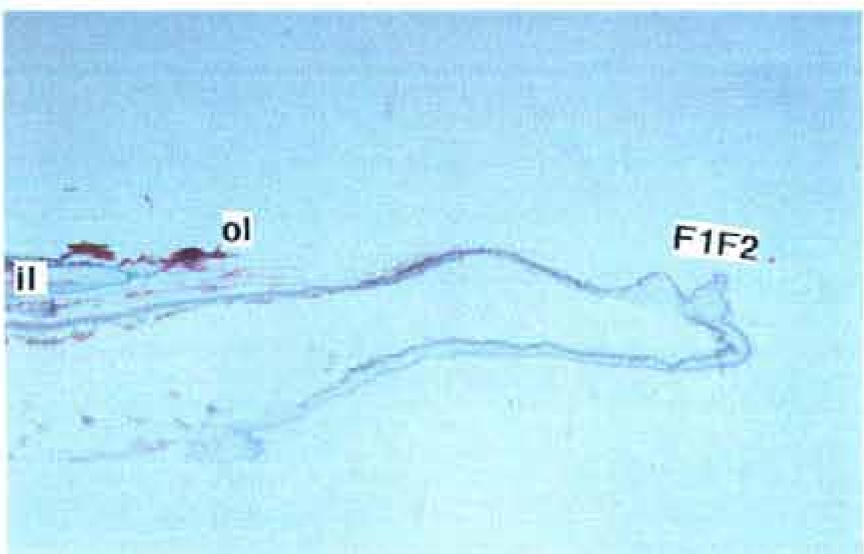
FIGURE 5.181. *Mactra obesa* and *Mactra dissimilis.* - External Mantles, Mantle Margins and decalcified Shell Layers.

a. L.M. x 46 A.B./M.S.B. *Mactra obesa.*

b. L.M. x 46 Azan *Mactra dissimilis.*



ax46



bx46

Figure 5.181

5.16. CORBICULOIDEA.

5.16.1. *GELOINA COAXANS.*

In Figure 5.182, L is lateral and M medial.

The bilaminate outer non-calcareous Shell Layer has an outer lamina which stains pink-red with all stains used (ol in Figure 5.182, a). This lamina is produced by the proximal half of Groove F1F2, originating at Apical Groove F1F2., (aF1F2 in Figure 5.182, b).

The distal part of MedF1. and Terminal F1. produced the dense inner lamina of the bilaminate outer non-calcareous Shell Layer (il in Figure 5.182, a).

The outer epithelium of LatF1. (oe in Figure 5.182, c), appears to produce two different types of organic matrix alternately (1 and 2 in Figure 5.182, c).

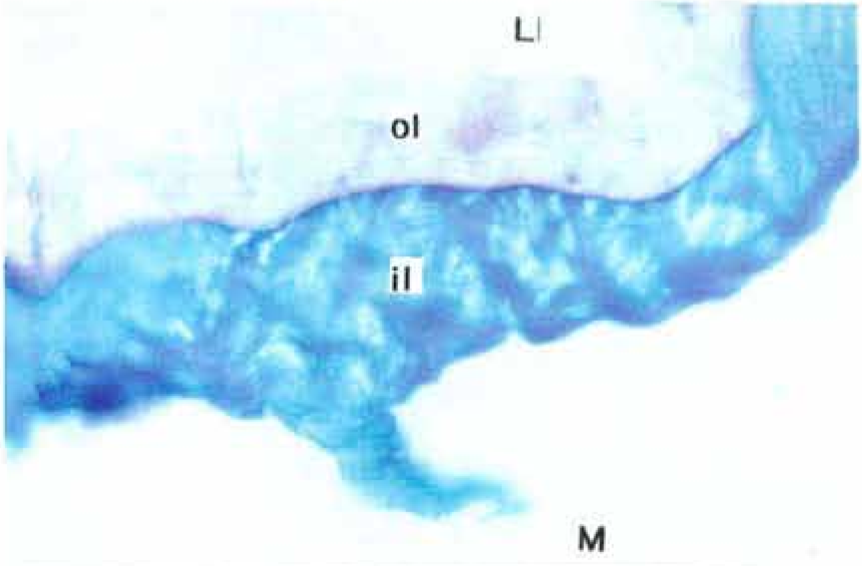
FIGURE 5.182. *Geloina coaxans* - Shell Layers and Mantle Margin.

a. L.M. x 1200 Steedmans Outer non-calcareous Shell Layer.

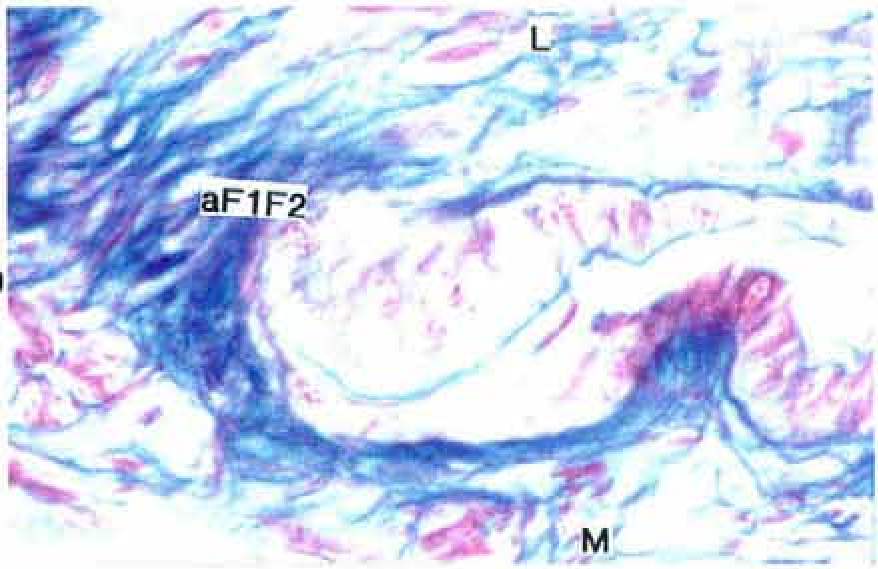
b. L.M. x 1200 Steedmans apical Groove F1F2..

c. L.M. x 1200 Steedmans LatF1. and organic matrices of outer calcareous Shell Layer.

ax1200



bx1200



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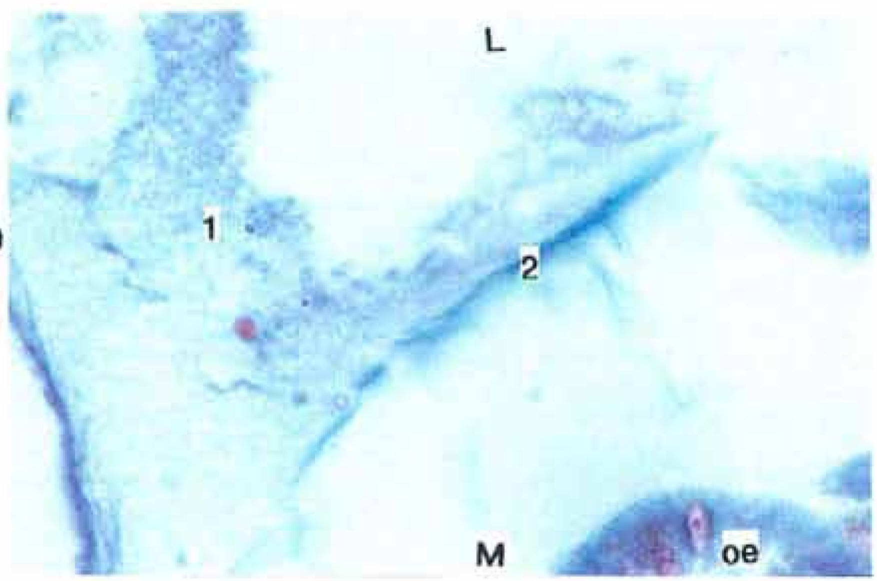


Figure
5.182

5.17. GASTROCHAENOIDEA.

5.17.1. *GASTROCHAENA CUNEIFORMIS*

In Figure 5.183 L is lateral, M. medial.

There appear to be four Marginal Mantle Folds in this species (F1., F2., F3. and F4. in Figure 5.183. a and d). The outer lamina of the trilaminate outer non-calcareous Shell Layer is produced at the apex of the most medial Marginal Mantle Groove at 0 in Figure 5.183. a and d. The middle lamina which stains turquoise with A.B./M.S.B. is secreted by the second most medial Marginal Mantle Groove between Folds F2. and F3. in Figure 5.183. a and d. The medial of the three laminae of the trilaminate outer non-calcareous Shell Layer is secreted by the outer Marginal Mantle Groove (gF1F2. in Figure 5.183. a and d).

The Outer of the two calcareous Shell Layers (ocl in Figure 5.183. b and c) is generated by the secretory structures of the medial lining of the Extrapallial Space (eps in Figure 5.183. a). The inner calcareous Shell Layer is generated by the Pallial Mantle (icl and pm respectively in Figure 5.182. c).

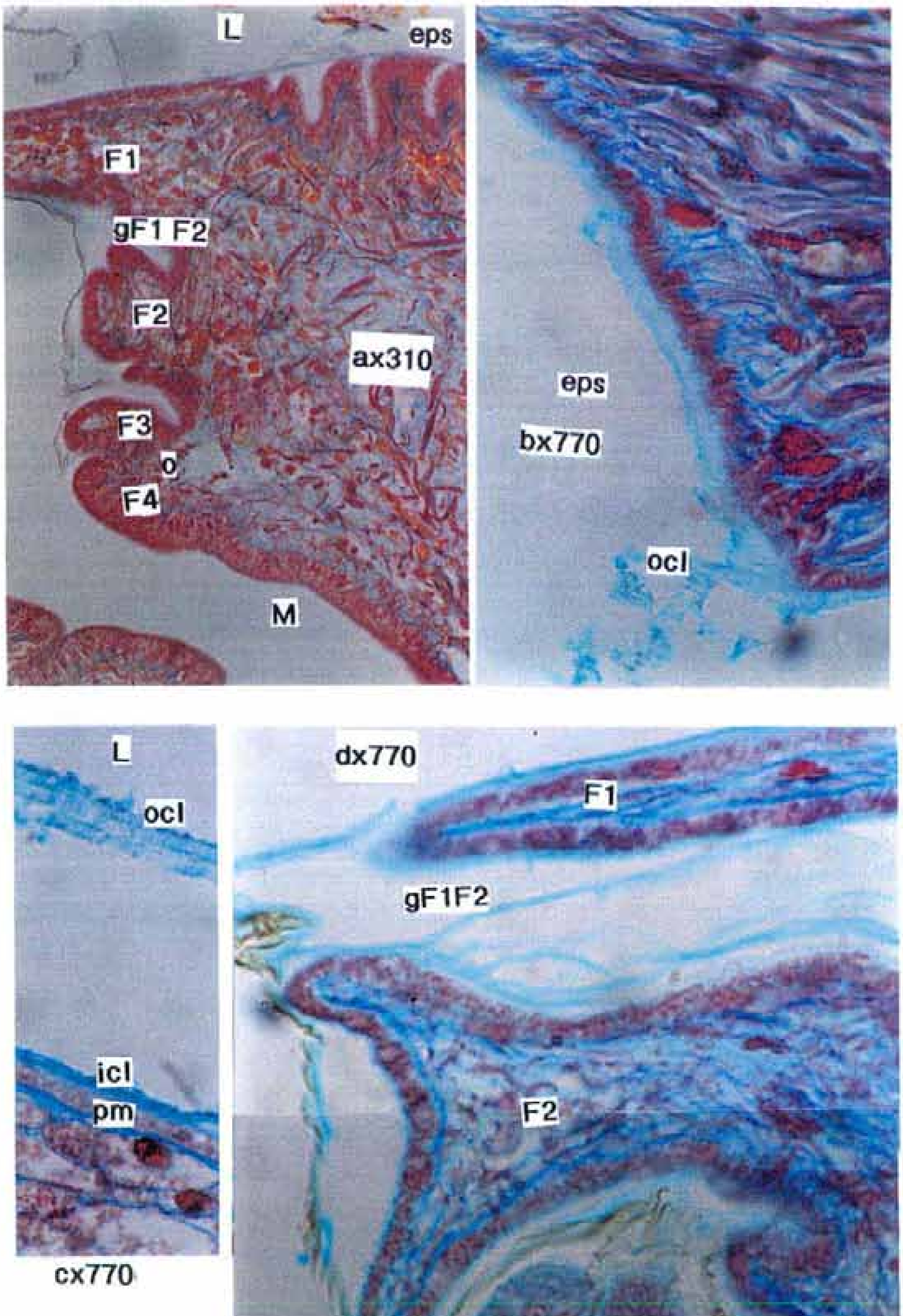
FIGURE 5.183. *Gastrochaena cuneiformis* - L.M. Shell Layers. External Mantle and Mantle Margin.

a. x 310 Mallory's. Mantle Margin and outer non-calcareous Shell Layer secretion.

b. x 770 A.B./M.S.B. Medial surface of Extra-pallial Space and secretion of outer calcareous Shell Layer.

c. x 770 A.B./M.S.B. Pallial Mantle and secretion of Inner calcareous Shell Layer.

d. x 770 A.B./S.B. Mantle Margin and secretion of trilaminate outer non-calcareous Shell Layer.


L
eps
F1
gF1 F2
F2
ax310
F3
o
F4
M
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cx770
dx770
F1
gF1F2
F2
F4
F3
M
o

Figure5.183

CHAPTER 6. DISCUSSION

6.1. THE ORIGIN OF THE SHELL LAYER ORGANIC MATRICES IN PEARL OYSTERS AND OTHER BIVALVES.

6.1.18.7. The Sources of the Organic Matrix Precursors of the Outer Nacreous Layer.

6.1.18.8. The Sources of the Organic Matrix Precursors of the Prismatic Layer.

6.1.THE ORIGIN OF THE SHELL LAYER ORGANIC MATRICES IN PEARL OYSTERS AND OTHER BIVALVES.

6.1.1.Scleroproteins and their Production in Biological Systems.

Some of the chemical constituents of the Shell Layer Organic Matrices of pearl oysters are scleroproteins and, as with other scleroproteins, are relatively very insoluble (Waite, 1983). This insolubility is achieved by a series of chemical steps during their synthesis which necessarily take place extracellularly - they involve cytotoxic intermediaries such as aldehydes, semiquinones and activated oxygen. This in turn requires that the precursors of the scleroproteins be stored in separate compartments e.g., unicellular secretory glands. Waite (1983) points out that there are four compartmentalised precursors in the formation of the scleroprotein fibrin, and essentially similar steps are required for the formation of the scleroproteins collagen, elastin, keratin, resilin and silk fibroin.

6.1.2.Scleroproteins in Bivalves.

As Waite (1983) considers it likely that the formation of the scleroproteins of molluscan Shell Layers involves "quinone tanning" a very similar situation must be required for their production. A model suggesting four precursors in separate compartments for this process to produce molluscan "periostracum" and byssis is given (Waite, 1983). Discrete unicellular secretory glands which take part in the production of "periostracum" have been identified in Groove F1F2, in marine bivalves' Marginal Mantle (Bubel, 1973). Thus essential morphological features for production of Shell Layer Organic Matrix scleroproteins are secretory glands which discharge the precursors into an extracellular reaction chamber.

6.1.3.Scleroproteins and Bivalve Shell Layers.

Bivalve shells are said to be covered externally by a "periostracum", (Watabe, 1984), with between one and three calcified Shell Layers medial to it (Taylor, 1973). The "periostracum" is discussed in detail by Watabe, (1984) and its formation in bivalves described by Saleuddin and Petit (1983). Waller (1978) suggests that the only constant of all bivalve Mantle Margins is a "periostracum" which is produced in the "periostracal" groove i.e., between the outer two Marginal Mantle Folds. While the accuracy of this description and the usefullness of the concept of a periostracum in pearl oysters and other bivalves will be discussed below, for the moment following the above authors, it can be defined as a more or less non-mineralised scleroproteinaceous Shell Layer produced in Groove F1F2, which recurves around the distal extremity of F1, and covers the lateral surface of the valve.

Taylor (1973) shows the shell of the *Pteriacea* as composed of an outer calcitic Prismatic Layer, lateral to two Nacreous Layers - the Outer Nacreous Layer and the Inner Nacreous Layer.

calcite Simple Prismatic Layer - lateral
aragonite Outer Nacreous Layer - middle
arogonite Inner Nacreous Layer - medial

Other Superfamiles, involved in this study and the Shell Layers assigned them by Taylor (1973), and described by Watabe (1983), with, where seen in this work an outer non-calcareous layer added, are the following:

Unionoidea
outer non-calcareous Shell Layer
aragonite Simple Prismatic Layer - lateral
aragonite Outer Nacreous Layer - middle
arogonite Inner Nacreous Layer - medial

Nuculoidea
aragonite Composite Prism Layer - lateral
aragonite Outer Nacreous Layer - middle
aragonite Inner Nacreous Layer - medial

Mytiloidea

calcite Mytilid Prisms - or trilaminate outer non-calcareous Shell Layers - lateral

aragonite Outer Nacreous Layer - middle

aragonite Inner Nacreous Layer - medial

Pinnoidea

calcite Prismatic Layer - lateral

aragonite Outer Nacreous Layer - middle

aragonite Inner Nacreous Layer - medial

Ostreoidea

calcite Prismatic Layer - lateral

calcite Foliated Structure - medial

Anomioidea and Pectinoidea

calcite Foliated Structure -lateral

aragonite or calcite Crossed Lamellar Structure - medial

Limopsoidea and Arcoidea

trilaminate non-calcareous outer layer -lateral

aragonite or calcite Crossed Lamellar Structure - middle

aragonite or calcite Complex Crossed Lamellar Structure - medial

Tellinoidea and some *Veneroidea*

unilaminate non-calcareous layer

aragonite Composite Prismatic Layer -lateral

aragonite or calcite Crossed Lamellar Layer -middle

aragonite or calcite Complex Crossed Lamellar Layer - medial

Veneroidea

unilaminate non-calcareous layer
fromaragonite Composite Prismatic Layer -lateral
aragonite or calcite Crossed Lamellar Layer -middle
aragonite or calcite Complex Crossed Lamellar Layer - medial
to unilaminate non-calcareous layer.
aragonite Homogenous Structure -

Corbiculoidea, Gastrochaenoidea, Carditoidea, and Mactroidea

bilaminate non-calcareous layer
aragonite or calcite Crossed Lamellar Structure-lateral
aragonite or calcite Complex Crossed Lamellar Structure - medial

In summary the calcareous Shell Layers of the animals used in this study according to Taylor (1973), are the following:
Simple Prismatic Layer, calcite.
Simple Prismatic Layer, aragonite.
Nacreous Layer, aragonite.
Composite Prismatic Layer, aragonite.
Mytilid Prismatic Layer, calcite.
Foliated Structure, calcite.
Crossed Lamellar Structure, aragonite or calcite.
Complex Crossed Lamellar Structure, aragonite or calcite.
Homogenous Structure, aragonite.
To these nine calcareous Shell Layers, for the purposes of this discussion, surface "periostracum" or non-calcareous outer Shell Layer, (which may be either uni-, bi- or trilaminate), must be added.
The Shell Layers of most interest in this work are the Nacreous Layers and Prismatic Layer of pearl oysters. Origin, ultrastructure and staining affinity of the other Shell Layers in the other bivalves given above are included here only to the extent that their study sheds light on the origin of the precursors or mode of

synthesis of the pearl oyster Shell Layers.

Weiner et al., (1983), have shown that the process of crystallisation of the inorganic phase of Shell Layers is controlled by the Shell Organic Matrices - including the determination of the direction of the crystallographic axes. It has been shown here that the geometric shape of Free and Partly Bound Nacre Tiles is almost invariably the same at any one locality on the medial surface of any Nacreous Layer studied. Further, the orientation of the axes of the geometric shapes of the Free and Partly -bound Nacre Tiles to the direction of the associated Nacre Sheet Edge is standardised at any location. This indicates a very close control over the micromorphololology of Nacreous Layers by their Organic Matrices. The exactness of the replication from specimen to specimen, in any one species of bivalve of the micromorphology of the different Shell Layers indicates a similar degree of control to that of Nacreous Organic Matrices by the Organic Matrices of these other Shell Layers.

It is not clear from Waite (1983) (nor from any reports sighted), whether most, or all, of the different components of Shell Organic Matrices are thought to consist largely of scleroproteins. In this study the Shell Organic Matrices of members of different Superfamilies were found to have a wide variety of responses to solubilisation by acid hydrolysis. Never-the-less, all Shell Layers tested had Organic Matrices which, given the fineness of structure of such features, were relatively very resistant to solubility in an acid solution and hence at least part of their protein component can be regarded as sclerified.

6.1.4.Scleroproteins and Pearl Shell Layers.

As above, (Taylor, 1973) the members of the *Pterioidea* have a shell consisting of a Prismatic Layer lateral to an Outer Nacreous Layer and an Inner Nacreous Layer.

Acid decalcification and subsequent sectioning and staining for light microscopy revealed a variety of different scleroproteins in the Shell Layers of pearl shells which was augmented by S.E.M study of the shells of the same species.

6.1.4 (a). The Scleroproteins of the Prismatic Layer of Pearl Shells.

As shown in Results, Figures 5.40, 5.97-5.99 the acid decalcified Prismatic Layers of the shells of the members of the *Pterioidea* studied consist of the following components which are held to be discrete scleroprotein entities on the bases of morphology, resistance to acid solubility and/or reaction to staining procedures:

1 - Outer Fibrous Sheath,
2 - Outer Prismatic End Plate.
3 - Inner Lateral Structure,
4 - Transverse Parallel Side Walls,
5 - Intraprismatic Organic Matrix
6 - Inner Prismatic End Plate,
7 - Inner Fibrous Sheath
In addition,
8 - Reteform Interprismatic Organic Matrix and
9 - Linear Pattern Organic Matrix
were seen with S.E.M..

Of these, the Outer Prismatic End Plate and the Inner Lateral Structure appear to be secreted by tissues at the same locus and where one stains aberrantly the other invariably, in the animals studied here, does the same. It is therefore likely that they have at least some secreted precursors in common.

Similarly Intraprismatic Organic Matrix appears indistinguishable from Reteform Inter-prismatic Organic Matrix under S.E.M. and both of these structures appear to be secreted at the same locus as Linear Pattern Organic Matrix. As above these latter two are invariably dissolved during the proceedure of acid decalcification employed in this study and the former only survives this procedure as a wispy deformed remnant. It may well be that it is exactly the same material as the Reteform Inter-prismatic Organic Matrix but survives due to relatively decreased acid strength between the calcite tablets during decalcification.

Thus while it is considered likely that there are nine chemically distinct scleroproteins in the Prismatic Layer of any member of the *Pterioidea* studied it is possible that "2" and "3" above are chemically very similar but morphologically distinct and that the same is true of "5", "8" and "9" above. If it only requires two precursors to react to form each scleroprotein species and one precursor is common to each of them, this still would require a minimum of seven discrete secretory entities to produce a pearl shell Prismatic Layer. If in fact all nine entities listed above are discrete scleroproteins and if as Waite (1983) suggests, each scleroprotein requires four different precursors and if none are used in the synthesis of more than one scleroprotein then the production of pearl shell Prismatic Layer might require thirty six chemically discrete secreted precursors. As seen here, a type of acidmucopolysaccharide unicellular secretory gland is present in the Mantle Margin of pearl oysters in every tissue of LatF2., Med F2. and LatF3., and unique to these locations. It would therefore appear likely that at least one precursor is common to all Prismatic Layer scleroproteins of any pearl oyster, (see Chapter 6.6).

1.4 (b) Scleroproteins of the Nacreous Layers of Pearl Shells.

Nakahara (1991) states that bivalve Nacreous Layers consist of Nacre Tiles individually enclosed by envelopes of one Organic Matrix in Nacre Sheets separated by layers of a different Organic Matrix. It appears more likely, (Figures 5.98, a and 5.135, a), that the Nacre Sheets are separated medio-laterally by sheets of one Organic Matrix scleroprotein, and separated from other Nacre Tiles in the same Nacre Sheet by side walls of a different, and more easily acid-hydrolysed scleroprotein. On both Nakahara's and others' descriptions and results seen in this study it is very likely that there is an Organic Matrix entity within the Nacre Tiles. The explanation of origin of free Nacre Tiles given here (Chapter 6.5), suggests that the Organic Matrices of these are identical to those of the adjacent Nacre Sheet so that no distinct intra-Nacre Tile Organic Matrix is postulated here. On either of these interpretations there are considered to be two or three different scleroproteins in a Nacreous Layer.

Since two physically different Nacreous Layers are distinguished in pearl oysters the constituent Organic Matrices are in all probability also different.

Again, as with the Prismatic Layer, it appears that there is one type of acid-mucopolysaccharide secretory gland common to every tissue of the External Mantle and Mantle Margin Fold F1, in every species of pearl oyster studied. It is thus likely that the Organic Matrices of the Nacreous Layers of any one pearl oyster share at least one chemical precursor.

6.1.5.Essential Bases of Synthesis of Shell Organic Matrix Scleroproteins.

From all the above it is clear that there are certain requirements for the production by the External Mantle and Mantle Margin of bivalves of the scleroproteins of their Shell Layers. These are:

1.That the scleroprotein precursors be stored in discrete compartments.
2.That there is a suitable extracellular reaction chamber into which the precursors can be discharged.
3.There must be a competence to control the discharges so that the correct sequences of events can occur in the production of the scleroproteins.
4.There must be some mechanism whereby the intricate shapes of the Shell Organic Matrix scleroproteins can be generated.
5.There must be some mechanism whereby these shapes are either built *in situ* on the shell or are placed in position on the shell following assembly.
6.There must be either physically or physiologically closed spaces so that the precursors of the various organic matrices can be kept discrete from each other, or a control over timing of secretion, or both.

6.1.6.Previously Published Views of Shell Formation in Bivalves.

In a review of Shell Formation, Wilbur and Saleuddin (1983) gave the following diagrammatic representation:-

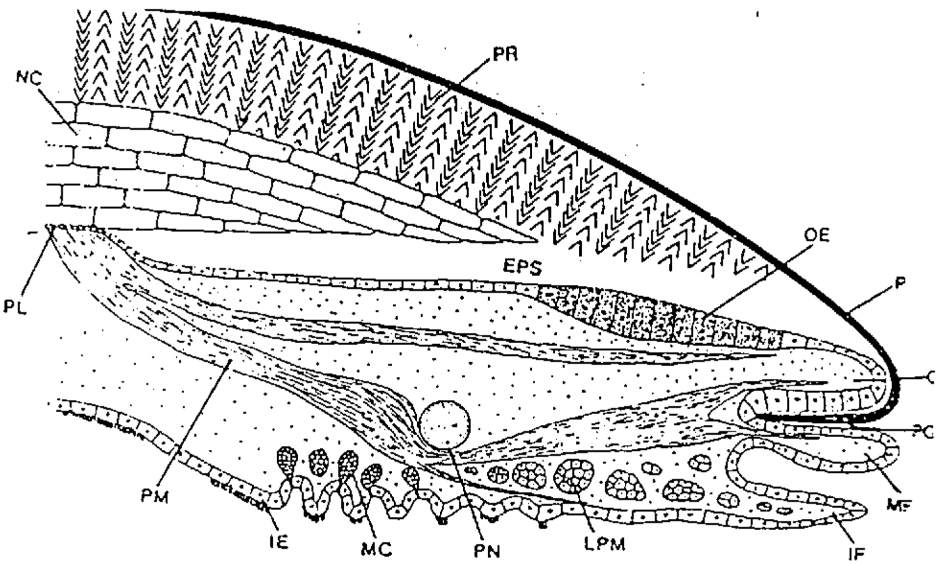


Fig. 1. Radial section of the mantle edge of a bivalve to show the relationship between the shell and mantle. (Not to scale.) EPS, Extrapallial space; IE, inner epithelium; IF, inner fold; LPM, longitudinal pallial muscle; MC, mucous cell; MF, middle fold; NC, nacreous shell layer; OE, outer epithelium; OF, outer fold; P, periostracum; PC, periostracal groove; PL, pallial line; PM, pallial muscle; PN, pallial nerve; PR, prismatic shell layer.

FIGURE 6.1. Diagrammatic representation of bivalve shell formation after Wilbur and Saleuddin, (1983), repeat of Figure 1.5.

This diagram can be criticised as deficient in that there is no mechanism shown for thickening the shell proximal to the Pallial Line. Further the illustration would be of more general use were the shell layers designated outer layer, middle layer and inner layer (Taylor, 1972). Thus a diagram illustrating a bivalve with three calcareous shell layers inside a "periostracum" with a mantle-shell junction at the Pallial Line and a peripheral adherence of forming "periostracum" to the outer surface of the valve can be shown as follows:-

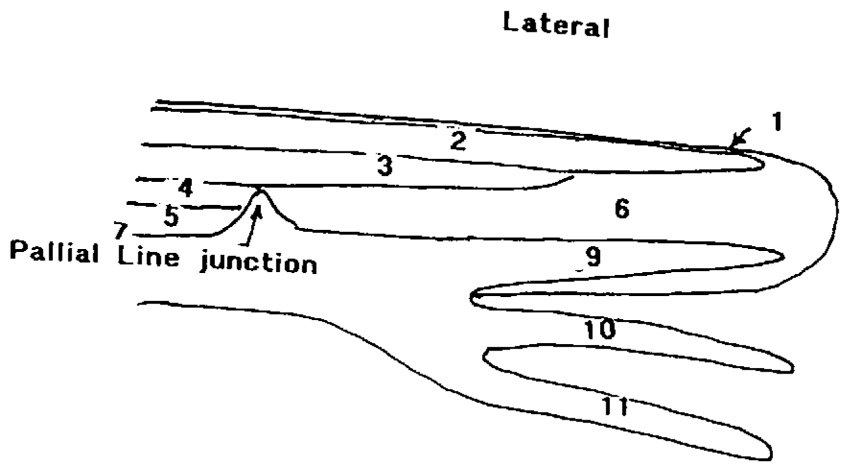


FIGURE 6.2. Diagrammatic representation of production of a Bivalve shell with an outer non-calcareous Shell Layer, an outer Shell Layer, a middle Shell Layer, and an Inner Shell Layer by a Bivalve with a Pallial Line junction and three Marginal Mantle Folds. 1 = outer non-calcareous Shell Layer; 2 = outer calcareous Shell Layer; 3 = middle calcareous Shell Layer; 4 = inner calcareous Shell Layer; 5 = Pallial Space; 6 = Extra-pallial Space; 7 = External Mantle; 9 = F1.; 10 = F2.; 11 = F3..

Such a Mantle Margin forming a scleroprotein "periostracum" is given in Waite's review of the quinone tanned scleroproteins in molluscs, (Waite, 1983):-

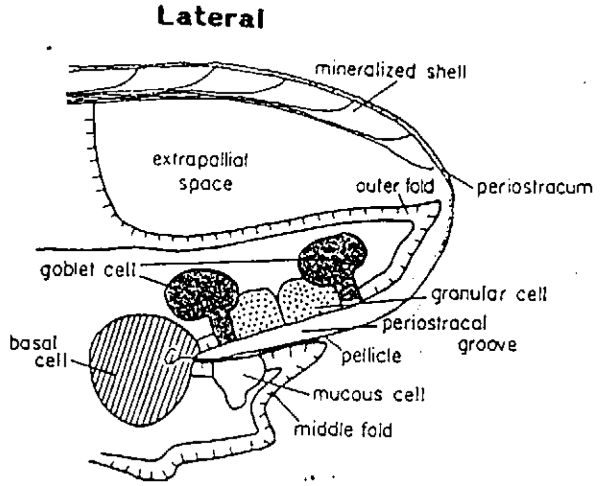


Fig. 4. Schematic illustration of the cells in the mantle epithelium presumed to participate in periostracum formation in bivalves. Each cell type is grossly exaggerated to emphasize its role.

FIGURE 6.3. "Schematic Illustration" of formation of periostracum in a periostracal groove - after Waite, (1983), repeat of Figure 1.7.

6.1.7.Shell Production in Pearl Oysters, and Tissue Differentiation of Their External Mantle and Mantle Margin.

Nakahara and Bevelander, (1971) adopted a similar view of production of "periostracum" in a "periostracal groove" to explain the production of Growth Processes in the pearl oyster *Pinctada radiata*. They attributed secretion of Prismatic Layer to the secretory activity of LatF1.:-

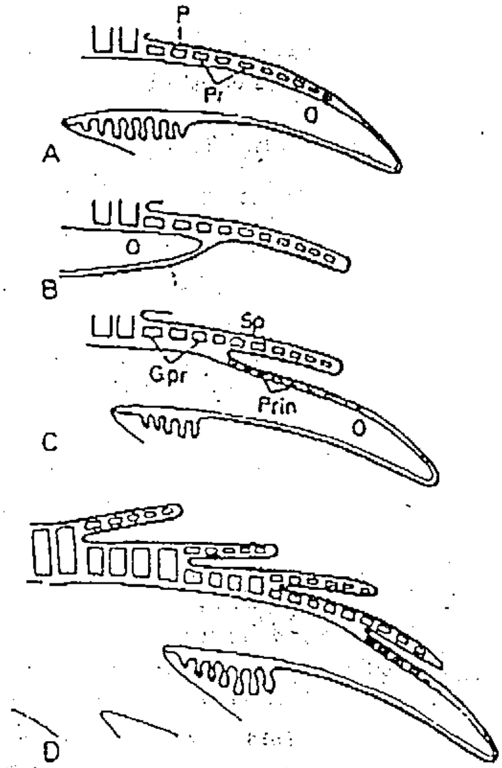


Fig. 16 A–D. This series of diagrams represents our interpretation of the important steps that occur in the formation of the prismatic layer (including spurs) of *P. radiata* and illustrates the relationship between the outer fold, the periostracum and the prisms. To simplify the illustrations, proportions of the structures were modified slightly. A. This diagram illustrates a completed single array of prisms located between the outer fold and the periostracum. Following the establishment of these prisms the mantle retracts. *P*, periostracum; *Pr*, prism array; *O*, outer fold. B. Showing new relation of the distal portion of the prism layer now enveloped on both upper and lower surfaces by the periostracum. *O*, outer fold. C. Mantle fold now in extended position again. The marginal portion of newly-formed array is enveloped by the periostracum to form a new spur. The proximal portion of the array is in continuous contact with the mantle epithelium and undergoes growth similar to proximal prisms previously formed. Also, prism formation is again initiated in the newly formed space between the margin of the outer fold and the periostracum. The formation of additional spurs consist of a repetition of the cycle A→B→C. *G.pr*, growing prisms; *O*, outer fold, *Pr in*, initiation of prisms; *Sp*, spur. D. Structure of shell and margin of the mantle following the formation of several spurs and additional prisms located proximally to the spurs. These prisms increase in height which results in a thickening of the prismatic layer. The periodic formation of additional proximal prisms also results in marginal growth

FIGURE 6.4. Production of periostracum in a periostracal Groove and Prismatic Layer by LatF1 in the pearl oyster *Pinclada radiata* - after Nakahara and Bevelander, (1971), repeat of Figure 1.9.

Dix, (1972), Jabbour-Zahab et al, (1992), and Garcia Gasca et al. (1994), present a picture of pearl shell formation essentially in agreement with Nakahara and Bevelander (1971).

Jabbour-Zahab et al. (1992), suggest dividing what they call the mantle edge into six areas with several sub-areas according to the following:-

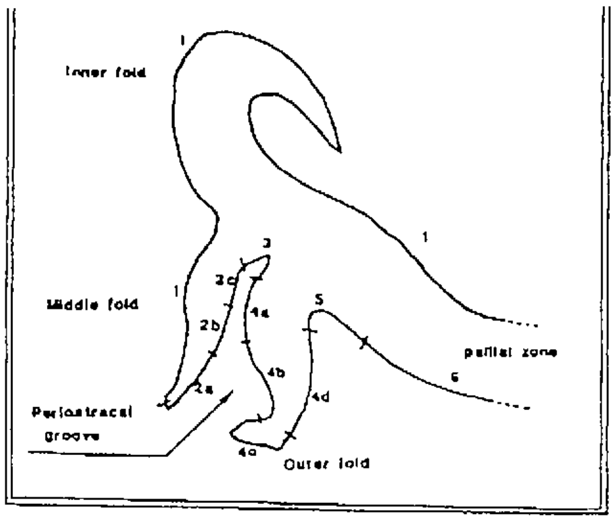


Figure 2. — Radial section through the mantle edge of *Pinctada margaritifera*.

FIGURE 6.5. Radial Section of Mantle Margin of *Pinctada margaritifera* showing suggested division into tissue areas, after Jabbour-Zahab et al.,(1992).

As below, many Bivalves have no equivalent to the third most lateral Marginal Mantle Fold of *Pinctada margaritifera* but all do have structures analogous to the two most lateral Marginal Mantle Folds. To apply the nomenclature of Jabbour-Zahab et al. (1992) to Bivalves in general would thus result in further confusion and ambiguity of nomenclature.

In this study in an attempt to maintain coherence with previous schemes of nomenclature as well as to generate a nomenclature which was at the same time strictly accurately descriptive and generally applicable throughout the Bivalvia, on anatomical, histological and morphological grounds, the following names were given to a pearl oyster's External Mantle and Mantle Margin Regions and parts of its shell.

1. = Isthmus
2.= Shoulder Region
3.= Pallial Gland Region
4.= Proximal Pallial Region
5.= Middle Pallial Region
6.= Distal Pallial Region
7.= Distal Folded Region
8.= Fold F1.
9.= Fold F2.
10.= Fold F3.
11. = Hinge
12.= Inner Nacreous Layer
13.= Outer Nacreous Layer
14.= Prismatic Layer
15.= Growth Scale
16.= Growth Process
17.= Major Prism
18.= Growth Process Prism

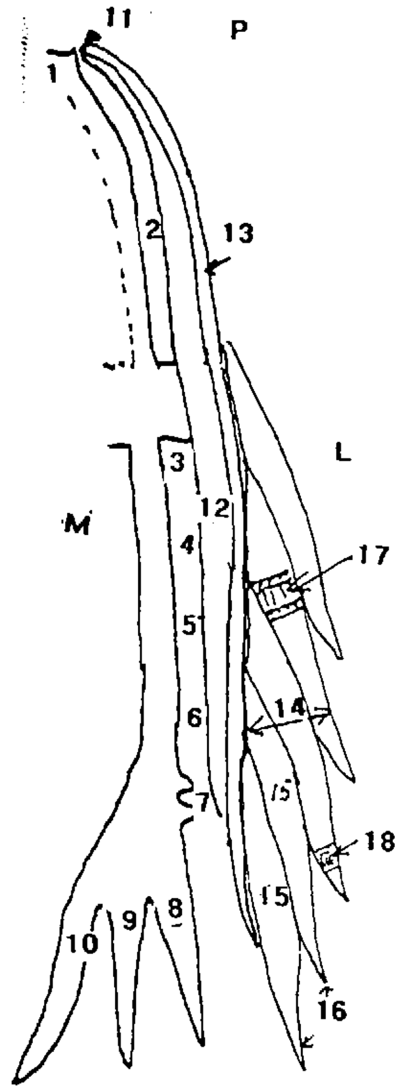


FIGURE 6.6. Diagrammatic representation of radial section of External Mantle and Mantle Margin and Valve of a Pinctada. L = lateral; M = medial; P = proximal; D = distal.

The components of a Major Prism and its lateral and medial coverings are illustrated in Figure 6.7.

lateral

1.= Outer Fibrous Sheath
2.= Outer Prismatic End Plate
3.= Inner Lateral Structure
4.= Parallel Transverse Prismatic Side Walls
5.= Linear Pattern Inter-prismatic Organic Matrix
6.= Reteform Inter-prismatic Organic Matrix
7.= Intra-Prismatic Organic Matrix
8.= Calcite Tablet
9.= Inner Prismatic End Plate
10.= Inner Fibrous Sheath

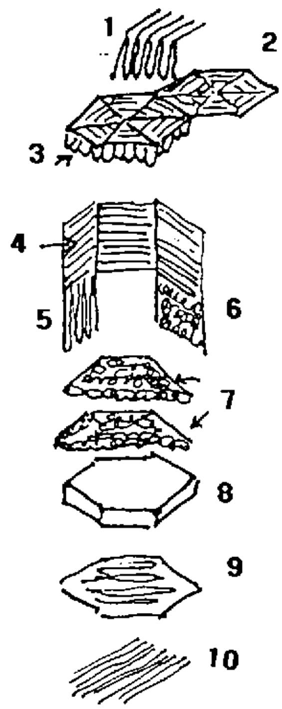
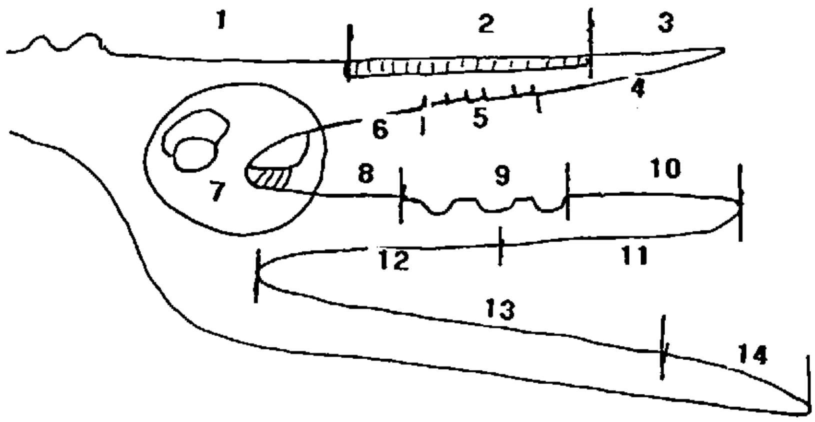


FIGURE 6.7. Expanded diagram showing the components of a Major Prism and its lateral and medial coverings.

The Mantle Margin was then subdivided on histological grounds into the following:



1.= Proximal LatF1. = Proximal Mantle Edge Gland
2.= Mantle Edge Gland (Proper)
3.= Terminal Region of LatF1.
4.= Distal MedF1.
5.= Middle MedF1.
6.= Proximal MedF1.
7.= Apical F1F2.
8.= Proximal LatF2.
9.= Middle LatF2.
10.= Distal LatF2.
11.= Distal MedF2.
12.= Proximal MedF2.
13.= Proximal LatF3.
14.= Distal LatF3.

FIGURE 6.8. Histological Regions of the Mantle Margins of pearl oysters.

It will be seen that 4, 5, 6, 7, 8, 9, and 10 of this study coincide exactly with the medial part of 4c, and 4b, 4a, 3, 2c, 2b, and 2a respectively of Jabbour-Zahab et al (1992).

Considering LatF2., Middle LatF2. Region is very similar on all members of the *Pinctada* studied and unique to this genera. The surface epithelium in fixed specimens displays a unique plateau-semicircular infolding-plateau conformation. The subepithelium contains Light Blue Glands - also unique to this area and ubiquitous in this area in the *Pinctada*. Amongst the Light Blue Glands are a kind of Trabecular Turquoise Gland unique to the Mantle Margin of LatF2. and to MedF2. and Proximal LatF3. i.e., that part of the Mantle Margin medial to the Pleated Secretion of Groove F1F2..

While the surface epithelial secretory structures and their relationship to the Subepithelial Glands of LatF2. in the genera *Pteria, Isognomon* and *Malleus* have an overall similarity to the picture seen in the *Pinctada* there are fundamental differences in the arrangement of the structures between the *Pinctada* and these other groups. For example, in all species of the *Pinctada*, the Proximal LatF2. bears an epithelium with a dense mat of apical microvilli but is almost devoid of cilia. Located in this epithelium are the Receptacle Glands of this genus. This situation abruptly changes just proximal to the traversing of the LatF2. epithelium by the secretory ducts of a large specialised type of Subepithelial Gland. Distal to this point the apical surface of the epithelial cells bears numerous elongate cilia as well as the dense mat of apical microvilli. The junction of Proximal LatF2. and Middle LatF2. is defined herein as the line of junction of the non-ciliated epithelium with the ciliated epithelium, as the presence of the cilia must denote a change in physiological function. In contrast, the surface epithelium of Latf2. of the *Pteria, Isognomon* and *Malleus* studied all bore the elongate cilia as well as the apical microvilli for the entire LatF2. and the Subepithelial Gland-Receptacle Gland relationship extended over the greater part of the LatF2. surface.

Similarly the relationship of Receptacle Glands to subepithelial secretory Glands are histologically unique to Proximal LatF2. in the *Pinctada* and an equivalent position in the *Ostreoidea, Arcoidea* and *Limopsoidea* and the LatF2 more generally in *Pteria, Isognomon, Malleus* and *Pinna* studied - ie. those genera which uniquely amongst the Bivalvia studied have the particular type of simple calcitic Prismatic Layer external to two aragonite Nacreous layers, and the *Ostreoidea, Arcoidea* and *Limopsoidea*. The relationship of the Prismatic Layer of pearl oysters to the outer Shell Layer of members of these other three Superfamilies will be returned to later.

The secretory glands and tissues and associated musculature of the Proximal and Medial LatF2. regions of pearl oysters (and those of the Ostreacea and Pinnacea studied), are emmeshed in branches of the F2. Ramus of the Circum-pallial Nerve.

Again, the surface epithelium and Subepithelial secretory structures of Distal LatF2. are very similar in all members of the *Pinctada* and unique to this location as are those of Distal MedF2., Proximal MedF2., Proximal LatF3. and Distal LatF3.. The latter four Regions are not distinguished by Jabbour-Zahab et al. but instead are included in their area 1 which also includes the medial surface of F3. This is histologically a very different tissue from any of the secretory tissues lateral to it on the Mantle Margin. It is held in this study that it plays no secretory part in Shell Layer formation.

Jabbour-Zahab et al., (1992), offer no explanation for the existence of any of these highly differentiated tissues located on the Mantle Margin of pearl oysters between Apical Groove F1F2. and the distal extremity of F3..
If the description of the formation of Prismatic layer and Growth Processes of *Pinctada radiata* given by Nakahara and Bevelander, (1971), is accepted i.e., if the Pleated Secretion of GrooveF1F2. is considered to be "forming periostracum" which then recurves around Terminal F1, to form the outer ("periostracal") layer of the Prismatic Layer, this also necessarily excludes all the highly differentiated secretory glands and tissues of LatF2., MedF2., and LatF3., from participation in shell formation. This acceptance also requires that the lateral surface of the Outer Fibrous Sheath be structurally the same as the medial Surface of the Inner Fibrous Sheath. This is not so in any pearl oyster studied.

No tissues on the External Mantle and Mantle Margin of a pearl oyster more closely fit the requirements for the production of a diverse and complicated scleroprotein structure, such as the Prismatic Layer Organic Matrix, than these tissues and glands of LatF2..

That a population of monotonously similar elongate columnar epithelial cells such as the cells of the Mantle Edge Gland could produce a structure as complex as the Prismatic Layer of a pearl oyster is to suggest a degree of "multi-skilling" unknown to this author in any highly differentiated tissue with a single cell type. The Mantle Edge Gland cells resemble the cells of the Isthmusistic Epithelium, which produce the hinge - the morphology of the constituent parts of which reflects the monotonous similarity of the cells which secrete the precursors of their protein rubber.

Similarly, Nakahara's (1991) description of Nacre formation takes account of neither location nor species specificity of Nacre morphology nor histological differentiation into the specific tissues of the different regions of the pearl oyster External Mantle.

6.1.8.Types of Physical Relationships Between Bivalves' Mantles and Mantle Margins and The Organic Matrices of Their Shells.

In determining the source of the Organic Matrix of a particular part of a shell or similar structure, the certitude attending the designation of tissue of origin is dependant on the exclusivity of the physical relationship between the forming shell layer (or structure) and the tissue responsible for secretion of the precursors of its Organic Matrices. Thus there can be little doubt that the Organic Matrices of the Nacreous Layer of a cultured pearl are formed from secretions of the pearl sac epithelium. The pearl sac epithelium is formed from pallial epithelium. Pallial epithelium underlies and is immediately adjacent to the Inner Nacreous Layer of the pearl shell. Similarly the accuracy with which the surface morphology of the Isthmusistic epithelium reflects the overlying shape and extent of the inner surface of the Hinge leaves very little doubt that this epithelium is involved in Hinge formation. No such certitude attends the relationships of Shell Layers and originating tissues for the more peripheral parts of a pearl shell and peripheral Mantle. The relationship between the various morphologically distinct areas of Inner Nacreous Layer and the histologically distinct areas of External Mantle immediately adjacent to them will be discussed more fully later (Chapter 6.3, 6.5).

The morphological relationships between the shells (and hence Shell Layers) and the tissues of the External Mantle and Mantle Margin of the members of the Bivalvia studied here are of three basic types:

1.Those Bivalves where the Pallial space is sealed distally by adherence of the External Mantle and medial surface of the Valve at the Pallial Line; and whose Extrapallial Space is sealed distally by the recurvature of a scleroprotein sheet, (whether forming part of the Organic Matrix of calcified Shell Layer or not), generated by the Mantle Margin, and its adherence to the shell periphery (as with the "periostracum" of Wilbur and Saleuddin, (1983)), Figure 6.9..

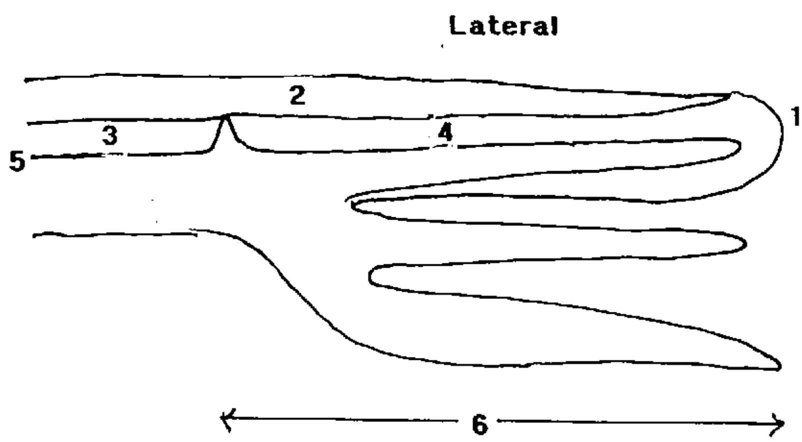


FIGURE 6.9. Diagram of shell/mantle relationships of Bivalves with the Pallial Space sealed at the Pallial Line and an Extra-pallial Space sealed by a recurving outer non-calcareous shell layer. 1 = recurved outer layer; 2 = Valve; 3 = Pallial Space; 4 = Extra-pallial Space; 5 = External Mantle; 6 = Mantle Margin.

This is the situation in all the heterodant bivalves studied as well as in the Unionoidean *Velesunio ambiguus*, and the *Mytilids*.

2Where a bivalve is attached at the periphery of Shell and Mantle but not at the Pallial Line this relationship is illustrated in Figure 6.10. :

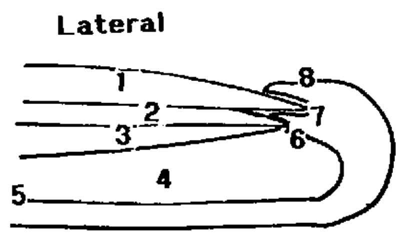


FIGURE 6.10. Diagram of Shell/Mantle relationships of Bivalves with Mantle Margin adherent to the Valve periphery but no Pallial Line junction. 1 = outer Shell Layer; 2 = Outer Nacreous Layer; 3 = Inner Nacreous Layer; 4 = Pallial Space; 5 = External Mantle; 6, 7and 8 = Mantle Margin Folds F1., F2. and F3. respectively.

This is the situation in the *Nuculoidea*

3.In a pearl oyster the most distal point of attachment is the Adductor Muscle and hence neither the Pallial Space nor the Extra-pallial Space are physically sealed as in the above cases (Figure 6.11.).

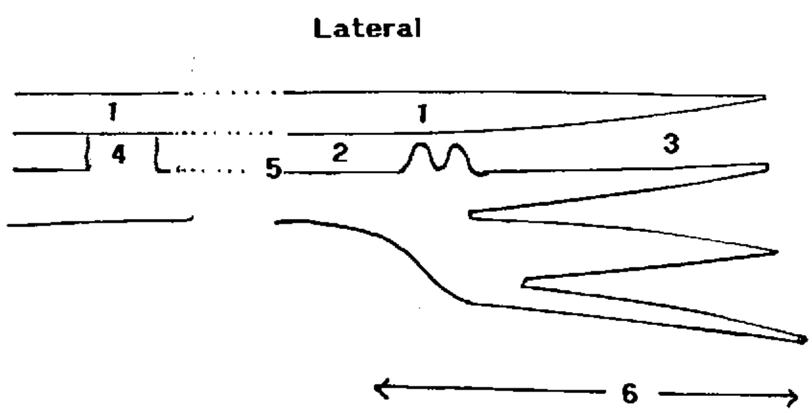


FIGURE 6.11. Diagram of radial section showing the shell/Mantle relationships of the External Mantle, Mantle Margin and Valve of the pearl oysters. 1 = Valve; 2 = Pallial "Space"; 3 = Extra-pallial "Space"; 4 = Adductor Muscle; 5 = External Mantle; 6 = Mantle Margin.

(The "Space" is in italics because it is not physically confined as in the other cases).

This is also the situation in those bivalves taxonomically close to pearl oysters - the *Pinnoidea*, *Ostreoidea*, *Anomioidea* and *Pectinoidea* (Waller 1978).

The examples of these three types of Mantle-shell morphological relationships will now be considered in detail.

6.1.9.Shells and Mantles in the Heterodonta.

Species which were members of the following seven Heterodant Superfamilies were included in this work - *Tellinoidea*, *Veneroidea*, *Solenoidea*, *Carditoidea*, *Mactroidea*, *Corbiculoidea* and *Gastrochaenoidea*.

Schematic diagrams showing the relationships of the External Mantles and Mantle Margins and some of their secretions found in these Superfamilies in this study and the Shell Layers described for them by Taylor, (1973) are presented below.

All twelve Genera of the Heterodonta from seven different Superfamilies included in this study had a junction of Mantle and Valve at the Pallial Line thus forming an enclosed Pallial Space. Further all these Genera showed an outer layer which resulted from the recurving of a scleroprotein sheet generated in a Marginal Mantle Groove. This means that all the above also had an enclosed Extrapallial Space.

As can be seen in Figures 6.12 to 6.18 of the six Genera of the Superfamily *Veneroidea* studied one (*Gafrarium*) has a Mantle Margin with four Marginal Mantle Folds and the other five have three Marginal Mantle Folds.

VENEROIDEA.

Dosinia juvenilis.

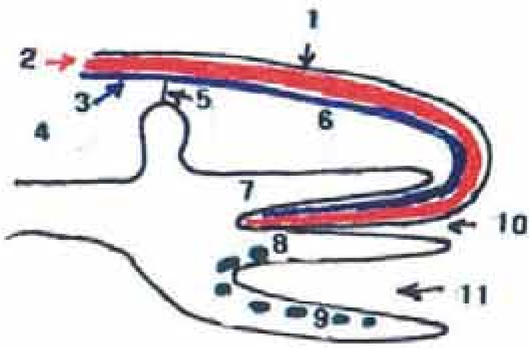


FIGURE 6.12

Circe trigona.

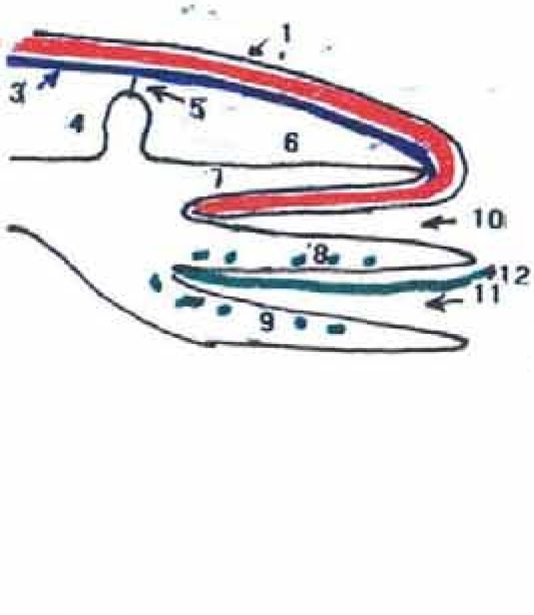


FIGURE 6.13

Figures 6.12 and 6.13. 1 = outer lamina of outer non-calcareous Shell Layer. 2 = middle lamina of outer non-calcareous Shell Layer. 3 = inner lamina of Outer Non-calcareous Shell Layer. 4 = Pallial Space. 5 = Pallial Line. 6 = Extra-pallial Space. 7 = F1.. 8 = F2.. 9 = F3.. 10 = Groove F1F2.. 11 = Groove F2F3.. 12 = acid muco-polysaccharide secretion of Groove F2F3..

Gafrarium divaricatum.

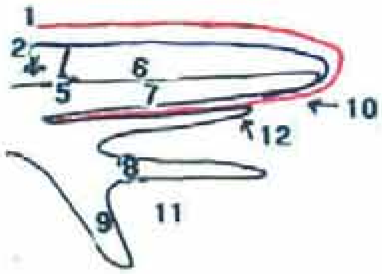


FIGURE 6.14

Gafrarium ? sp.

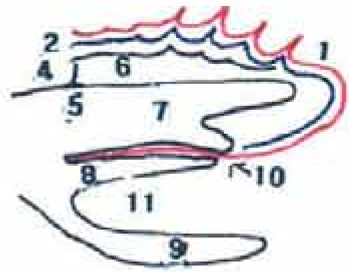


FIGURE 6.15

FIGURES 6.14 and 6.15. 1 = outer lamina of non-calcareous outer Shell Layer. 2 = medial lamina of non-calcareous outer Shell Layer (= middle lamina of trilaminate outer Shell Layer). 4 = Pallial Space. 5 = Pallial Line. 6 = Extra-pallial Space. 7 = F1.. 8 = F2.. 9 = F3.. 10 = Groove F1F2.. 11 = Groove F2F3.. 12 = Ancillary F2..

Placamen calophyllum.

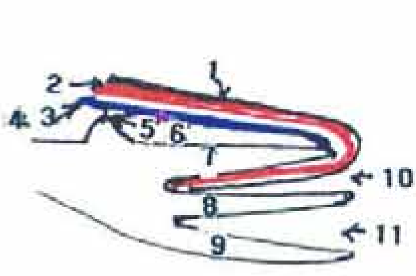


FIGURE 6.16.

FIGURE 6.16. 1 = outer lamina of non-calcareous outer Shell Layer. 2 = middle lamina of non-calcareous outer Shell Layer. 3 = inner lamina of non-calcareous outer Shell Layer. 4 = Pallial Space. 5 = Pallial Line. 6 = Extra-pallial Space. 7 = F1.. 8 = F2.. 9 = F3.. 10 = Groove F1F2.. 11 = Groove F2F3..

Globivenus embrithes.

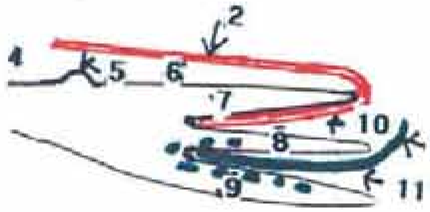


FIGURE 6.17

Tapes sp.

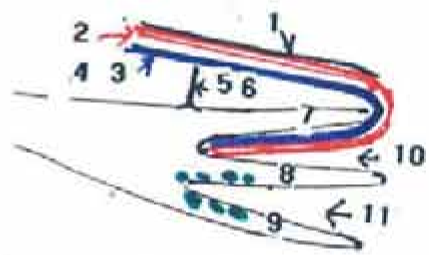


FIGURE 6.18.

FIGURES 6.17 and 6.18. 1 = outer lamina of outer non-calcareous Shell Layer. 2 = middle lamina of outer non-calcareous Shell Layer (*Globivenus Embrithes.* No inner lamina of outer non-calcareous Shell Layer observed in *Tapes sp.*, 3 = inner lamina of outer non-calcareous Shell Layer. 4 = Pallial Space. 5 = Pallial Line. 6 = Extra Pallial Space. 7 = F1.. 8 = F2.. 9 = F3.. 10 = Groove F1F2.. 11 = Groove F2F3..

In all six Genera a scleroprotein which stains amber with Mallorys, yellow with M.S.B and A.B./M.S.B red with Azan and red with Steedmans (illustrated in red in the Figures 6.12-6.18 above) is sythesiesed in the groove between the outer two Marginal Mantle Folds (Groove F1F2.) and recurves around the terminal part of Fold F1. to form part of the Outer Shell Layer. For this reason the second most lateral Marginal Mantle Fold of *Gafrarium* being medial to this scleroprotein is here held to be Ancillary F2..

Given this, it can then be said that there is in all members of the *Veneroidea* studied a heavy concentration of acid-mucopolysaccharide secretory glands in Groove F2F3.

At least in *Globivenus embrithes* these glands appear to form a lateral lamina of the outer Shell Layer and they may also do so in the other genera.

Thus even in the Superfamily whose many varied genera best fit the standard shell forming diagram of Wilbur and Saleuddin (1983), the name "periostracum" may well be a misnomer and the Lateral lamina at least of the outer Shell Layer may originate not in the "groove located between the outer fold and the middle fold" (Jabbour - Zahad et al, 1992, Waller, 1978) but in the Groove F2F3..

With this qualification, the six genera of the superfamily *Veneracea* have a Mantle Margin - Shell Spatial Relationship in accord with that in Figure 6.2 above, adapted from the schematic diagrams of Wilbur and Saleuddin, (1983) and Waite, (1983).

TELLINOIDEA.

Phylloda foliacea.

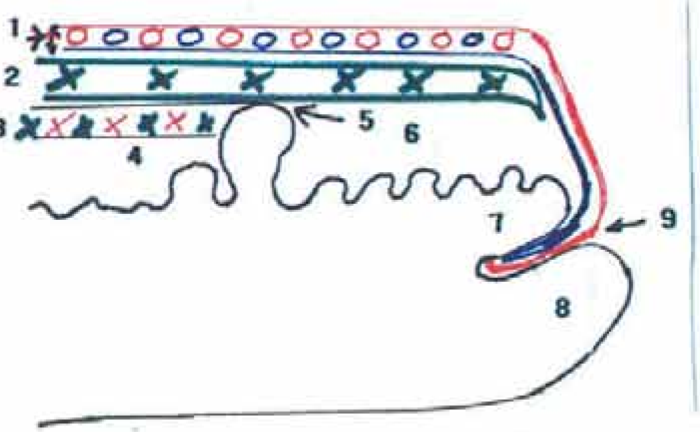


FIGURE 6.19. *Phylloda foliacea* 1 = Composite Prismatic Layer (Aragonite). 2 = Crossed Lamellar Structure (aragonite or calcite). 3 = Complex Crossed Lamellar Layer (aragonite or calcite). 4 = Pallial Space. 5 = Pallial Line. 6 = Extra-pallial Space. 7 = F1.. 8 = F2.. 9 = Groove F1F2..

The *Tellinacea* studied have only one Marginal Mantle Groove. Proximally it generates a sheet of scleroprotein which stains amber with Mallory's. Yellow with M.S.B. and A.B./M.S.B. and red with azan. These colour reactions to these stains are the same as seen with one of the scleroproteins generated by the Mantle margin of all the members of the Veneroidea studied, and as with these latter it recurves around the Terminal part of Fold F1. to form the outer layer of the shell. It is joined on what becomes, following recurvature, its medial surface by another scleroprotein which originates at least in part from the medial and terminal parts of Fold F1. and may well form part of the Organic Matrix of the Composite Prismatic Layer - the outermost of the three calcareous layers of the tellinoidean shell (Taylor, 1972). This has elements in common with the description of the fine structure of "periostracum" given by Saleuddin and Petit (1983) pages 222 - 224. The Organic Matrices of the middle calcareous layer of the Tellinoidea, a Crossed Lamellar Structure, (Taylor, 1972), appear to be secreted by the lateral surface of F1.. The Inner Shell Layer Organic Matrices (Complex Crossed Lamellar Structure), are necessarily secreted by the Pallial Mantle.

SOLENOIDEA.

Solen vagina.

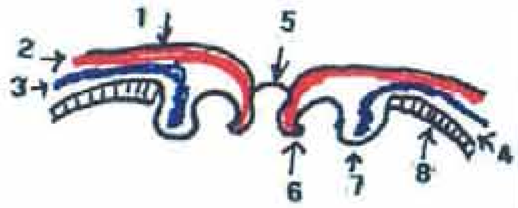


FIGURE 6.20. *Solen vagina* 1 = outer lamina of non-calcareous outer Shell Layer. 2 = inner lamina of bilaminate non-calcareous outer Shell Layer. 3 = Organic Matrix of outer calcareous Shell Layer. 4 = Extra-pallial Space. 5 = (probably) fused F2.s. 6 = (probably) Groove F1F2.. 7 = (probably) invagination of Terminal F1.. 8 = Mantle Edge Gland.

The Solenoidea studied, *Solen vagina* and *S. grandis* have near identical Mantles, Mantle Margins and Shell Layers. The Mantle Margins of these animals are fused in the Ventral Midline. The midline fusion may probably be properly regarded as a fusion of left and right F2s. In any case scleroprotein sheets which stain amber with Mallorys, yellow with M.S.B and A.B./M.S.B.. and red with Azan are produced in the grooves on either side of the midline fusion and recurve outwards to form the outer layers of the valves. They are underlain on the valve surfaces by a fairly dense calcareous layer whose organic matrices are in part produced by a groove lateral to the groove which produced the outer scleroprotein Shell Layer. From a consideration of the histologies and staining affinities of this area of the Mantle Margin of related Heterodont Bivalves, this groove is probably more properly considered an invagination of Terminal F1. than a structure analogous to Groove F1F2.. Lateral to this the columnar cells of the Mantle Edge Gland of LatF1. also appear to contribute to the Organic Matrices of this inner Shell Layer.

CARDITOIDEA.

Cardita variegata.

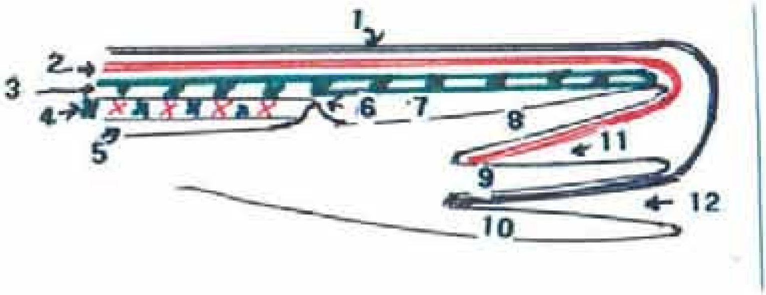


FIGURE 6.21 *Cardita variegata*. 1 = outer lamina of non-calcified outer Shell Layer from Groove F2F3.. 2 = inner lamina of bilaminate non-calcified outer Shell Layer from Groove F1F2.. 3 = Crossed Lamellar Structure (aragonite or calcite). 4 = Complex Crossed Lamellar Structure (aragonite or calcite). 5 = Pallial Space. 6 = Pallial Line. 7 = Extra-pallial Space. 8 = F1.. 9 = F2.. 10 = F3.. 11 = Groove F1F2.. 12 = Groove F2F3..

The carditoidean *Cardita variegata* has a double outer non-calcified Shell layer, the outer lamina of which is a product of Groove F2F3.. It recurves around the extremity of Fold F1. outside the non-calcified layer from Groove F1F2. which stains amber with Mallorys, yellow with M.S.B and A.B./M.S.B and red with Azan. The organic matrices of the two calcareous layers - Crossed Lamellar Structure laterally and Complex Crossed Lamellar Structure medially - appear to be produced by LatF1. and the Pallial Mantle respectively.

MACTROIDEA.

Mactra abbreviata.

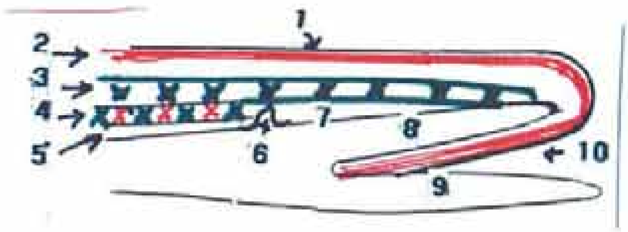


FIGURE 6.22 1 = outer lamina of non-calcareous outer Shell Layer from Groove F1F2.. 2 = inner lamina of bilaminate non-calcareous outer Shell Layer from Groove F1F2.. 3 = Crossed Lamellar Structure (aragonite or calcite) from LatF1.. 4 = Complex Crossed Lamellar Structure (aragonite or calcite) from Pallial Mantle. 5 = Pallial Space. 6 = Pallial Line. 7 = Extra-pallial Space. 8 = F1.. 9 = F2.. 10 = Groove F1F2..

The Mactroidea appear to have a very thin scleroprotein outer lamina of the Non-calcareous Outer Shell Layer secreted by the only Marginal Mantle Groove, Groove F1F2.. Inside this are scleroprotein secretions which appear to form an inner lamina of the outer non-calcareous Shell Layer and the Organic Matrix of the outer calcareous Shell Layer (Crossed Lamellar Structure). The Organic Matrices of the inner calcareous Shell Layer, being pallial, necessarily must originate from the Pallial epithelium.

CORBICULOIDEA

Geloina coaxans.

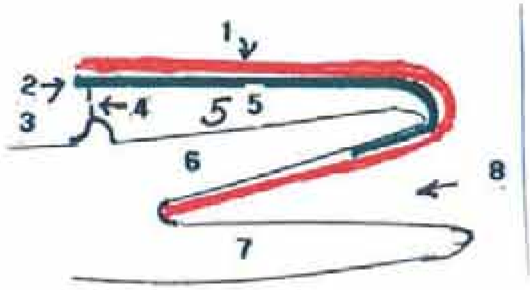


FIGURE 6.23. *Geloina coaxans.* 1 = outer lamina of bilaminate non-calcareous outer Shell Layer from Proximal Groove F1F2.. 2 = inner lamina of non-calcareous outer Shell Layer from Distal MedF1. and Terminal LatF1.. 3 = Pallial Space. 4 = Pallial Line. 5 = Extra-pallial Space. 6 = F1.. 7 = F2.. 8 = Groove F1F2..

The corbiculoidean *Geloina coaxans* is of interest in that the outer lamina of the trilaminate non-calcareous outer Shell Layer seen in nearly all other Heterodont Bivalves is missing. The outer lamina of the bilaminate Non-calcareous Outer Shell Layer recurves from the single Mantle Margin Groove F1F2. and has the same staining affinities as the middle lamina of the trilaminate Non-calcareous outer Shell Layers of other Heterodont Bivalves. A thicker, denser, non-calcified layer which lies inside this is added at Distal MedF1. and Terminal LatF1.. This latter lamina stains dark blue/green with Steedmans whereas the outer lamina stains red with this stain. Medial to this bilaminate outer Shell Layer, the two calcareous layers appear distinct - the medial surface of the more lateral lining the lateral Extra-pallial Space and the medial surface of the more medial lining the lateral Pallial Space.

GASTROCHAENOIDEA.

Gastrochaena cuneiformis.

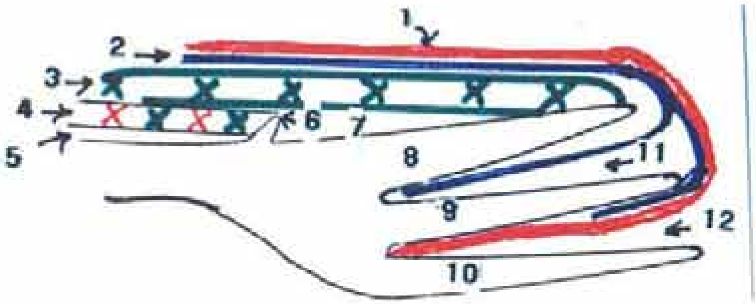


FIGURE 6.24. *Gastrochaena cuneiformis.* 1 = lateral lamina of bilaminate non-calcified outer Shell Layer from Groove F2F3.. 2 = inner lamina of bilaminate non-calcified outer Shell Layer from Grooves F1F2., F2F3.. 3 = Crossed Lamellar Strucutre. 4 = Complex Crossed Lamellar Structure. 5 = Pallial Space. 6 = Pallial Line. 7 = Extra-pallial Space. 8 = F1.. 9 = F2. (or Ancillary F1.). 10 = F2. (or F3.). 11 = Groove F1F2. (or Groove F1 AncillaryF1.). 12 = Groove F2F3. (or Groove AncillaryF1F2.).

Finally for the Heterodonta, the gastrochaenoidean *Gastrochaena cuneiformis* has a shell with a bilaminate outer Non-calcareous Shell Layer outside two calcareous Shell Layers. The Mantle Margin has three Marginal Mantle Folds (plus an ancillary Fold F3). A scleroprotein sheet which stains amber with Mallorys, yellow with M.S.B. and A.B./M.S.B. and red with Azan is produced in the more medial of the two Marginal Mantle Grooves and recurves around the termini of Folds F2. and F1. to form the outer lamina of the outer Non-calcareous Shell Layer. Because of its similarity to the Scleroprotein normally found emanating from Groove F1F2. in the heterodonta an alternative nomenclature for the Marginal Mantle is given in the legend to Figure 6.24 above. Another scleroprotein sheet which stains shades of blue with all the above stains is produced in part by the medial surface of the middle Marginal Mantle Fold and by Groove F1F2. and recurves inside the scleroprotein sheet from Groove F2F3. referred to above so that it forms the medial lamina of the Outer non-calcareous Shell Layer. The Crossed Lamellar Structure which forms the outer calcareous Shell Layer is produced by the LatF1. and the inner Complex Crossed Lamellar Shell Layer by the Pallial Mantle.

In summary, in the Heterondonta studied, with the possible exception of the Solenoidea, the Valve and Mantle are physically attached at both the Pallial Line and the shell-mantle periphery.

In every species studied there is a scleroprotein which stains amber with Mallory's, yellow/gold with M.S.B. and A.B./M.S.B., red with Azan and red with Steedman's which recurves from its site of synthesis in a Marginal Mantle Groove to lie lateral to the calcareous layers of the valve. Where this scleroprotein forms part of a trilaminate outer Non-calcareous Shell Layer it is invariably the middle lamina, and in these cases the lamina lateral to it is invariably a thin lamina fairly refractory to most stains used. The medial lamina of these trilaminate Non-calcareous outer Shell Layers invariably stains shades of blue with all stains used. Where this scleroprotein forms part of a bilaminate Non-calcareous Outer Shell Layer it may be either the more medial of the two laminae, (as in the Veneroideans *Gafrarium divaricatum, Cardita variegata or Mactra obesa)* or it may be the more lateral of the two laminae as in *Geloina coaxans* or *Gastrochaena cuneiformis.* It is usually formed in Groove F1F2, but in some cases is formed in Groove F2F3.. In the only instance in the Heterodonta studied where this scleroprotein did not take part in the formation of a trilaminate or bilaminate Non-calcareous Outer Shell Layer it appeared to join with the blue staining secretion secreted inside it to form the Organic Matrices of the Composite Prismatic Layer on the lateral surface of the valves of the Tellinoidea.

It thus is a misnomer to call it "periostracum" and its site of origin, even in the Heterodonta is not exclusively the most lateral of the Marginal Mantle Grooves.

As will be seen, a similarly staining scleroprotein is formed in the Mantle Marginal Grooves of every bivalve studied with the exceptions of those of the Limopsoidea, Arcoidea, Ostreoidea and Pectinoidea. While of course many histological features stain red with Azan and Steedman's, and some fewer amber with Mallory's, the yellow/gold colour displayed by this scleroprotein secretion after staining with M.S.B. or A.B./M.S.B. is unique to itself and the Shell Organic Matrices formed from it in the entirety of tissues and decalcified Shell Organic Matrices investigated in this work, with two exceptions : A similar yellow/gold colour was displayed by glands in the External Mantle of *Isognomon isognomon*, and on MedF2. of the Limopsoidea and Arcoidea following staining with A.B./M.S.B.. For this reason these scleroproteins will be referred to as martius yellow positive scleroproteins.

To highlight the part played by these scleroproteins in Shell Layer formation they will be depicted in red in all sketches in Chapter 6.1.

6.1.10.Shells and Mantles in the Limopsoidea and Arcoidea.

The higher order Taxonomy of the Limopsoidea and Arcoidea has been a matter of considerable confusion. As above, Waller (1978) classifies them as close to the Mytiloidea and Pinnoidea in the Pteriomorphia while Taylor, (1973) suggests that their shells are derived by part of the same evolutionary trend which gave rise to numerous heterodont superfamilies.

The Mantle Margin of these animals appears more variable in morphology than that of any other group studied but the histology of the secretory structures of the Mantle and Mantle Margin, taken sequentially from the Isthmus to the Medial Surface of the Mantle is surprisingly similar through the two Superfamilies and seven genera.

LIMOPSOIDEA.

Melaxinaea vitrea

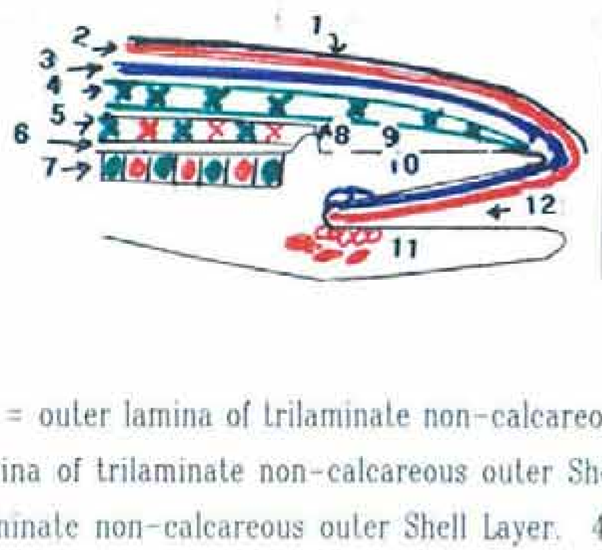


FIGURE 6.25. 1 = outer lamina of trilaminate non-calcareous outer Shell Layer. 2 = middle lamina of trilaminate non-calcareous outer Shell Layer. 3 = inner lamina of trilaminate non-calcareous outer Shell Layer. 4 = Crossed Lamellar Layer. 5 = Complex Crossed Lamellar Layer. 6 = Pallial Space. 7 = Beeswax Glands of External Pallial Mantle. 8 = Pallial Line. 9 = Extra-pallial Space. 10 = F1.. 11 = F2..12 = Groove F1F2..

ARCOIDEA.

Anadara antiquata/ pilula.

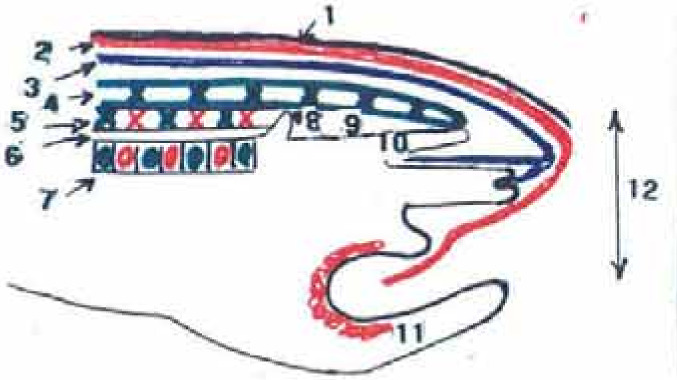


FIGURE 6.26. 1 = outer lamina of trilaminate non-calcareous outer Shell Layer. 2 = middle lamina of trilaminate non-calcareous outer Shell Layer. 3 = inner lamina of trilaminate non-calcerous outer Shell Layer. 4 = Crossed Lamellar Layer. 5 = Complex Crossed Lamellar Layer. 6 = Pallial Space. 7 = Beeswax Glands of External Pallial Mantle. 8 = Pallial Line. 9 = Pallial Space. 10 = F1.. 11 = F2.. 12 = Groove F1F2..

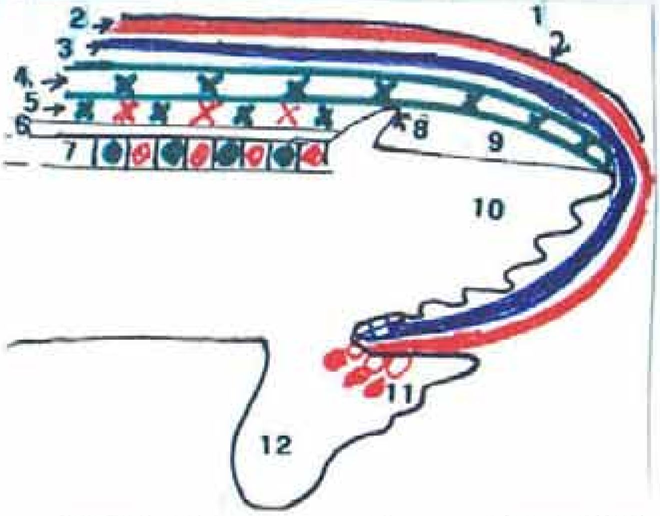


FIGURE 6.27. *Arca aladdin*

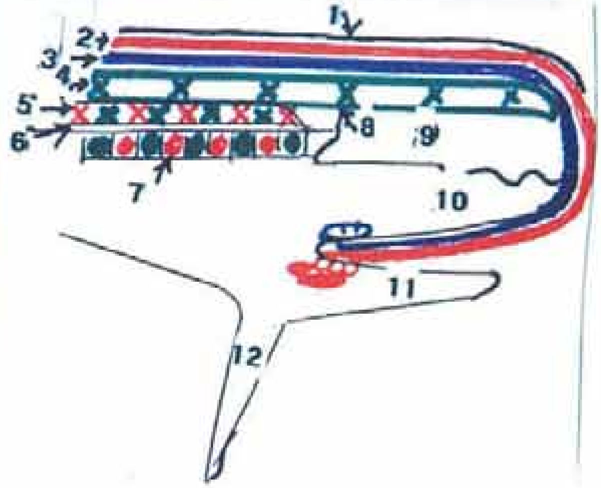


FIGURE 6.28. *Barbatium amygdalumtostum*

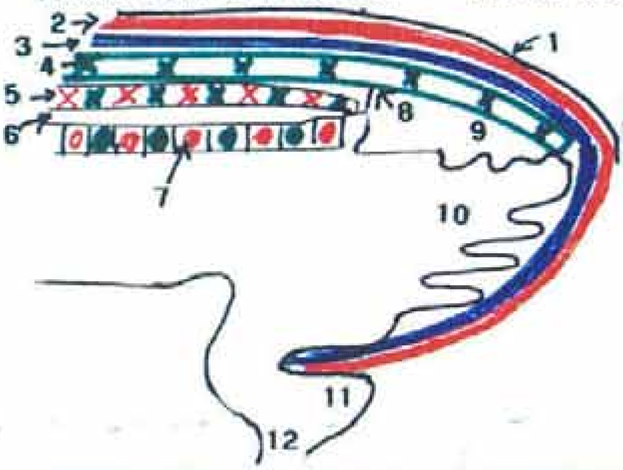


FIGURE 6.29. *Mesocibota luana*

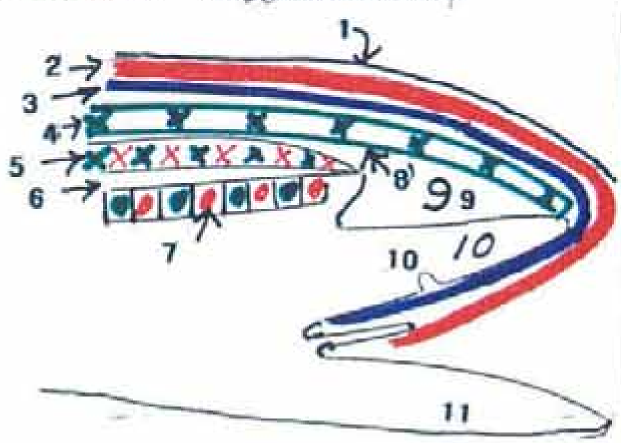


FIGURE 6.30. *Trisidos tortuosa*

Figures 6.27.-6.30.1 = outer lamina non-calcareous outer Shell Layer. 2 = middle lamina non-calcareous outer Shell Layer. 3 = inner lamina non-calcareous outer Shell Layer. 4 = Crossed Lamellar Structure. 5 = Complex Crossed Lamallar Structure. 6 = Pallial Space. 7 = Beeswax Glands of Pallial Mantle. 8 = Pallial Line. 9 = Extra-pallial Space. 10 = F1.. 11= F2.. 12 = F3..

Again, as for the Heterodonta, this group has a Pallial Line Junction of the Medial surface of the Valve to the lateral surface of the External Mantle thereby generating a closed Pallial Space. Similarly the Extrapallial Space is sealed by the recurving of a trilaminate Outer Non-calcareous Shell Layer.

The External Mantle of the Pallial Region is covered by a remarkable secretory epithelium which is very similar in all members of these Superfamilies and quite distinct from any secretory structure encountered elsewhere in the Bivalvia. It consists apparently almost exclusively of secretory glands of almost equal size. The side walls of each gland are normal to the basal lamina and hence the glands present a honeycomb-like picture. Each gland is filled with spherical secretory granules which stain at least three different colours with each trichrome stain used. The cells are further differentiated by granule size. This unique epithelium (Beeswax Glands) is most pronounced in the limopsacean *Melaxinaea vitrea* and the arcacean *Anadara pilula* but is present in every species of these two Superfamilies studied.

There can be little doubt that the function of this very specialised tissue is the secretion of the Inner Calcareous Shell Layer Organic Matrices precursors. This Shell Layer, throughout these groups, is Complex Crossed Lamellar Layer. Again the array of secretory glands between the Pallial line and the origin of the medial lamina of the outer non-calcareous Shell Layer is quite similar throughout the group as is the prominent and brightly staining Mantle Edge Gland covering the distal half of the LatF1..

Again, since these secretory structures line the enclosed Extra-pallial Space it would seem unlikely that their major function was other than the secretion of the outer of the two calcareous Shell Layers of these groups. Throughout these two groups the outer Calcareous Shell Layer is a Crossed Lamellar Structure (Taylor !973).

This arrangement of a Crossed Lamellar Structure secreted by the tissues lining an enclosed Extrapallial Space lateral to a Complex Crossed Lamellar Structure secreted by the External Mantle of the Pallial Region lining an enclosed Pallial Space is precisely the same as that seen in the Tellinoidea, some Veneroidea, the Carditoidea, Mactroidea, and the Gastrochaenoidea yet there would appear to be no resemblance whatsoever between the Pallial Region histology and Extrapallial Region histology of the Limopsoidea and Arcoidea on the one hand and the equivalent tissues of members of the heterodont superfamilies on the other (save that they both have Mantle Edge Glands, but very different Mantle Edge Glands). This finding seriously prejudices the use made of Shell Layers as taxonomic tools by Taylor, (1972) and Waller (1978) unless similarity in Shell

Layers is coupled with similarity in the histological structures which secrete their precursors..

The origins of the two inner laminae of the outer non-calcareous layer are also quite similar throughout these groups.

The medial of the two laminae is produced, in part, by an epithelial glandular structure which occupies the same position on the lateral aspect of Apical Groove F1F2, as the Omega Gland in Pearl Oysters. The secretions here produced stain shades of blue or purple with all stains used throughout the members of these Groups.

The middle lamina of the trilaminate outer non-calcareous Shell Layer of these groups is produced unequivocally on LatF2.. It is secreted through a variant of the Receptacle Glands found in exactly this location in the *Pinctada* from Subepithelial glands of Proximal LatF2.. In every member of these groups examined this middle lamina of the outer non-calcareous Shell Layer stained amber with Mallorys, yellowish to faint pink with M.S.B. and A.B./M.S.B. and red with Azan and Steedmans. This is thought to be related to the martius yellow positive scleroproteins of other groups.

The Limopsoidea and Arcoidea have an outer lamina of their trilaminate outer Shell Layer which is refractory to staining with any stain used. Its origin is uncertain but necessarily must be medial to the source of the middle lamina of the Outer Shell Layer.

The significance of these findings re the relationships between the Mantle and Mantle Margin and the Shell Layers in the Limopsoidea and Arcoidea are twofold: Firstly they place these two Superfamilies firmly in the Pteriomorphia as the tissues which produce those calcareous Shell layers (because of which Taylor (1973) placed them in the Heterodonta), are utterly distinct from the Heterodont tissues which produce similar Shell Layers - and the tissues which produce the Non-calcareous Shell Layers, in location and histology, are unmistakably akin to those which produce the Outer Shell Layer in the Pterioidea.

Secondly the production of an outer Shell Layer from Receptacle Gland-like structures on Proximal LatF2 and structures medial to this on the Mantle Margin is of great significance in respect of the sources of the precursors of the Shell Organic Matrices of the outer Prismatic Shell Layer in the Pterioidea. This will be returned to later (Chapter 6.6.)

6.1.11.Shell and Mantle of a Unionoidean.

Velesunio ambiguus.

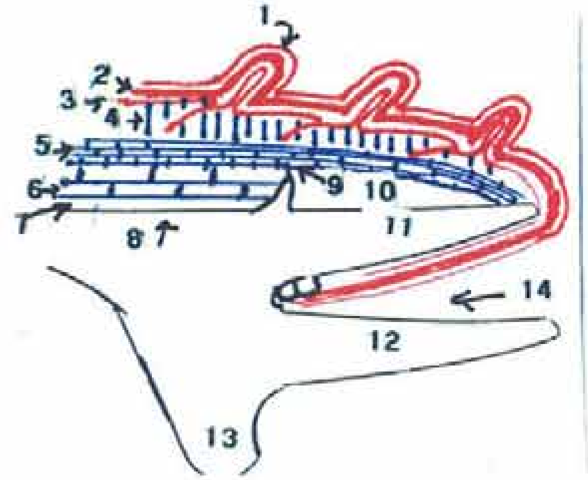


FIGURE 6.31. 1 = lateral lamina of trilaminate non-calcified outer Shell Layer from Apical Groove F1F2.. 2 = middle lamina of trilaminate non-calcified outer Shell Layer from Proximal Med F1.. 3 = inner lamina of trilaminate non-calcified outer Shell Layer from MedF1.. 4 = Aragonite Prisms which form-near inner lammina of outer non-calcified Shell Layer -Organic Matrices from F1.. 5 = Outer Nacreous Layer Organic Matrices from F1.. 6 = Inner Nacreous Layer Organic Matrices from External (Pallial) Mantle. 7 = Pallial Space. 8 = Pallial Mantle. 9 = Pallial Line. 10 = Extra-pallial Space. 11 = F1.. 12 = F2.. 13 = F3.. 14 = Groove F1F2..

The Valve/Mantle configuration in *V. ambiguus* displays a Pallial Line Junction between the medial surface of the valve and the lateral surface of the External Mantle forming a sealed Pallial Space, and a recurved trilaminate non-calcareous outer Shell Layer which joins the Mantle Margin from its origins in Groove F1F2. to the lateral periphery of the Shell. There is thus a sealed Extrapallial Space.

This trilaminate non-calcareous outer Shell Layer is thrown into elongate folds whose internal sides touch each other just distal to the growing shell periphery.

The middle lamina which appears to be secreted by the distal part of the tuft of elongate columnar cells on the lateral aspect of Apical Groove F1F2, stains amber with Mallory"s, yellow with M.S.B. and A.B./M.S.B. and red with Azan and Steedman's. It is thus the martius yellow positive protein in this species. In a way that is obscure, the medial lamina of this trilaminate outer Shell Layer forms a flat sigmoid structure which bends proximally out of the fold of the outer Shell Layer and then bends medially and then recurves on itself thus forming a bilaminate flattish sigmoid. The aragonite Prismatic Layer which is the outer calcareous Shell Layer lines both surfaces of this sigmoid structure and the medial surface of the medial lamina of the Outer Shell Layer where it is not forming a sigmoid strucutre.

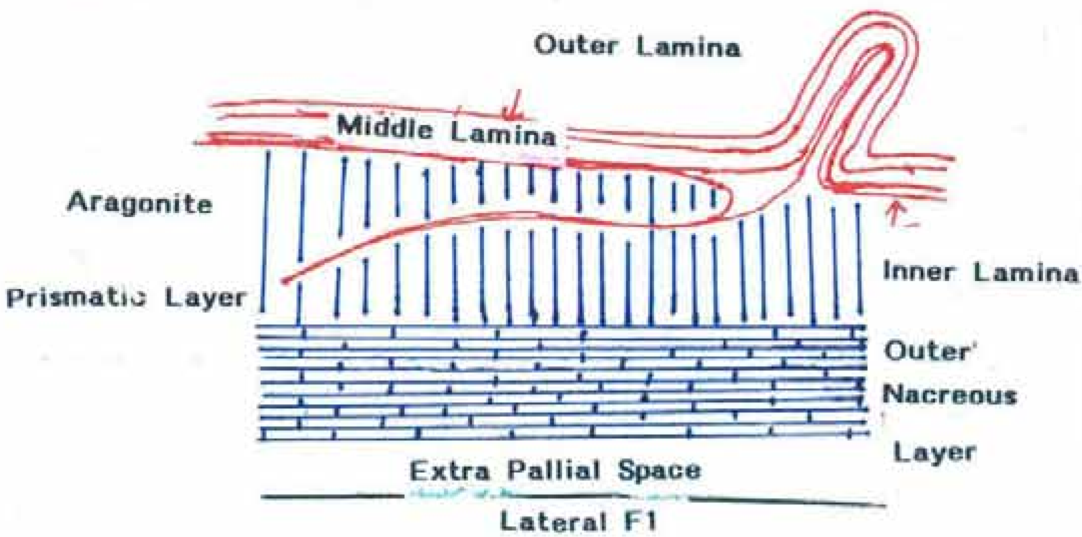


FIGURE 6.32. *Velesunio ambiguus* Schematic diagram showing relationships of aragonite Prismatic Layer to underlying Outer Nacreous Layer and laterally, to the overlying inner lamina of the trilaminate outer non-calcareous Shell Layer.

Medial to this aragonite Prismatic Layer, the Outer Nacreous Layer is necessarily formed by secretory structures of LatF1, (the medial surface of the Extrapallial Space) with the possible addition of material from distal MedF1.. Proximal to the Pallial Line attachment the Inner Nacreous Layer lines the lateral surface of the Pallial Space and hence is almost certainly produced by the secretory functions of the pallial Mantle.

These latter are all but unique in this study in that from L.M. it appears that the Pallial surface epithelial cells are undifferentiated squamous epithelium and hence very unlikely to be on their own the source of scleroprotein precursors. It may be that the array of spherical Granular Cytoplasm Secretory Glands in the trabecular walls of the submucosal sinus produce the scleroprotein precursors which are then transferred to the overlying epithelium and thence to the Pallial Space..

Concerning the aragonite Outer Prismatic Layer of *V.ambiguus* Taylor. (1972) lists the arrangement of calcareous layers in the Unionoidea of an outer aragonite Prismatic Shell Layer outside two aragonite Nacreous Shell Layers as an ancestral primitive condition from which all other types of bivalve shells derive. I am not in a position to dispute this. Species of the other three Superfamilies said by Taylor to exhibit this primitive condition have not been available for this study.

However Taylor closely relates the outer aragonite Prismatic Shell Layer of the Unionoidea to the outer calcite Prismatic Shell Layer of the Pterioidea.

The outer aragonite Prismatic Layer of *V.ambiguus* appears likely to bear the same relationship to the underlying Outer Nacreous Layer in *V.ambiguus* as does the inner aragonite prismatic layer of a cultured pearl to the overlying nacreous layer. That is, it is intimately associated with it, and structurally determined by similar scleroproteins from largely the same source as the adjacent nacreous layer Organic Matrix scleroproteins. As will be seen, it seems likely that the calcitic Prismatic Layer of pearl oysters has no relationship at all to the aragonite Prismatic Layer of *V.ambiguus.* but rather is closely related to the outer non-calcareous Shell Layer of this animal. (ref. the martius yellow positive scleroproteins throughout the taxa studied)

6.1.12Shells and Mantles in the Nuculoidea.

Nucula superba / Nucula sp.

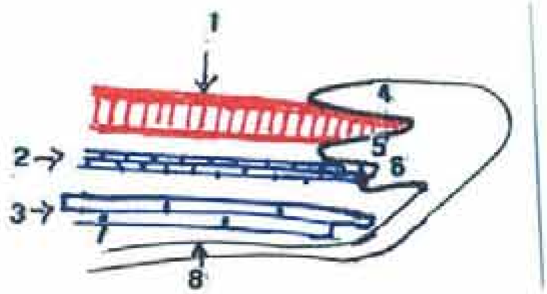


FIGURE 6.33 1 = trilaminate Composite Prism Layer from "Groove F2F3.". 2 = Outer Nacreous Layer from "Groove F1F2." (aragonite). 3 = Inner Nacreous Layer from Pallial Mantle (aragonite). 4 = "F3.". 5 = "F2.". 6 = "F1.". 7 = Pallial Space. 8 = Pallial Mantle.

In the Nuculoideans *Nucula superba* and *Nucula sp.* the shell/mantle configuration is unique amongst the Bivalves studied in that the Mantle Margin is recurved so that, of the three Marginal Mantle folds "F3". is lateral and "F1." medial. As in Chapter 5, the nomenclature of the parts of the Mantle Margin will be as for the rest of the Bivalvia so as to preserve the relationship between nomenclature and histology, but all Mantle Margin Nomenclature will be in inverted commas to signify that eg. in these cases "LatF2." is in fact on the medial side of F2..

It is also unique amongst the genera studied in that there is no shell - mantle junction at the Pallial Line but there are two shell - mantle junctions at the Mantle Margin - the forming Composite Prismatic Layer remains within the recurved Groove F2F3. throughout the animal's life and the forming Outer Nacreous Layer appears to remain adherent to the notch like Groove F1F2. which produces it. The outer Nacreous Layer separates from the Inner Nacreous Layer with acid decalcification, the former growing progressively thicker from the Hinge to the Ventral Margin of the Valve and the latter vice versa. In no other animal studied are the origins of the Shell Layer Organic Matrices precursors as certain as they are in these animals. From serial sectioning the animals and their decalcified shells, the following tissues and forming surfaces of Shell Layer Organic Matrices are seen to be coextensive peripherially:

"LatF3." and the lateral lamina of the trilaminate outer Shell Layer (i.e. the lateral lamina of the Composite Prism Layer);

"Proximal MedF2" and the underlying subepithelial glands and the middle striated lamina of the trilaminate outer Shell Layer (i.e. the middle lamina of the Composite Prism Layer);

"Distal MedF2" and the medial lamina of the trilaminate outer Shell Layer (i.e. the medial lamina of the Composite Prism Layer);

"Groove F1F2' and the Outer Nacreous Layer;

Pallial Mantle and the Inner Nacreous Shell Layer.

Further the nacreous Lateral Denticles are closely approximated to an epithelium and subepithelial glands very similar to the External Mantle Pallial epithelium and glands, which secrete the precursors of the Inner Nacreous Shell Layer. Further still, each of the dramatically differently staining parts of the hinge is closely applied to a morphologically different secretory epithelium.

All three lamina of the Composite Prism Layer after acid decalcification stain amber with Mallory's, yellow with M.S.B. and A.B./M.S.B. and red with azan and Steedman's and thus are the martius yellow positive scleroproteins of this species.

6.1.13 Shells and Mantles of the Mytiloidea.

Trchomya hirsuta, *Lithophaga teres* and *Botulopa silicula infra*.

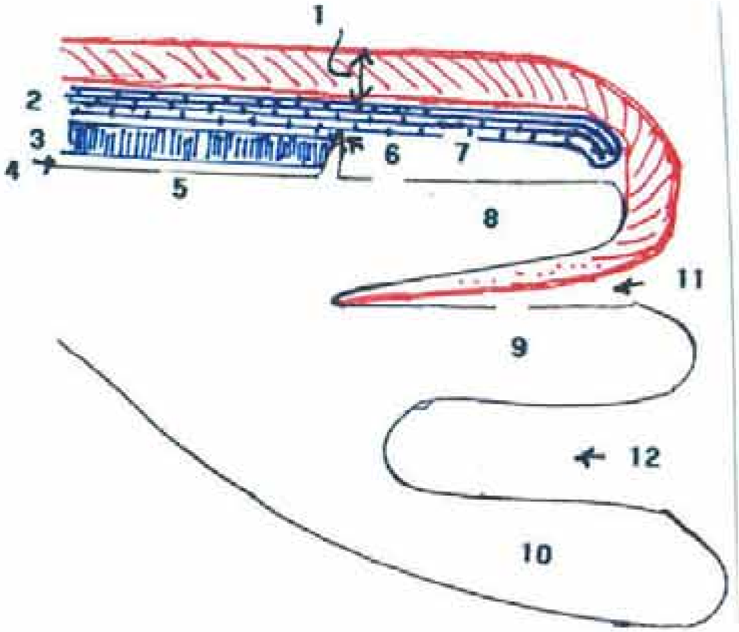


FIGURE 6.34. Shell and Mantle of *Lithophaga teres* 1 = Mytilid Prism Shell Layer (Calcitic), or trilaminate non-calcified outer Shell Layer. 2 = Outer Nacreous Shell Layer. 3 = Inner Nacreous Shell Layer. 4 = Pallial Space. 5 = Pallial Mantle. 6 = Pallial Line. 7 = Extra-pallial Space. 8 = F1.. 9 = F2.. 10 = F3.. 11 = Groove F1F2.. 12 = Groove F2F3..

Taylor (1972) lists two conditions of the shells of the Mytiloidea - a three layered shell consisting of an external Shell Layer of Mytilid Prisms outside an Outer and Inner Nacreous Layer, and, alternatively, a condition which he says is found in some tropical members of the Superfamily of a two layered shell of Outer and Inner Nacreous Layers from which the Outer Mytilid Prismatic Layer has been lost.

The three species of the superfamily studied here were *Trichomya hirsuta*, *Lithephaga teres* and *Botulopa silicula infra*. As the aim was to prepare decalcified shells and External Mantle and Mantle Margin tissues *in situ* for L.M. it is not known which of the two conditions as described by Taylor were present in these animals. What was present in all three species was a dense trilaminate outer Shell Layer, elaborated by the secretory tissues of Groove F1F2. and recurving around terminal F1.. There is also a junction of medial valve surface to External Mantle at the Pallial Line hence both the Pallial Space and the Extrapallial Space are discrete, enclosed spaces.

Especially in *Lithophaga teres* the exclusivity of these spaces is reflected in distinctly different, associated Shell Layers. The Inner Nacreous Layer, which is coextensive with the Pallial Space displays a structurally different Nacreous Layer Organic Matrix from that of any other studied - the decalcified Shell Layer, while staining similarly to all stains used as any other Nacreous Shell Layer Organic Matrix, morphologically consists, in radial section, of fine structures normal to the Shell Layer surface and hence normal to the usual direction of Nacre Sheets. As above, this Shell Layer terminates peripherally at the Pallial Line.

Beyond this and hence forming the lateral surface of the Extrapallial Space is the Outer Nacreous Layer which, in sharp contradistinction to the Inner Nacreous Layer, has a decalcified Shell Organic Matrix which under L.M appears structurally indistinguishable from the Nacreous Organic Matrices of other Superfamilies studied - i.e., Nacre Sheets about parallel to the surface of the Shell Layer.

Lateral to the Outer Nacreous Layer in all three species of the Mytiloidea studied is a complex trilaminate outer Shell Layer. In each species the outer lamina (lateral on the shell surface) appears to be secreted by an Apical Channel at the proximal extremity of Groove F1F2.; The middle Lamina appears to be added by MedF1.; and the medial Lamina by the elongate columnar epithelium of Terminal F1.. This epithelium is separated from the elongate columnar epithelium of the Mantle Edge Gland of LatF1. by a deep notch. All three laminae of this outer Shell Layer stain amber-brown with Mallory's, yellow with M.S.B. and A.B./M.S.B. and red with azan and Steedman's, and hence are considered martius yellow positive proteins in these species.

6.1.14. Shells and Mantles in the Ostreoidea.

Hyotissa hyotis

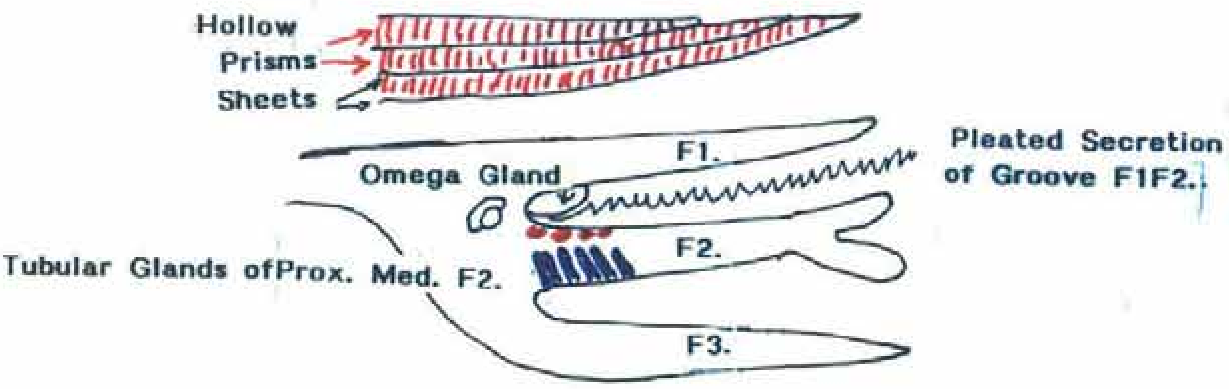


FIGURE 6.35.

Ostrea sp.

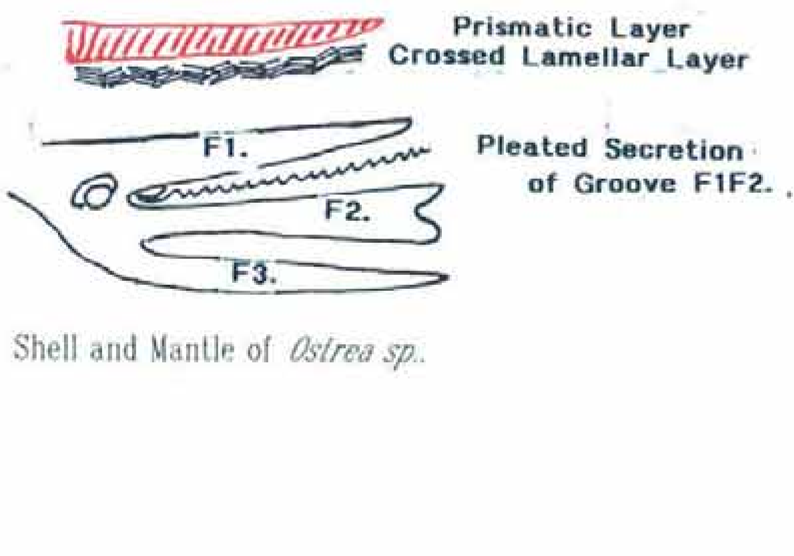


FIGURE 6.36. Shell and Mantle of *Ostrea sp.*

Saccostrea echinata

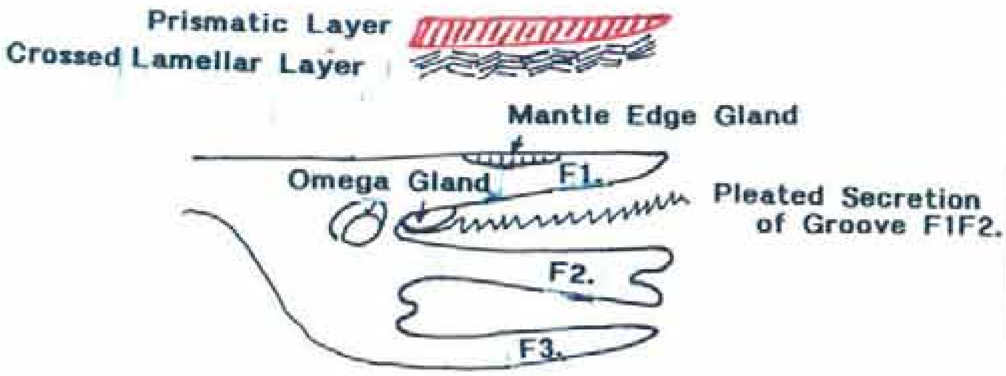


FIGURE 6.37. Shell and Mantle of *Saccostrea echinata*.

Saccostrea cuccullata

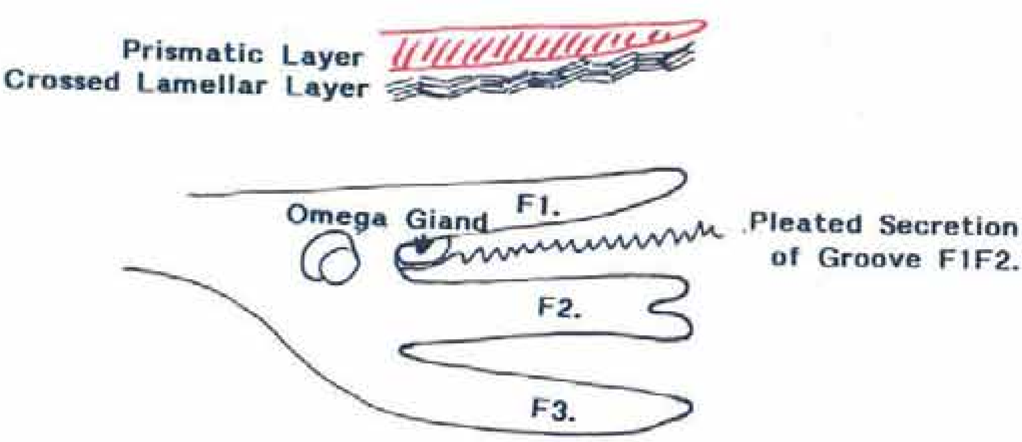


FIGURE 6.38. Shell and Mantle of *Saccostrea cuccullata*

The Ostreoidea, Anomioidea, Pectinoidea and Pterioidea differ from all other bivalves studied in that there is no attachment between shell and Mantle either at a Pallial Line or at the periphery. As above, the Pallial Space and Extrapallial Space are therefore not physically defined by tissue /shell junctions .

The members of the Ostreoidea investigated were the pycnodontid *Hyotissa hyotis* an *Ostrea sp., Saccostrea echinata* and *Saccostrea cucculata*. Of these, according to Taylor, (1973) the Pycnodontidae have a valve consisting only of Foliated Structure. This is not the case with the Pycnodontid examined. The valve of *Hyotissa hyotis*, after drying, floats in sea water because it is composed of alternating sheet like calcified Shell Layers and layers of hollow prisms with long axes normal to the valve surface and transverse parallel side walls as in the Prismatic Layer of the Pterioidea. As in Chapters 3, (Classification), and 5, (Results) the anatomy and histology of the External Mantle and Mantle Margin of especially this species, (but also the other members of the Ostreoidea studied), so closely resembles that of the Pterioidea as to cogently suggest their reclassification towards the Pterioidea and away from the Pectinoidea.

The *Ostrea sp.* appears to have no prismatic layer on the valve and therefore according to Taylor, (1972) has a valve consisting entirely of Foliated Structure. *Ostrea sp.* was found to have a Mantle Margin with three folds and a Pleated Secretion issued from the Apical Channel of Groove F1F2.. That LatF1. bore a well defined Mantle Edge Gland and MedF1. a highly differentiated secretory structure, while LatF2., Med F2. and LatF3. were largely undifferentiated is in keeping with the relatively simple shell structure in this animal (Taylor, 1973).
According to Taylor *Saccostrea echinata* has a lower (left) valve without prismatic layer and an upper (right) valve with an outer Prismatic Layer. In *Saccostrea cucculata* the outer Prismatic Layer is present on both the upper (right) valve and the posterior part of the lower (left) valve, which part is normal to the surface of attachment.

Saccostrea echinata has highly differentiated secretory structures in both the Pallial Region and the three Marginal Mantle Folds. By far the most striking secretory structure of the Mantle Margin is the dense mass of secretory glands in the Subepithelium of MedF1.. In contradistinction to *Ostrea sp.* Proximal MedF2. and Proximal MedF3. are highly differentiated glandular regions. Again a large Circum-pallial Nerve is intimately associated with the secretory strucutres.

The Anterior left (lower) Mantle Margin of *Saccotrea cucculata* which secretes a shell allegedly lacking an outer Prismatic Layer (Taylor, 19773) is dramatically different from either the right (upper) Mantle Margin or the Posterior left (lower) Mantle Margin which do secrete shells with an Outer Prismatic Layer. Part of the difference in the differentiation and distribution of secretory glands and tissues is in those of MedF1.

In summary, the degree of differentiation into secretory tissues and the abundance or otherwise of secretory glands on the External Mantles and Mantle Margins of the species of the Ostreacea studied closely reflected the degree of morphological complexity of their valves. In particular, where a Mantle Margin was associated with a valve with a well developed Prismatic Layer, it bore highly differentiated secretory tissues and abundant secretory glands on LatF2., MedF2. and LatF3. - that is on those Regions of the Mantle Margin medial to the Pleated Secretion of Groove F1F2. and lateral to Terminal F3.. Where a Prismatic Layer was lacking on the valve, the associated Mantle Margin had a largely undifferentiated mucosa on LatF2., MedF2. and LatF3. and inconspicuous secretory glands in these Regions.

Attempts to decalcify the valves of the Ostreacea to yield Shell Organic Matrices for histological study proved abortive, so that no secreted scleroprotein was found in these animals with similar staining affinities to the martius yellow positive scleroproteins of the other groups. To pursue the matter further was outside the scope of this investigation.

6.1.15.Shells and Mantle in the Anomioidea.

Placuna placenta.

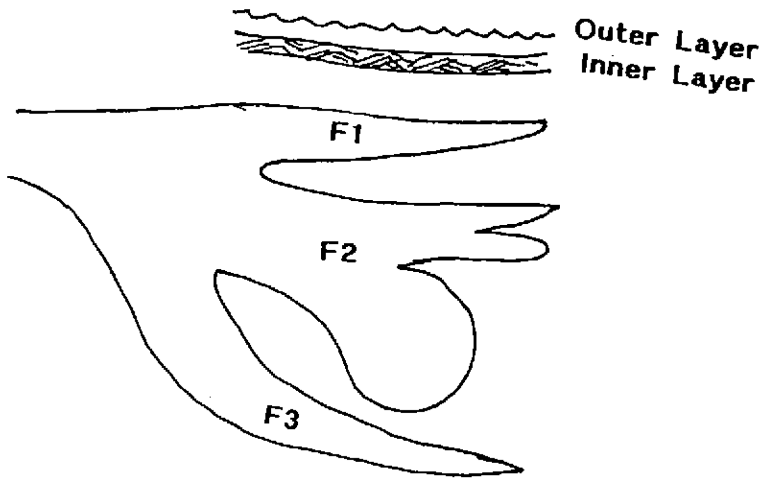
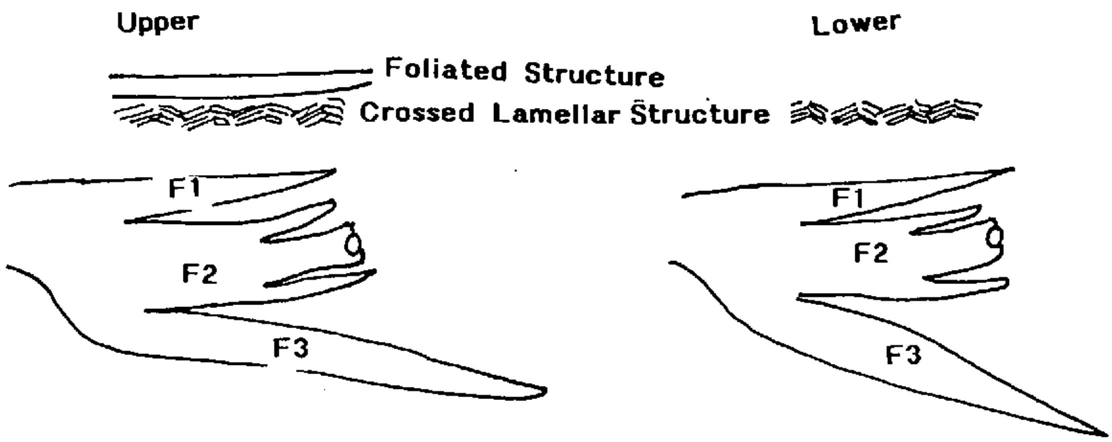


FIGURE 6.39. Shell and Mantle of *Placuna placenta*

The Left and Right Mantle Margins of *Placuna placenta* each appear to consist of three Marginal Mantle Folds with F2. apparently trilobate. There is no immediately apparent differences between the secretory structures of the two mantles and the shells they secrete are structurally very similar consisting of Foliated Structure outside Crossed Lamellar Structure (Taylor, 1973).

6.1.16.Shells and Mantles in the Pectinoidea.

Amusium pleuronectes, Spondylus lamarcki



6.40. Shells and Mantles in *Amusium pleuronectes* and *Spondylus lamarcki.*

As with *Placuna placenta* the Pectinoidea studied, the scallop *Amusium pleuronectes* and *Spondyles lamarcki* have three Marginal Mantle Folds on both upper and lower mantles and in all cases F2. is multilolate bearing such structures as highly developed eyes. However, whereas similar shell structures of Foliated Structure lateral to Crossed Lamellar Structure (Taylor, 1973) in both valves of *Spondylus lamarcki* suggested the similar degree of complexity of secretory strucutres found in the left and right Mantle Margins, the distinctly different left and right valves of *Amusium pleuronectes* was not reflected in obvious differences in secretory differentiation of the Pallial Mantle or Mantle Margin in these animals. The study of the relationship between Mantle secretory structures and Shell Layer organic Matrices in these animals is complicated by both the lack of physically defined compartments within which the Shell Layers are generated and the complexity of the Mantle Margin of these actively swimming bivalves. To pursue the relationship further was outside the scope of this study.

6.1.17.Shells and Mantles in the Pinnoidea.

Pinna bicolor.

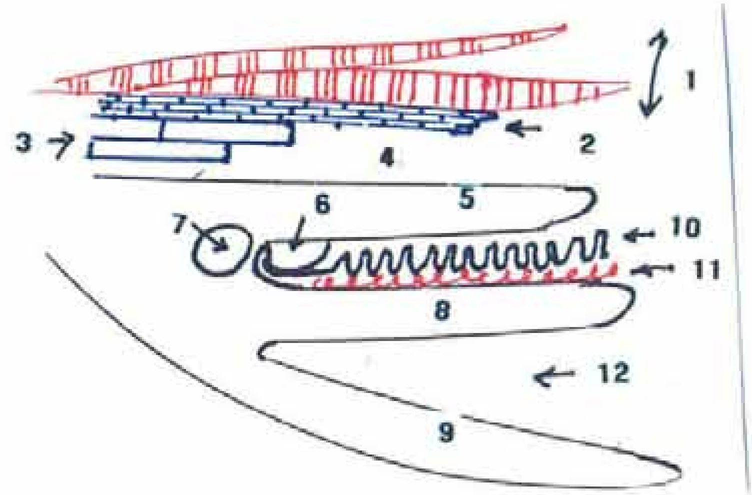


FIGURE 6.41. *Pinna bicolor.* 1 = calcitic Prismatic Shell Layer laid down in Growth Scales. 2 = Outer Nacreous Layer, (aragonite). 3 = Inner Nacreous Layer, (aragonite). 4 = Potential Extra-pallial Space. 5 = F1.. 6 = Omega Gland. 7 = Circumpallial Nerve. 8 = F2.. 9 = F3.. 10 = Pleated Secretion of Groove F1F2.. 11 = Vesiculate Secretion of Groove F1F2.. 12 = Groove F2F3.

According to Taylor (1973) the Pinnoidea and Pterioidea both have an outer calcitic simple Prismatic Layer outside an Outer Nacreous Layer and an Inner Nacreous Layers. Of the twenty eight Superfamilies of bivalves whose shell structures are listed in his work they are the only two with this arrangement of Shell Layers.

As with the Pterioidea, the calcitic Prismatic Layer of the Pinnoidea consists of Growth Scales superimposed on each other medially with the periphery of each Growth Scale formed, at intervals going peripherally, into a Growth Process. These Growth Processes, again as in the Pterioidea, come to lie on the outer surface of the Valve so that those from successively more recent Growth Scales form radiating lines from the Umbonal region to the Valve periphery. The decalcified Prismatic Layer Shell Organic Matrices are morphologically very similar to those of the pearl oysters but much less robust.

As with all members of the Pterioidea studied, the Mantle Margin of *P.bicolor* bears highly differentiated secretory glands and tissues. The Mantle Edge Gland is not only highly developed as a specialised elongate Columnar Epithelium but has a relatively large number of Subepithelial Trabecular Turquoise Glands.

Distal to the part of the mantle involved in Nacreous Shell Layer production the MedF1. is virtually devoid of secretory glands.

There is however a very well developed Omega Gland which with the opposed cells of Proximal LatF2. form the Apical Channel from which issues a voluminous Pleated Secretion. This stains purple with Mallorys and turquoise turning to pink with distance from the Apex of Groove F1F2. with A.B./M.S.B.

The Circum-pallial Nerve and Circum-pallial Sinus are both closely associated with the secretory tissues of the Apical Channel.

The F2. Ramis of the Circum-pallial Nerve closely invests the secretory glands of Proximal LatF2.

The entire surfaces of LatF2., MedF2. and LatF3. are differentiated into a complex set of secretory strucutres with numerous Trabecular Turquoise Glands.

The decalcified Shell Organic Matrices of the Prismatic Layer stain amber with Mallorys. Yellow with M.S.B. and A.B./M.S.B. and red with azan and Steedman's. They therefore constitute the martius yellow positive scleroproteins of this species.

6.1.18.Shells and Mantles in the Pterioidea.

6.1.18.1. The Genera of the Pterioidea Studied.

The Genera of the Pterioidea included in this work were *Pinctada.* (the genus of the commercial pearl oysters *P.maxima* and *P.margaritifera*), *Pteria* (the genus of the commercial pearl oyster *Pteria penguin*), *Isognomon* and *Malleus.*

6.1.18.2. Shells of the Pterioidea Studied.

All the shells of the Pterioidea studied consisted of valves with three shell layers - an outer calcitic simple Prismatic Layer and two nacreous layers - the Outer Nacreous Layer and the Inner Nacreous Layer. In every specimen studied the Prismatic Layer formed the entire outer Shell Layer (except where removed) and also constituted the medial periphery.

The only Outer Nacreous Layer normally visible in a member of the Superfamily is the most distal part of the nacre on the medial surface, and on the lateral surface where erosion or abrasion of the Prismatic Layer reveals the underlying Outer Nacreous Layer.

The Inner Nacreous Layer covers the greater part of the medial nacreous surface of a pearl shell and, in a mature specimen, forms by for the greater part of the thickness of most of the valve.

PTERIOIDEA.

Pinctada maxima.

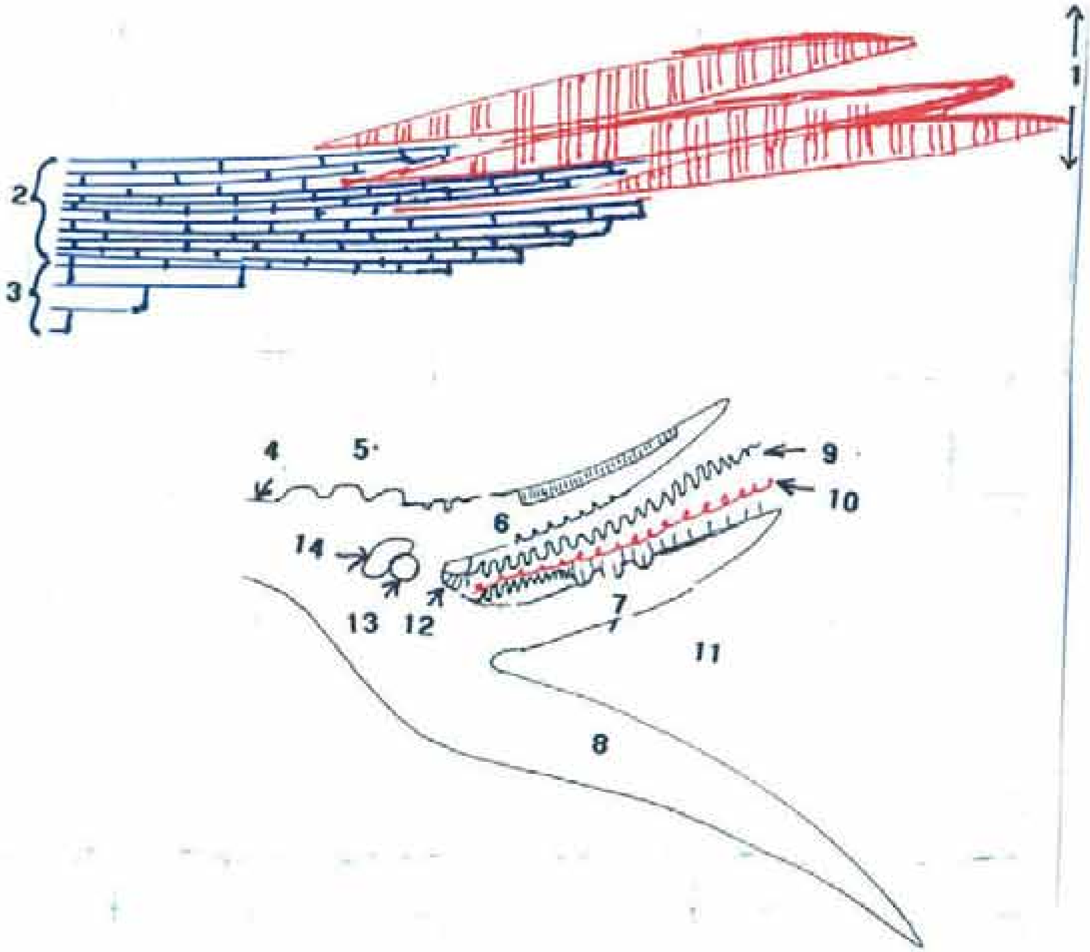


FIGURE 6.42. *Pinctada maxima* Shell and Mantle. 1 = Prismatic Layer laid down in somewhat ill-defined Growth Scales (3 shown). (calcite). 2 = Outer Nacreous Layer (aragonite). 3 = Inner Nacreous Layer (aragonite). 4 = Distal Pallial External Mantle. 5 = Distal Folded Region. 6 = F1.. 7 = F2.. 8 = F3.. 9 = Pleated Secretion of Groove F1F2. 10 = Vesiculate Secretion of Groove F1F2.. 11 = Groove F2F3.. 12 = Apical Groove F1F2.. 13 = Circumpallial Nerve. 14 = Circumpallial Sinus.

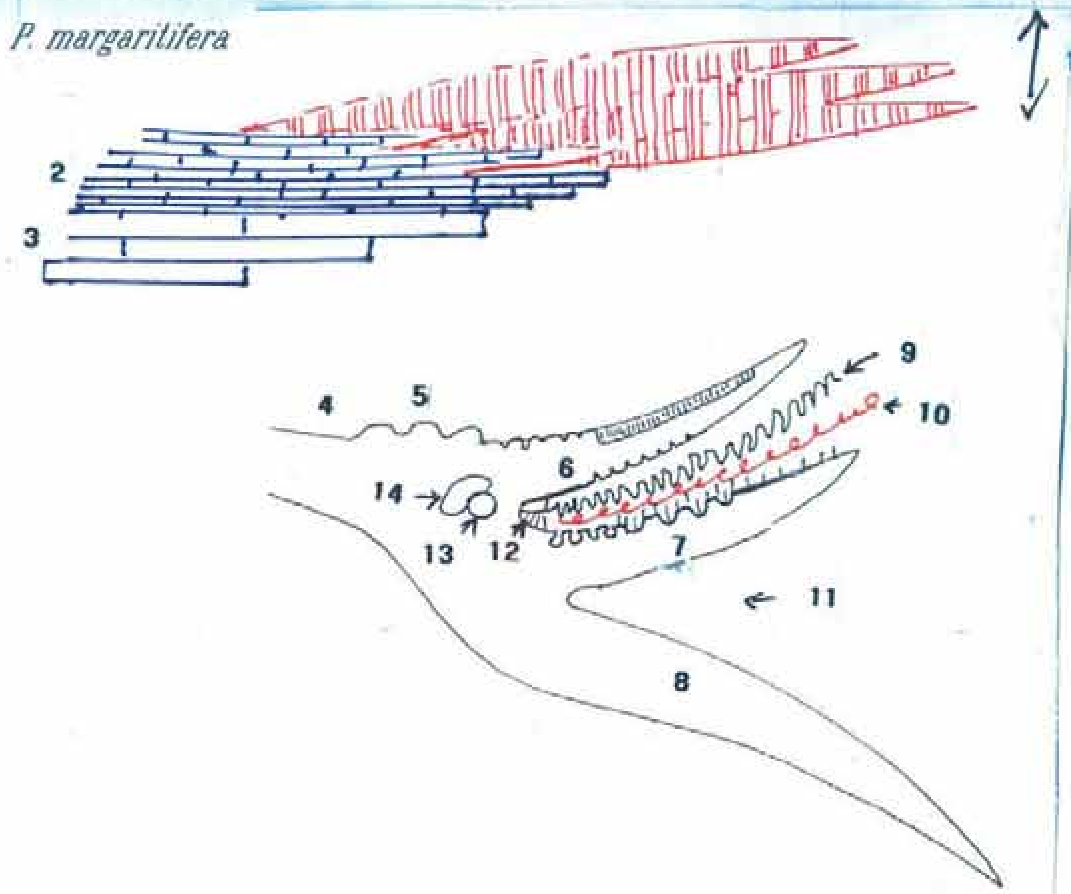


FIGURE 6.43. *P. margaritifera.* 1 = Prismatic Layer laid down in very poorly defined Growth Scales (three shown). (Calcitic). 2 - 14 = as for *P.maxima* above. Note Proximal LatF2. histological differences between these species.

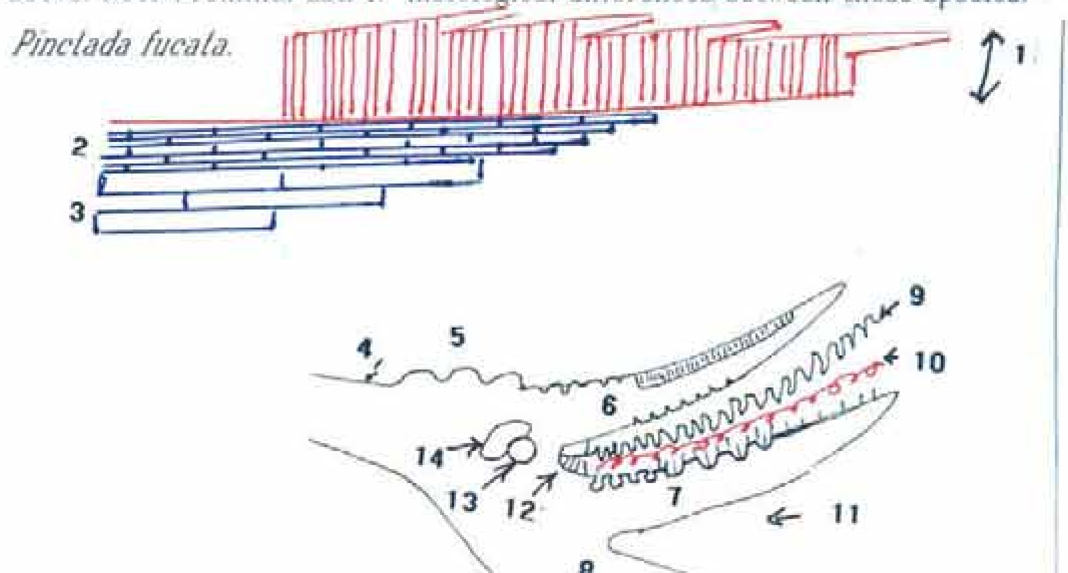


FIGURE 6.44. *P. fucata.* 1 = Prismatic Layer with no defined Growth Scales except for staining (calcite). 2 - 14 = as for *P. maxima.*

Pinctada sp. 1.

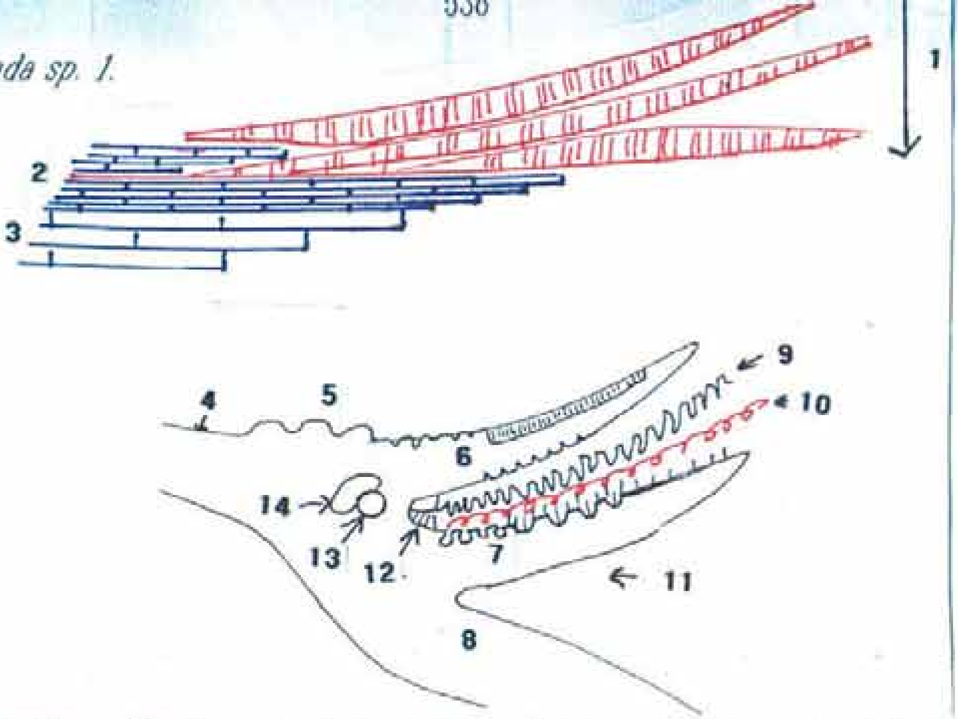


FIGURE 6.45. *Pinctada sp. 1.* 1 = Prismatic Layer laid down in well defined Growth Scales with relatively very elongate Growth Processes, three shown, (calcite). 2 -14 = as for *P. maxima* above.

Pinctada sp.2.

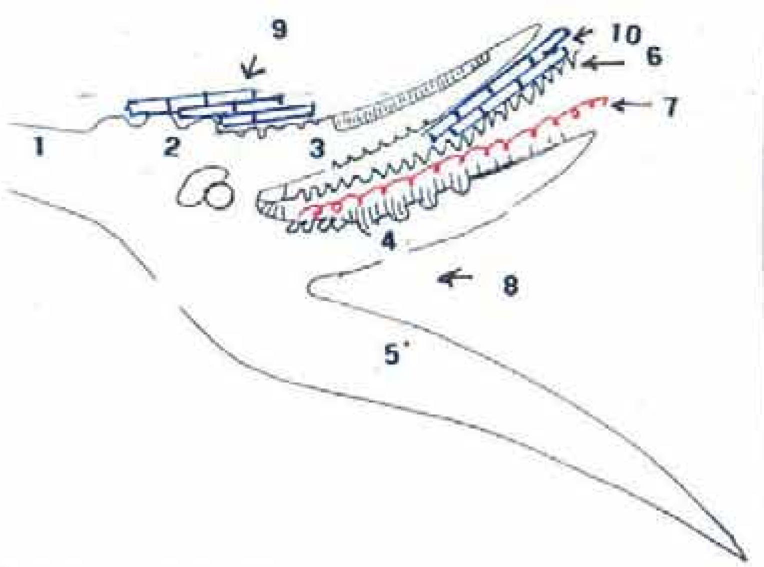


FIGURE 6.46. *Pinctada sp. 2.* 1 = Distal Pallial Region. 2 = Distal Folded Region. 3 = F1.. 4 = F2.. 5 = F3.. 6 = Pleated Secretion of Groove F1F2.. 7 = Vesiculate Secretion of Groove F1F2.. 8 = Groove F2F3.. 9 = Forming Nacreous Organic Matrices on Distal Folded Region. 10 = Forming Nacreous Organic Matrices Between MedF1. and Pleated Secretion of Groove F1F2..

Pinctada sp.3.

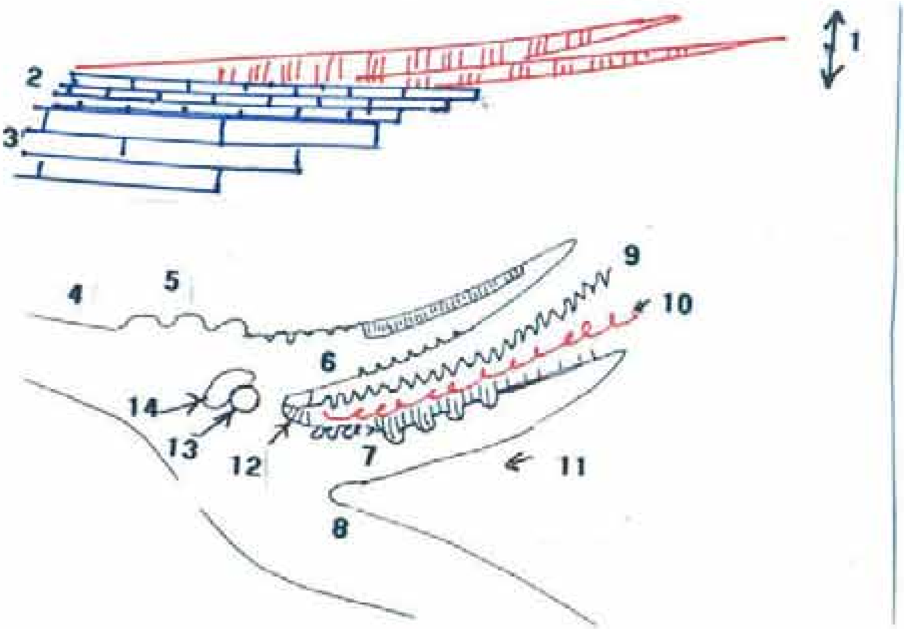


FIGURE 6.47. *Pinctada sp.3*1 = Prismatic Layer in defined Growth Scales, usually only two thick. (Calcitic). 2 - 14 = as for *P. maxima* above.

Pinctada albina sugillata

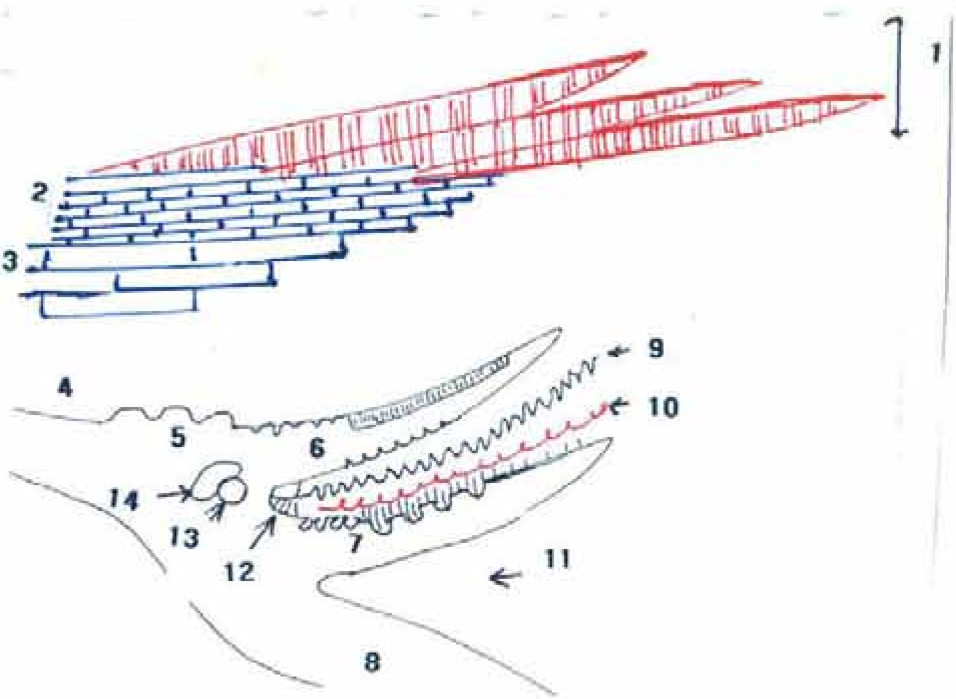


Figure 6.48. *P. albina sugillata.* 1 = Prismatic Layer in ill-defined Growth Scales seldom more than three thick. 2 - 14 = as for *P. maxima.*

6.1.18.3. External Mantles and Mantle Margins of the Pterioidea Studied.

All the Pterioidea studied have External Mantles and Mantle Margins which (apart from some ligaments near the Adductor Muscle) are free from physical attachment to the valve peripherally to the area of the Adductor Muscle.

As previously, the areas of the External Mantle have been divided into Shoulder Region, Pallial Gland Region, and the Pallial Region which is subidvided into nine regions - Anterior, Ventral and Posterior, Proximal, Middle and Distal Pallial Regions. Between the Pallial Regions and the Mantle Margin, the Distal Folded Region is distinguished. The Mantle Margin in the *Pinctada* has been divided into Folds F1., F2. and F3., and their enclosed Grooves F1F2., F2F3.. LatF1. is further subdivided into Proximal LatF1., Mantle Edge Gland, Terminal F1.. MedF1. is then subdivided into Distal, Middle and Proximal F1. An Apical F1F2. was recognised. LatF2. was subdivided into Proximal, Middle and Distal LatF2. Med F2. was subdivided into Distal and Proximal MedF2. and finally, LatF3. was subdived into Proximal LatF3. and Distal LatF3..

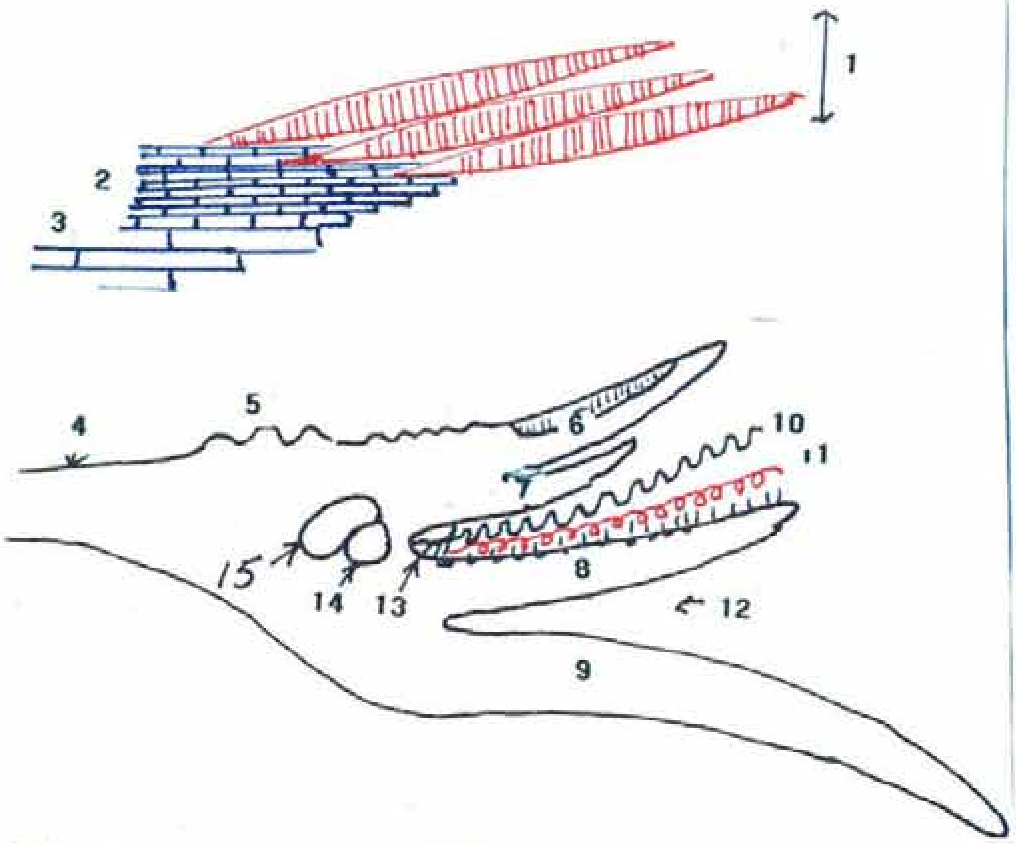


FIGURE 6.49. *Pteria, Isognomon* and *Malleus*: 1 = Prismatic Layer laid down in usually very well defined Growth Scales. 2 = Outer Nacreous Layer. 3 = Inner Nacreous Layer. 4 = Distal Pallial Region. 5 = Distal Folded Region. 6 = F1.. 7 = Ancillary F1.. 8 = F2.. 9 = F3. 10 = Pleated Secretion of Groove F1F2.. 11 = Vesiculate Secretion of Groove F1F2.. 12 = Groove F2F3.. 13 = Apical Groove F1F2.. 14 = Circumpallial Nerve. 15 = Circumpallial Sinus.

In *Pteria, Isognomon* and *Malleus* the Mantle Margin was subdivided into F1., Ancillary F1., F2. and F3 because the presence of the Mantle Edge Gland on the lateral surface of the outer Marginal Mantle Fold, and the issuance of the Pleated Secretion from an Apical Channel, (unmistakably histologically analogous to Apical F1F2. in the *Pinctada*), at the proximal apex of the Groove between the second most lateral and third most lateral Marginal Mantle Folds dictated this nomenclature.

6.1.18.4. The Inner Nacreous Layer and the External Mantle.

1. *Pinctada*.
2. *Pteria*.

From the close approximation of the External Mantle to the overlying medial surface of the Inner Nacreous Layer and the production of cultured pearls using transplants of Pallial epithelium to generate the pearl sacs there can be little argument with the proposition that the Inner Nacreous Layer is generated by the External Mantle between the Hinge and the Distal Folded Region. This relationship will be considered in greater detail later (Chapter 6.3.).

6.1.18.5. The Outer Nacreous Layer, Prismatic Layer and the Mantle Margin.

This leaves the origins of the precursors of the Outer Nacreous Layer and the various parts of the Prismatic Layer to be found amongst the secretory structures between the Distal Folded Region and the distal extremity of F3..

As with the Ostreoidea, Anomioidea and Pectinoidea the major difficulty in determining the source of the Shell Layer scleroprotein precursors of the Outer Nacreous Layer and Prismatic Layer in the Pterioidea stems from the lack of attachment and the lability of the distal Mantle and Mantle Margin in the members which were studied of this Superfamily .

The evidence presented below is both direct and indirect, positive and negative. Before proceding it is necessary to establish that in the Pterioidea there are two distinct Nacreous Layers, and this is dealt with more fully in Chapter 6.2. below.

6.1.18.6. The Outer Nacreous Layer in the Pterioidea.

Taylor, (1973) has stated that the valve in Pterioidea consisted of an outer calcitic Prismatic Layer and Outer Nacreous Layer and an Inner Nacreous Layer. Neither in the acid decalcified shell nor in S.E.M of the radial broken surface of pearl shells has any discontinuity (used in the geological sense) between the Outer Nacreous Layer and the Inner Nacreous Layer been observed. However the common names of the large *Pinctada, P.maxima* and *P.margaritifera* - gold lip pearl oyster and black lip pearl oyster respectively, indicate that the periphery of the nacre on the medial surface is differently coloured from that more proximal. That the difference is not due to the Prismatic Layer backing and the relative thinness of the peripheral part of the shell is shown by removal of the prismatic layer. This reveals that e.g., in *P.maxima* (gold lip) the gold colouration covers the entire outer surface of the Outer Nacreous Layer.

The matter is even more definate in *Pteria penguin* where the Outer Nacreous Layer is bronze coloured and strongly opalescent. In this species it fractures far more easily than the "milky" Inner Nacreous Layer and is sufficiently distinct from the Inner Nacreous Layer for the shell to be cameoed.

Under S.E.M of the medial surface of the shell periphery the Outer Nacreous Layer has edges to its Nacre Sheets which are usually a far shorter distance apart and usually more in parallel straight lines compared with the adjacent Inner Nacreous Layer. Hence Taylor's description is held to be accurate despite the lack of a dintinct line of demarcation between the two layers.

6.1.18.7 The Origin of the Organic Matrix Precursors of the Outer Nacreous Layer.

6.1.18.7.1. Direct positive evidence that the F1. fold of the Mantle Margin of the Pterioidea is involved in production of the precursors of the scleroprotein Organic Matrices of the Outer Nacreous Layer is the following:

1. Brickwork patterns of what are thought to be forming Nacre Tile Organic Matrices have been seen under T.E.M on the lateral surface of F1. in one member of one species of *Pinctada.*

2. On the same animal similar material was found on Middle MedF1. in secreted material between the surface epithelium and the Pleated Secretion of Groove F1F2..

3. In all Pterioidea studied the mature secreted material from the whole of F1. stains in the same way as does the acid resistant scleroproteins of the Nacreous Layers of the same animals.

4. The Trabecular Turquoise Glands of both LatF1. and MedF1. stain very similarly to those of the External Mantle and distinctly differently from those of F2. and F3..

6.1.18.7.2. Direct negative evidence that the quite elaborate secretory structures of F1. in the *Pinctada* and F1. and Ancillary F1. in the *Pteria* are not involved in production of scleroproteins of the Prismatic Layer is the following:

1. No mature secretory material with similar staining affinities to those of the scleroproteins of the Prismatic Layer (excluding the Intraprismatic Organic Matrix) has ever been observed in the secretions between the Pleated Secretion of Groove F1.F2 and MedF1., or on the surface of LatF1. in members of these Genera.

2. No secreted structure which appeared to be a forming or formed part of the Prismatic Layer scleroproteins has ever been observed between the Distal Folded Region and the Lateral Surface of the Pleated Secretion of Groove F1F2. in members of these Genera.

6.1.18.7.3. Indirect Positive Evidence.

In the figure giving a generalised schematic representation of formation of a bivalve shell (Figure 6.1). Wilbur and Saleuddin (1983) show an Outer Prismatic Layer being formed in the distal part of an Extrapallial Space and proximal to this a Nacreous Layer being formed in the proximal part of an Extrapallial Space.

The present study failed to reveal with certainty one example of a shell - mantle arrangement exactly like this where the Prismatic Layer was calcitic. It is not however disputed that if for the "Prismatic Layer" of Wilbur and Saleuddin "Outer Calcareous Shell Layer" is substituted and if for their "Nacreous Layer" "Middle Calcareous Layer" is substituted there may be Heterodont Bivalves which form their shells according to this scheme, i.e. with the Shell Organic Matrtices of the outer two calcareous Shell layers being produced in the Extra-pallial Space.

In the Arcoidea and Limopsoidea the secretion which begins contiguous with a glandular structure like the Omega Gland of the Pterioidea and the Proximal MedF1. continues distally as a sheet of secreted material, recurves around Distal F1. and joins the Valve inside the Outer Shell Layer or as the medial laminate of a trilaminate outer Shell Layer - which is not absolutely clear in some acid decalcified shells - but probably largely the latter. This secretion and the Inner Lamina of the trilaminate Outer Shell Layer stain various shades of blue and purple with all stains used depending on species and stain. Whether secretory material accruing to the inside of this secreted sheet as it passed the secretory strucutres of MedF1. and terminal F1. becomes part of the inner lamina of a trilaminate Outer Shell Layer or part of the Organic Matrix of the outer of the two calcareous layers of the shells of members of these superfamilies is unclear. The Complex Crossed Lamellar Layer of the Arcoidea and Limapsoidea is unquestionably secreted by the "Beeswax Glands" of the enclosed Pallial Epithelium. Just as certainly, the Crossed Lamellar Structure of the outer calcareous Shell Layer is secreted between the Pallial Line and the Apex of Groove F1F2. and in all probability except for perhaps an outer layer, almost entirely by the copious and varied secretions of the highly differentiated secretory tissues of LatF1..

(The Outer Shell Layer (or middle lamina of a trilaminate Outer Layer) is secreted exclusively by tissues Medial on the Mantle Margin to the tissues which produce the blue staining secretion described above. This will be described more fully when considering Prismatic Layer formation in pearl oysters, Chapter 6.1.18.8. and Chapter 6.6.).

Thus in these Superfamilies, the two inner lamina of the trilaminate outer Shell

Layer are a product of LatF2., (middle lamina) and apical Groove F1F2. (inner Lamina); the Inner Shell Layer is the Product of the External Mantle in the Pallial Region; and the outer calcareous shell layer is produced by LatF1. with perhaps a contribution from MedF1..

In the Unionoidean *Velesunio ambiguus* Taylor (1973) lists the shell layers as consisting of an outer Prismatic Layer of aragonite simple prisms outside two Nacreous Layers. He then suggests that the simple aragonite prismatic layer is the ancestral prismatic condition to the calcitic simple Prismatic Layer of the Pinnoidea and Pterioidea.
This is disputed.
The aragonite prisms forming the so called "Prismatic Layer" of a cultured pearl (i.e., the most medial calcareous layer - here called the Radial Layer (Figure). precisely to save this confusion). have Organic Matrices indistinguishable in their colour reactions to all stains used from the Organic Matrices of the Nacreous Layer of the pearl overlying the Radial Layer. The same is true of the Organic Matrices of the aragonite Simple Prismatic Layer of *Velesunio ambiguus* and the underlying Outer Nacreous Layer. Again the same is true of the Organic Matrices of aragonitic Myostracal Prisms of the Pterioidea and those of the surrounding Inner Nacreous Layers.

As seen here the outer aragonite simple Prismatic Layer in fact invests the inverted sigmoid part of the inner lamina of the trilaminate Outer Shell Layer (non-calcareous in this species) for a short distance on either side, while its own internal organic matrices are shown by staining affinities to be closely allied to those of the Outer Nacreous Layer.

In conclusion it is here held that the simple aragonite Prismatic Layer of *V.ambiguus* is to be regarded as a variant of the underlying Nacreous Layer generated by the effect of the presence of the Inner Lamina of the Trilaminate Outer Shell Layer.

It is hence here held that the aragonite Outer Nacreous Layer including its locally altered outer aragonite prism layer is secreted perhaps entirely by LatF1. but perhaps with some elements from distal MedF1. inside the recurving Trilaminate Outer Layer which is produced in Groove F1F2..

In the nuculoideans *Nucula superba* and *Nucula sp.* the outer nacreous layer is physically distinct in the acid decalcified shell from the Composite Prismatic Layer laterally, and the Inner Nacreous Layer medially, and indisputably produced by "F1". and Groove "F1F2". (here reduced to a notch) as the immature overlying Composite Prismatic Layer (Taylor, 1973) permanently occupies Groove "F2F3". throughout the life of the animal, and the medial "Inner Nacreous Layer" is produced by the adjacent External Pallial Mantle.

Similarly in the Mytiloidea the matter is beyond dispute. The Organic Matrices of the Inner Nacreous Layer, while accurately reflecting the staining affinities of the Organic Matrices of the Outer Nacreous Layer, have a quite distinct morphology from those of the latter and terminate abruptly at the Pallial Line. The part of the Outer Nacreous Layer which projects beyond the Pallial Line, and hence forms the Outer Surface of the Extra-pallial Space, is coextensive with LatF1. There is no sign of any secretion generated by any tissue of the Mantle Margin medial to the terminus of F1. which stains blue with any stain used - in fact the Outer Shell Layer which in the mytilids studied was in each case a complex trilaminate layer has its medial lamina added at distal MedF1. and terminal F1..

6.1.18.7.4. Indirect negative evidence supporting F1. as the source of the Organic Matrices of the Outer Nacreous Layer of the Pterioidea.

Nowhere throughout the Bivalvia has any part of a Shell Layer which stains other than blue with any of the stains used been seen to be generated or likely to be generated by LatF1. - with one clear exception. The mytilid *Botulopa silicula infra* has, like the other mytilids an ornate trilaminate outer Shell Layer outside two nacreous layers. However at the anterior and posterior terminals of this bullet shaped coral boring bivalve a sheet of the inner lamina of the trilaminate outer shell layer is recurved at regular intervals so that the anterior and posterior parts of the shell display interleaving sheets of Outer Nacreous Layer and sheets of the recurved part of the Inner Lamina of the Outer Layer. From the relationship of tissue to Shell Layer it appears beyond dispute in this animal that this part of the inner lamina of the Outer Shell Layer as well as the interleaving sheets of Outer Nacreous Layer has its source in the Lateral surface of F1. This is the only instance where credible evidence has been seen in this study that one tissue may take part alternately in the production of two Shell Layers with fundamentally different staining affinities. (The alternating Organic Matrices produced by LatF1. of *Gastrochaena cuneiformis* are morphologically different but stain similarly). Because of this a role for the Mantle Edge Gland in Prismatic Layer formation in the Pterioidea in some task aligned to its simple histological structure - e.g., the production of the Inner Fibrous Sheath - is not totally ruled out. However no evidence whatever of any such activity has ever been discerned in this study.

6.1.18.8. The Source of the Organic Matrix Precursors of the Prismatic Layer.

As with the Outer Nacreous Layer, the evidence for the source of the precursors of the Prismatic Layer Scleroprotein Organic Matrices is both direct and indirect and positive and negative.

6.1.18.8.1. Direct positive evidence for the source of the scleroprotein precursors from which the Prismatic Layer Organic Matrices are synthesised.

1. Massed secretory glands which secrete into individual Receptacle Glands in the surface epithelium are a feature of the subepithelium of Proximal LatF2. of all *Pinctada* studied with the single exception of some locations on the periphery of *Pinctada maxima* which have an alternative arrangement at that locus of elongate columnar epithelium between which the subepithelial Glands discharge. The resultant secretions stain, as do a majority of the granules in the subepithelial secretory glands themselves, amber with Mallorys and red with Azan. (Very few cytoplasmic granules whether before or after their secretion, stain

yellow with M.S.B or A.B./MS.B.). That is, these secretory glands, and more importantly their secretions, have similar staining affinities to the scleroproteins of most structures forming the Organic Matrices of decalcified Prismatic Layer.

No other mature secretory product of any part of the External Mantle or Mantle Margin has the same staining affinities, with the stains used, as do the greater part of the scleroproteins of the Prismatic Layer.

2. What are unmistakably forming Outer Prismatic End Plates and their association with forming Transverse Parallel Side Wall Prismatic Layer Organic Matrix have been seen infrequently inside the Pleated Secretion of Groove F1F2, between it and Middle LatF2.

3. The proximal limit of the distorted spindle structure which describes a Growth Scale in radial section is commonly a finely tapered structure which, after further deposition of Nacreous Layer, comes to lie between two lots of Nacre Sheets. Proximal to where the Growth Scale becomes continuous there are commonly seen intermittent depositions of Prismatic Layer material. Where folding in an acid decalcified shell resulted in a slanting section through this "pre-prismatic layer" material the result was material with the same general size and the same staining attributes as material seen forming from granular secreted material between the Pleated Secretion and the Lateral surface of F2, in another pearl oyster, (*Isognomon ephippium*, Figure 5.152, b and c, and *Pteria avicula* Figure 5.149, c and d).

6.1.18.8.2. Direct Negative Evidence that LatF2, MedF2, and LatF3, of the Pterioidea are the Source of the Precursors of their Prismatic Layers.

No evidence has ever been sighted by this author for a proposed function for the complex, and accurately replicated from species to species, secretory structures of LatF2., MedF2. and LatF3. in the *Pinctada* nor the equivalent tissues in these locations in the *Pteria*, *Isognomon*, *Malleus* or *Pinna*.

No vestige of a piece of secretion or secreted material which could have any function attributed to it other than formation of the Organic Matrices of Prismatic Layer has ever been observed in these locations.

6.1.18.8.3. Indirect Positive Evidence.

1. Unmistakably the outer Composite Prismatic Layer of *Nucula superba* and *Nucula sp.* are produced by the Secretory epithelium and associated subepithelial Secretory Glands of Med"F2". and Lat"F3"..

2. Similarly the middle lamina of the trilaminate non-calcareous layer outside the outer of the two calcareous layers in the Limopsoidea and Arcoidea stains, as do the Organic Matrices of most structures of the Prismatic Layer of pearl oysters, amber and red with Mallorys and Azan respectively. This middle lamina of the Outer Shell Layer is produced by the Secretory Structures of Proximal LatF2. and recurves around Terminal F1.. Thus the most lateral lamina of the Outer Shell Layer of the species of these Superfamilies, which is refractory to staining, is necessarily generated by tissues between Proximal LatF2. and Distal LatF3.. Similarly in some members of the Veneroidea and Carditoidea and Gastrochaenoidea the outer Shell Layer is produced by the secretory strucutres of Folds F2. and F3..

6.1.18.8.4. Indirect Negative Evidence.

Where the precursors of all Shell Layers are unmistakably accounted for lateral to the apex of Groove F1F2. there is either a reduction of F3. coupled with lack of differentiation and development of secretory structures of LatF2. and MedF2. or development of these tissues for some purpose other than shell production.

In the Corbiculoidea, Mactroidea, Solenoidea and Tellinoidea where the outer Shell Layer, whether calcareous as in the Tellinoidea or non-calcareous as in the remainder, is produced unmistakably by structures of Apical Groove F1F2. and those lateral to this, and the forming Organic Matrices of the Outer Shell Layer recurve around Terminal F1. the Mantle Margin is reduced to two folds - F3. does not exist and LatF2. and MedF2. have little apparent secretory function.

In the unionoidean *Velesunio ambiguus* Fold F3. is reduced physically and apparently devoid of specialised secretory strucutres. Proximal LatF2. may make a small contribution to the outer surface of the forming outer lamina of the Trilaminate outer layer, but this is uncertain.

In the mytiloidean *Trichomya hirsuta* the ornate secretory developments of F2. and F3. and the appearance of the parenchyma of these folds as inflatable tissues, (like F3. in the *Pinctada* they are spongiform with internally discharging secretory glands), together with the fact that the secretions of the surface secretory structures are shades of pink, red and purple with all stains used, all tend to suggest that these are structures associated with the production of the outer Shell Layer protrusions from which the specific name derives. The highly differentiated and ornate secretory structures of Fold F2. and F3. of the rock-boring mytilids *Lithophaga teres* and *Botulopa silicula infra* appear to be associated with acid secretion as a rock boring mechanism and the protective neutralisation of these secretions and not to be associated with shell production.

The mechanisms of production of the shells and more accurately of the Shell Layers in the Anomioidea and Pectinoidea have not been clarified by this study. The matter is inherently difficult for the same reasons that the Pterioidea presented a problem - lack of obvious enclosed spaces for production of precursors of the different Shell Organic Matrices. Here the matter is further complicated by the development of eyes and other sense organs on the multilobate F2. To pursue the matter further was outside the scope of this study.

N. Paspaley and D. Jackson (pers. comm.), have each independantly described the following sequence of events in pearl oysters on the ocean floor (Figure 6.50 :-

1.A pearl oyster with its valves gaping had the periphery of its right and left F3. joined in the midline with the outer surface of the Mantle Margin covered with secreted material.

2.The right and left F3. peripheries were then withdrawn from each other at fairly regular intervals around the periphery while remaining touching in between, thus forming the shapes of the Growth Processes.

3.The Mantles were then parted but the crenulated shape resulting from partial parting was maintained.

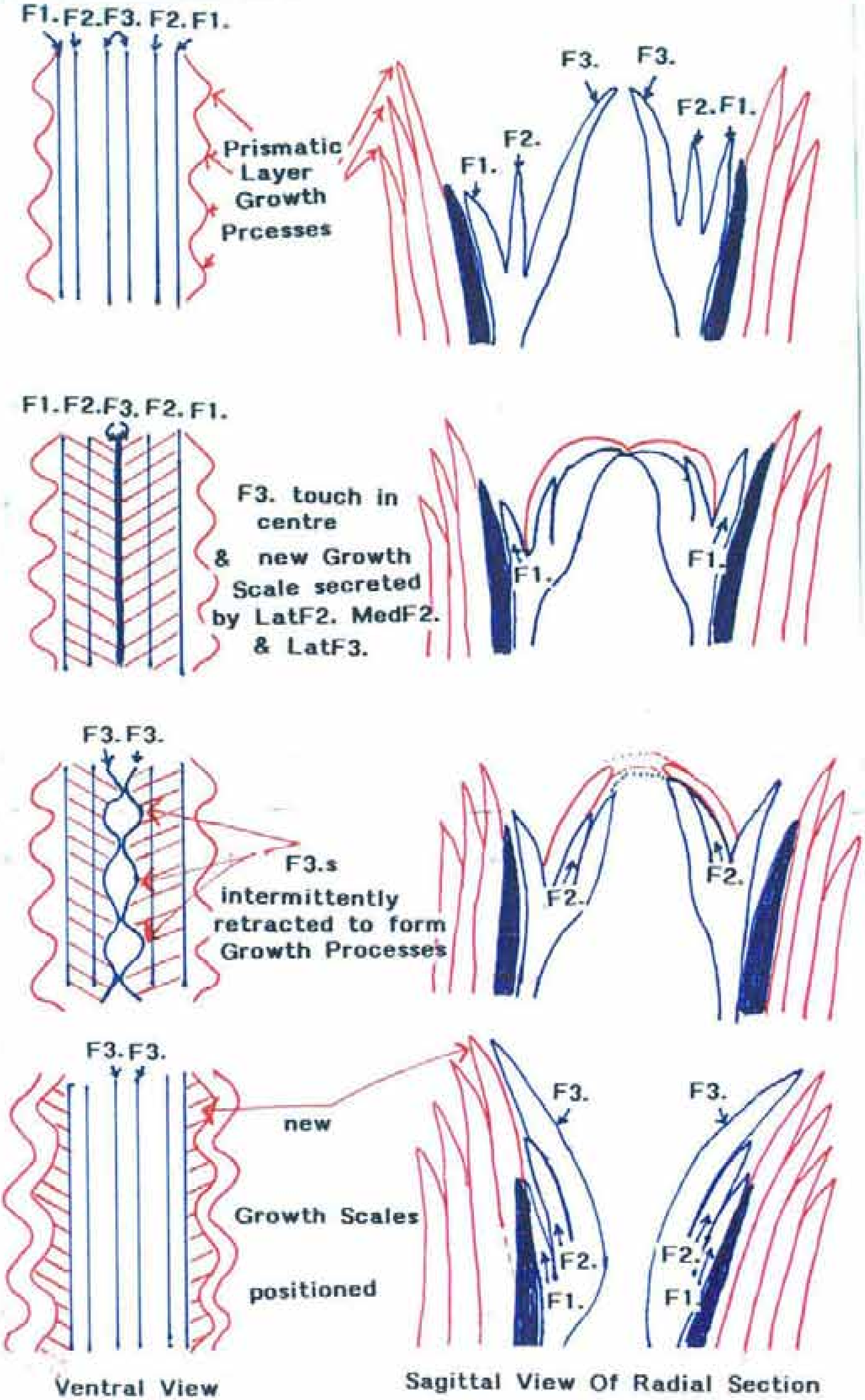


FIGURE 6.50. Diagrammatic representation of formation of Growth Scale and Growth Processes in *Pinctada*.

To finish the process of production of the organic matrices of a Growth Scale it would then only require that the Mantle Margin placed the ready-formed Growth Scale into position on the medial periphery of the extant shell. The process of sclerification may then be enzymatically completed when the Growth Scale is in position, (Waite,1983).

The Outer Composite Prism Layer of *Nucula superba* is fashioned in Groove F2F3. It abruptly changes staining affinities and morphology at the distal terminus of the Groove. This suggests that some final process of sclerification in this species is dependant on exposure to sea water and the same may be true in other species. Hence final sclerification of the Prismatic Layer Organic Matrices may also be hastened by exposure to sea water.

That some such as the sequence of events described by Paspaley and Jackson is indeed the process whereby Prismatic Layer Growth Processes are formed is made certain by the shape of the prisms near the sides of Growth Processes. While on all other surfaces they are irregularly polygonal with non-parallel sides, near the edge of the Growth Processes they are constrained to form parallelograms in the same way as if hexagonal chicken wire is stretched in one direction. The partial withdrawal of the right and left F3 peripheries must therefore be timed to occur while the scleroproteins are still sufficiently plastic for this distortion effect to occur, (*Pinctada sp.2*, 5.133,f and *g*, *Pinctada sp. 1*, 5.133, h and i).

Hypotheses concerning Prismatic Layer formation must take account of other evidence:-

Firstly, the Outer Fibrous Sheath is structurally different from the Inner Fibrous Sheath.

Secondly, the Outer Fibrous Sheath covers all lateral surfaces of the Prismatic Layer and Growth Processes - the Inner Fibroous Sheath is confined to the medial surfaces of Groth Processes and the distal medial peripheries of Growth

Scales.

Thirdly, the relative number of relatively very small prisms increases going distally.

Fourthly, the position of the underlying Major Prisms of a Major Prism Layer and that of the prisms of a Growth Process is related to the structure of the overlying Outer Fibrous Sheath. Evidence for this is:-

1. That in some species the Outer Fibrous Sheath has transverse fibres in patterns which quite accurately overlie the centres of the Outer Prismatic End Plates underneath them.

2. In other species where there are no patterns of transverse fibres, there is often a pattern of increasing and decreasing intervals between the surface "fibres" which again accurately indicates the position of the underlying prisms.

3. That in an instance where the lateral end of Major Prisms in one location had smaller cross-sectional area than the same prisms had more medially, the prisms were positioned such that the more medial parts of the prisms were norml for that species as to size and shape and relative positions.

Only two explanations of these phenomena are readily obvious. Either the Outer Fibrous Sheath in some way marks the proper location of an underlying prism which is subsequently built in the right place, or the underlying secretory tissues remain static with respect to their secretory products while they at first generate the Outer Fibrous Sheath and then the Outer Prismatic End Plate and perhaps some or all of the rest of the organic matrices of the prism.

Whatever view is taken of this last, the following seems beyond dispute.

Firstly, the Outer Fibrous Sheath of the Prismatic Layer in pearl shells is generated with what is its final lateral surface lateral - i.e., unlike the outer non-calcareous shell layers in many other bivalves it is not turned inside out by recurving around Terminal F1.

Secondly, since neither the Outer Fibrous Sheath nor the Inner Fibrous Sheath is morphologically compatiable with the Pleated Secretion of Groove F1F2., it would appear certain that this latter plays no part as Prismatic Layer organic matrix, the production of which is probably accomplished by the secretory activities of, especially, LatF2., MedF2. and LatF3. (with the possible exception of the Inner Fibrous Sheath which may derive From LatF1.. This will be discussed further below, Chapter 6.6).

In summary, in bivalves in general, the forming surface of each shell Layer is confined in an enclosed space with the secretory tissue which generates it - either in a Pallial Space, an Extrapallial Space a Marginal Mantle Groove or some such. In pearl oysters, where there is neither junction at the Pallial Line nor peripheral junction of Mantle to shell, it seems that the Distal Folded Epithelium can form a moveable outer boundary to the Pallial Space resulting in a somewhat indeterminate demarcation between the secreted precursors of the Organic Matrices of the Inner Nacreous Layer and those of the Outer Nacreous Layer; and the Pleated Secretion of Groove F1F2. forms a physical division between the secreted precursors of the Outer Nacreous Layer and those of at least the greater part of the Prismatic Layer.

No evidence is available from this work to decide between the Mantle Edge Gland and Distal LatF3. as the source of the Inner Fibrous Sheath of the Prismatic Layer except that the secretions seen on LatF1. stain differently from the Inner Fibrous Sheath in most instances.

Staining affinities suggest that the Light Blue Glands of Middle LatF2. may be one of the Sources of the Intraprismatic Organic Matrix, and if this is so, perhaps also of the Reteform Interprismatic Organic Matrix and Linear sSructure Interprismatic Organic Matrix which may be similar chemically

CHAPTER 6.2

DISCUSSION (Continued).

NACREOUS LAYERS IN THE PTERIOIDEA.

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CHAPTER 6 (2)
NACREOUS LAYERS IN THE PTERIOIDEA

6.2.1. The inner two Shell Layers of the Unionoidea, Pterioidea, Mytiloidea and Nuculoidea are Nacreous Layers (Taylor, 1973). In the Pterioidea these Nacreous Layers are not formed in discrete compartments as in the Unionoidea, Mytiloidea and Nuculoidea (Figures 5.3, 5.8 and 5.4 respectively) nor are they distinctly morphologically different as in the Unionoidea or Mytiloidea (Figures 5.1 and 5.7 respectively), nor do they separate with acid decalcification as in the Nuculoidea (Figure 5.)

6.2.2. Two factors militate against a clearly discernable boundary between the Inner Nacreous Layer and the Outer Nacreous Layer in the Pterioidea. Firstly, the intrinsic morphology of the two layers is essentially similar, and secondly, the tissue sources of the Organic Matrices of the two layers are freely mobile in the vicinity of the junction of the two layers. Considering the first of these, both Nacreous Layers in the Pterioidea are composed of Nacre Sheets which themselves are composed of Nacre Tiles surrounded by Organic Matrix.

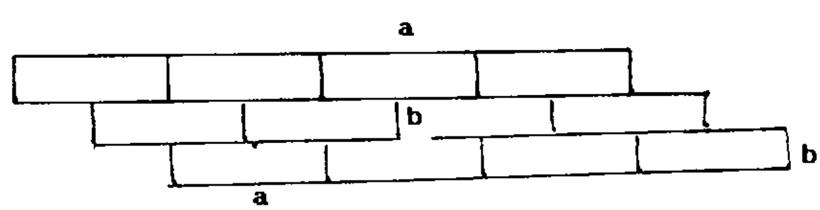

Figure 6.51. Schematic diagram of twelve Nacre Tiles in three Nacre Sheets. The Nacreous Organic Matrix between the Nacre Sheets, (a), is chemically different from that between the Nacre Tiles in any one Nacre Sheet,(b). This is the basic geometry of both Inner and Outer Nacreous Layers.

Viewed medially, the medial surface of each Nacreous Layer presents under L.M or S.E.M as a surface with linear patterns not unlike fingerprints; i.e., patterns which alter from place to place of straight, curved or anastomosing lines.

These lines are the edges of Nacre Sheets which, irrespective of the type of pattern they form, or the Nacreous Layer, are invariably so disposed that the next more lateral Nacre Sheet is more extensive in every visible surface dimension than the sheet more medial to it.

Figure 6.52. Concentric circular pattern of edges of Nacre Sheets of the Inner Nacreous Layer on left and parallel linear pattern of Outer Nacreous Layer on right, (repeat of part of Figure 2.9).

The radial broken face of the Nacreous Layers in the Pterioidea again shows no discontinuity (used in the geological sense) between the two layers. Moreover, stair patterns between Nacre Tiles in successive Nacre Sheets are the norm in both Nacreous Layers (Figures 5.29, d, 5.21, c).

6.2.3. Despite the above, as the common names of Gold Lip Pearl Oyster for one variety of *Pinctada maxima* and Black Lip Pearl Oyster for *P.margaritifera* imply, there are clear differences between the Inner Nacreous Layer and Outer Nacreous Layer in the Pterioidea. As well as the above, the difference is probably most marked in *Pteria penguin* where the Outer Nacreous Layer is noticeably more easily fractured than the Inner Nacreous Layer and the former is an opalescent bronze colour and the latter a milky white.

6.2.4. Under S.E.M the two Nacreous Layers in the Pterioidea are quite distinct. For the entire Ventral and most of the Posterior medial surface of the Outer Nacreous Layer the pattern of the Nacre Sheet edges closely approaches evenly and closely spaced parallel straight lines which are parallel to the adjacent Nacreo-prismatic Junction. (Figures 5.20 and 5.24). The adjacent pattern of Nacre Sheet edges on the medial surface of the Inner Nacreous Layer is of more widely spaced parallel straight line and curvilinear patterns. (Figure 5.19). At the dorsal half of the Anterior medial surface of the Outer Nacreous Layer the pattern of the Nacre Sheet Edges is of gently scalloped finely spaced near-parallel lines. The medial surface of the adjacent Inner Nacreous Layer shows a pattern of crooked anastomosing and discontinuous edges to Nacre Sheets characteristic of this region (Figures 5.89 and 5.90).

In some species e.g., *Pinctada maxima*, *P.fucata* and *Pteria penguin* the Nacre Tiles in the Outer Nacreous Layer are similar in habit to those of the adjacent Pallial Nacre of the Inner Nacreous Layer, but this is not the case in other species. Though the same habit, the Outer Nacreous Layer Nacre Tiles of *Pinctada maxima* are noticeably smaller than the Nacre Tiles of the adjacent Inner Nacreous Layer Pallial Nacre (Figures 5.19 and 5.20). In *P.margaritifera* the Nacre Tiles of the Inner Nacreous Layer have a diamond shaped habit and those of the Outer Nacreous Layer a truncated diamond shaped habit (Figures 5.89 and 5.90). In *Pinctada sp. 1* and *Pinctada sp. 3* the extended hexagon habit of the Inner Nacreous Layer Nacre Tiles is replaced by the regular hexagon habit of those of the Outer Nacreous Layer (Figure 5.131). The reverse is true of the habit of the Nacre Tiles in *Pinctada sp. 2* (Figure 5.133). In *Pinctada sp. 4* the regular hexagonal habit of the Nacre Tiles of the Inner Nacreous Layer is replaced by the truncated diamond shapes of the Nacre Tiles in the Outer Nacreous Layer.

Thus colouration of the Outer Nacreous Layer in some species, the changes in Nacre Tiles habit between Inner and Outer Nacreous Layer in others, and the patterns of Nacre Sheet Edges distinguish the Outer Nacreous Layer from the Inner Nacreous Layer in the Pterioidea studied.

The colouration of the Outer Nacreous Layer in *Pinctada margaritifera* is exploited in the production of so called black pearls in some Islands of the South Pacific. (Fougerouse-Tsing and Herbaut, 1994).

CHAPTER 6 (3).

DISCUSSION (Continued).

THE INNER NACREOUS LAYER AND THE ADJACENT EXTERNAL MANTLE IN THE PTERIOIDEA

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CHAPTER 6 (3).
DISCUSSION (Continued).
THE INNER NACREOUS LAYER AND THE ADJACENT EXTERNAL MANTLE IN THE PTERIACEA.

6.3.1. The Inner Nacreous Layer in the Pteriacea covers the entire medial surface of the Valve except for that covered by the Hinge, and the Adductor Muscle Scar, and peripherally, the Outer Nacreous Layer and the medial surface of the last produced Growth Scale.

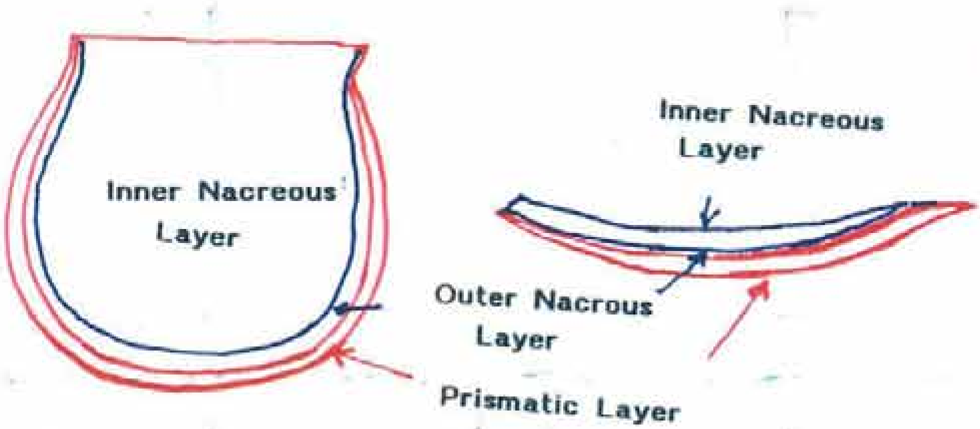

FIGURE 6.53 Schematic Diagram of medial surface of left Valve of young adult *Pinctada maxima* on left and radial section through a similar valve on right showing spatial relationships of Inner Nacreous Layer, Outer Nacreous Layer and Prismatic Layer.

6.3.2. The patterns of the edges of Nacre Sheets on the medial surface of the Inner Nacreous Layer are very similar in similar Regions not only within a species but also within the Genus *Pinctada* and, to a certain extent, throughout the Genera of the Pterioidea.

The Shoulder Region of the Inner Nacreous Layer is characterized medially by prominent and relatively frequent and extensive patterns of edges of Nacre

Sheets forming concentric circles and less frequently spirals, with intervening areas of curvilinear and parallel linear patterns (Figures 5.13, 5.14, 5.85 and 5.86).

The Pallial Gland Region Inner Nacreous Layer medial surface in the Pterioidea displays usually elongate ovoid patterns or strongly curved patterns (Figures 5.15, 5.87 and 5.131).

Generally speaking, the Inner Nacreous Layer medial surface patterns of edges of Nacre Sheets gradually become less curved with fewer anastomoses, and more closely approximate the parallel linear pattern of the Outer Nacreous Layer going ventrally and posteriorly from the Adductor Muscle Scar, (Figures 5.13 - 5.23). Conversely the Anterior Proximal and Anterior Middle Pallial Nacre usually shows a confused pattern of bent, anastomosing, crooked and discontinuous edges to Nacre Sheets (Figures 5.25 - 5.28).

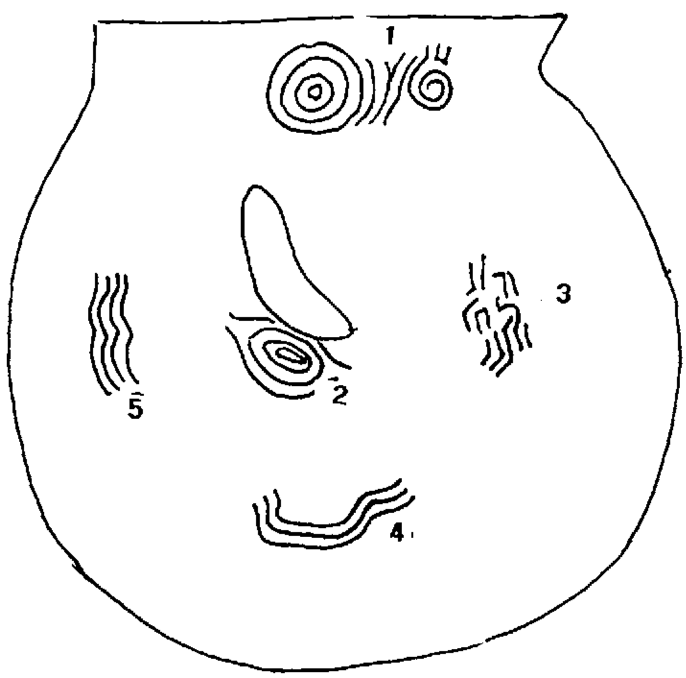

Figure 6.54. Typical *Pinctada* Inner Nacreous Layer patterns of edges of Nacre Sheets found at the indicated locations. 1 = Shoulder Region near Hinge. 2 = Pallial Gland Region. 3 = Anterior Middle Pallial Region. 4 = Ventral Middle Pallial Region. 5 = Posterior Middle Pallial Region.

An exception to the above general picture are superimposed tightly recurved tongues of Nacre Sheets projecting from the Pallial Gland Region ventrally and the Shoulder Region anteriorly and posteriorly (Figure 5.18,b and 5.28,b).

6.3.3. The edges of the Nacre Sheets represent the extent in that direction of the Nacre Tiles which lie within the fully formed Nacreous Organic Matrices of a Nacre Sheet. Beyond the edge of all Nacre Sheets are Nacre Tiles whose dimensions are comparable to those fully bound in the Nacre Sheets but are partially free of the Nacre Sheet Organic Matrices. Further away from the edge of the Nacre Sheet are Nacre Tiles which are located on the medial surface of the next most lateral Nacre Sheet, whose dimenesions diminish with distance from the ege of the Nacre Sheet. These free Nacre Tiles are not incorporated into the Organic Matrices of the Nacre Sheet although they appear to be enclosed in their own Organic Matrix. The partly bound and free Nacre Tiles are invariably more perfect specimens of the geometric shape to which they and the bound Nacre Tiles tend than are the latter (Figures 5.126,c and 5.126,k).

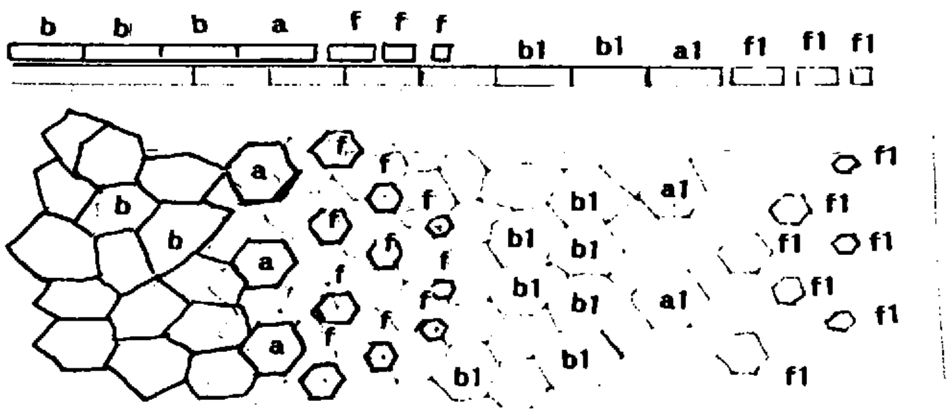

Figure 6.55. The black Nacre Sheet overlies the blue Nacre Sheet, (ie. is medial to it). Medial and sagittal views of two successive Nacre Sheets. The partly bound Nacre Tiles (a,a1) are about the same surface area as the adjacent bound Nacre Tiles (b,b1) but more perfect examples of the geometric shape to which they both tend, as are the free Nacre Tiles (f). These diminish in size with distance from the bound edge of the Nacre Sheet and usually don't occur beyond about half way between successive edges of Nacre Sheets.

6.3.4. In all species of the Pterioidea studied, the habit of the Nacre Tiles of the Inner Nacreous Layer was the same for members of the same species but in some cases differed from species to species. Thus while all other members of the *Pinctada* studied had Nacre Tiles in their Inner Nacreous Layers which tended to regular hexagons or to extended hexagons, the Inner Nacreous Layer of *P. margaritifera* studied here had diamond shaped Nacre Tiles in the Inner Nacreous Layer as had *Pteria penguin* and *Isognomon ephippium*. It is not known whether these shapes of Nacre Tiles are species specific from all areas under all conditions of normal nacre formation.

6.3.5. All Nacre Tiles forming part of any one pattern of edges of Nacre Sheets tend to not only the same size but the same orientation of the axes of their geometric shape to the edge of their Nacre Sheet. The orientation to the edge of the Nacre Sheet is usally the same whether the Nacre Tiles are free or bound. How this eventuates is not understood.

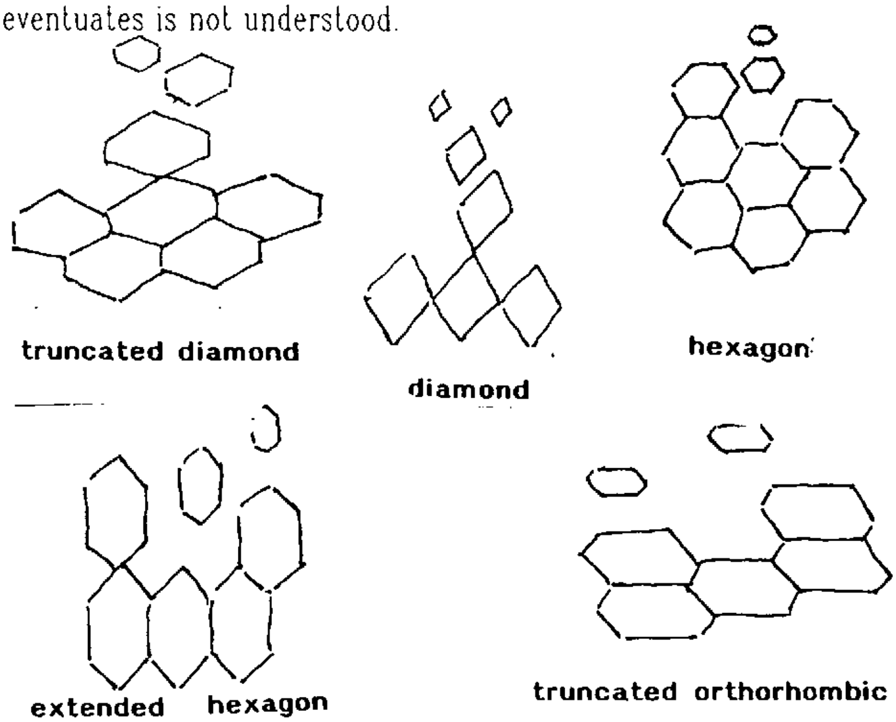

Figure 6.56. Medial view of Inner Nacreous Layers of different pearl oysters. Five different geometric forms of Nacre Tiles showing constancy of orientation of geometric axes to the direction of edge of the Nacre Sheets of both bound and free Nacre Tiles in each locus. This angle of orientation may vary from locus to locus even on the one valve.

This contrasts dramatically with the extensive spread of free Nacre Tiles over about 35 μm in width between the widely spaced tightly recurved edges of Nacre Sheets in the nearby Middle Pallial Region (Inner Nacreous Layer). (*Pictada sp.3*). Here the Free Nacre Tiles occur on about the most proximal ten, (going radially), of the medially visible Nacre Tiles of the underlying Nacre Sheet.

Similarly, where at the peripheral Nacreo-prismatic Junction the partly bound and free Nacre Tiles lie in the grooves between the medial ends of Major Prisms of the Prismatic Layer, the size of the free Nacre Tiles decreases with distance along the junctions of the Major Prisms from the bound edge of the most lateral Nacre Sheet. These phenomena will be discussed durther in Chapter 6.5.

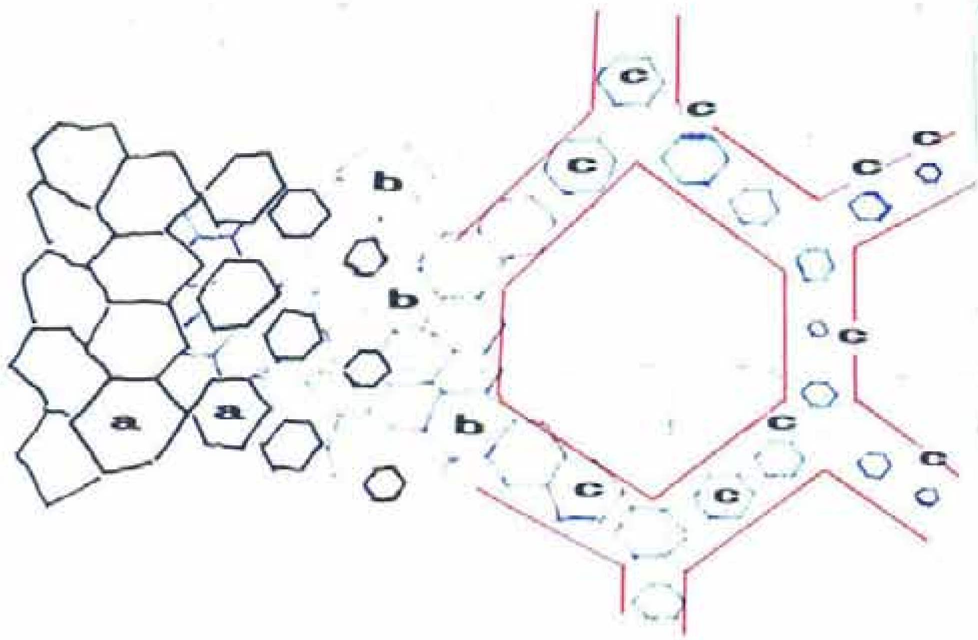

Figure 6.58. Peripheral Nacreo-prismatic Junction. Second most lateral Nacre Sheet of Outer Nacreous Layer (a) medially overlies most lateral Nacre Sheet (b). Partly bound and free Nacre Tiles of the latter lie in the grooves between the medial ends of Major Prisms of the Prismatic Layer(c). Size of free Nacre Tiles decreases with distance along the Grooves from the edge of the Nacre Sheet.

6.3.6. The sizes of bound and partly bound Nacre Tiles of Inner Nacreous Layer may remain fairly constant throughout the Regions and patterns of edges of Nacre Sheets of one specimen or may, as in some *Pinctada maxima* decrease in size from the Hinge to the periphery.

6.3.7. The Shoulder Region of the External Mantle and the Pallial Gland Region are both host to massive subepithelial glandular structures consisting of Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands. Although opposed to only about one fifth of the medial surface of the Inner Nacreous Layer in an adult pearl oyster the volume of subepithelial glandular tissue in these two areas is far greater than the combined total of that from all the remaining External Mantle. It is opposite these two Regions of the External Mantle that the concentric circular, large spiral and strongly recurved patterns of edges of Nacre Sheets are most commonly found. The significance of this will be discussed in Chapter 6.5.

6.3.8. The Shoulder Gland of the External Mantle is a fairly tightly packed mass of subepithelial glands whose lateral limits define the limits of the Shoulder Area. Beneath the surface epithelium and the subepithelial fibrous connective tissue, the space is largely occupied by the cell bodies of large Trabecular Turquoise Glands, and, deep to this, a mass of Granular Cytoplasm Secretory Glands.

6.3.9. The glands of the Pallial Gland are similar to those of the Shoulder Gland but the glandular structure of fibrous connective tissue is quite different. The functional significance of this is not clear. It may be related to the fact that the Shoulder Gland is fixed in position between the Isthmus and the Valve/Adductor Muscle junction, whereas the Pallial Gland is located on the free Mantle beyond the Adductor Muscle, or may be related to differences in secretory function.

6.3.10. The External Mantle of the Proximal Pallial Region, whether Anterior, Ventral or Posterior, is relatively poorly supplied with any secretory glands especially subepithelial Granular Cytoplasm Secretory Glands. The prevalence of all species of unicellular glands whether Trabecular Turquoise Glands or Granular Cytoplasm Secretory Glands increases going distally from the Proximal Pallial Region to the Distal Pallial Region anteriorly, ventrally and posteriorly. This is in inverse relationship to the degree of curvature generally seen in the opposing patterns of edges of Nacre Sheets in these Regions. The signifigance of this will be discussed further in Chapter 6.5.

6.3.11. The Nacreous Layer Organic Matrices residual following acid decalcification in all species of the Pterioidea studied, stain purple with Mallorys, pale mauve with M.S.B, deep blue with Azan and turquoise with A.B./M.S.B. On the surfaces of the External Mantle of all species studied there are secretions which stain in accord with the staining affinities of the Nacreous Layer Organic Matrices of acid decalcified shell. The greater part of the sheets of secretion where these are formed can be seen emanating from the Trabecular Turquoise Glands, and these glands seen under T.E.M. have a cytoplasm which consists largely of sheets of material. Under L.M. and T.E.M. these sheets of secretion from the Trabecular Turquoise Glands often have granules from the associated Granular cytoplasm Secretory Glands between them.

The entire surface of the External Mantle of all members of the Pterioidea studied is covered with apical microvilli which form vesicular enlargements at their distal ends. These bud off to form secreted vesicles. Above various tissues these vesicles at times form a secreted sheet but its function is not known.

As above, the cytoplasm of the Trabecular Turquoise Glands is refractory to all stains used with the exception of A.B./M.S.B.. It thus appears that the acid-mucopolysaccharide secretions of the Turquoise Glands after admixture with the secretions of the Granular Cytoplasm Secretory Glands, and possibly also the vesiculate secretion from the apical microvilli of the surface epithelium, form the scleroproteins of the Inner Nacreous Layer. That is that, for example with Mallory's Trichrome, the non-staining secretions of the acid-mucopolysaccharides from the Trabecular Turquoise Glands plus the magenta, purple, red and yellow staining granules secreted from the Granular Cytoplasm Secretory Glands with perhaps the addition of the vesiculate secretion from the

surface epithelium apical microvilli, generates scleroproteins, (presumably glycoproteins), which stain purple with Mallory's, (and pale mauve with M.S.B., Royal Blue with Azan, blue with Steedman's and still Turquoise with A.B./M.S.B.) as do the scleroproteins of the Inner Nacreous Layer Organic Matrices.

The morphologies of the Inner Nacreous Layer Organic Matrices will be discussed further in Chapter 6.5..

CHAPTER 6 (4).

OUTER NACREOUS LAYER ORGANIC MATRIX MORPHOLOGY AND THE MANTLE FROM THE DISTAL FOLDED REGION TO APICAL GROOVE F1.F2. IN THE *PINCTADA*.

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6.4.1. As previously, in the *Pinctada* the Outer Nacreous Shell Layer lies between the Inner Nacreous Layer and the Prismatic Layer for most of its extent, forming the medial surface of the Valve for a relatively short distance proximal to the peripheral Nacreo-prismatic Junction. (Figures 5.20 and 5.87).

6.4.2. On the medial surface, the edges of the Nacre Sheets of the Outer Nacreous Layer are arranged in a gently scalloped pattern anteriorly, and as closely spaced parallel line patterns ventrally and posteriorly. These parallel line patterns are about parallel to the adjacent Nacreo-prismatic Junction.

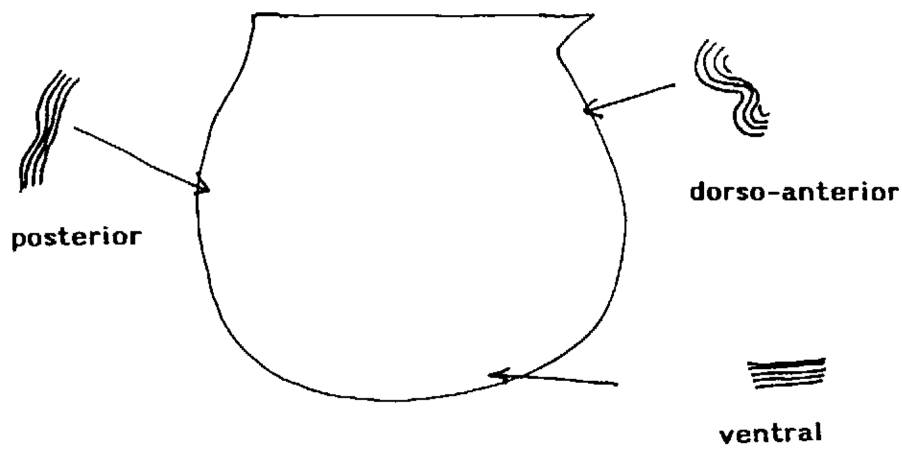

Figure 6.57. Schematic diagram of patterns of edges of Nacre Sheets of the Outer Nacreous Layer at the Dorso-anterior, Ventral and Posterior margins of the valve of a pearl oyster, Genus *Pinctada*.

6.4.3. In common with all patterns of edges of Nacre Sheets the parallel linear patterns of the Outer Nacreous Layer have both partly bound and free Nacre Tiles which accurately display a structural habit tended to by the adjacent bound Nacre Tiles. Further, the free Nacre Tiles which rapidly decrease in size with radial distance from the edge of the Nacre Sheet, are positioned on the junctions of the underlying Nacre Tiles over about the proximal half of the distance between the edges of the Nacre Sheets. Thus where these edges are about four Nacre Tiles apart in the Outer Nacreous Layer almost all the free Nacre Tiles occur on the two most proximal visible Nacre Tiles of the underlying Nacre Sheet. (*Pinctada sp. 3* Plate 8).

6.4.4 Wheras in some species e.g., *P.maxima*, *P.fucata* and *Pteria penguin* Nacre Tiles in the Outer Nacreous Layer are similar in habit to those of the Pallial Nacre of the Inner Nacreous Layer, this is not so in other species. In *Pinctada margaritifera* the Nacre Tiles of the Inner Nacreous Layer have a diamond shaped habit and those of the Outer Nacreous Layer a truncated diamond shaped habit. In *Pinctada sp.1* the extended hexagons of the Inner Nacreous Layer change to the regular hexagons of the Outer Nacreous Layer. In *Pinctada sp.2* the regular hexagonal habit of the Inner Nacreous Layer is replaced by the extended hexagonal habit of the Outer Nacreous Layer and the reverse is true in *Pinctada sp.3*. In *Pinctada sp.4*. the regular hexagons of the Inner Nacreous Layer are replaced by the truncated diamond shapes of the Nacre Tiles in the Outer Nacreous Layer.

6.4.5. As previously, the outer Nacreous Layer of *Pinctada margaritifera* is coloured and the common name of the species - black lip pearl oyster -derives from that fact. The cultured pearl industry of some South Pacific Islands is based on the production of so-called black pearls using this species. That part of the Mantle used for the tissue implant is taken from between the Distal Folded Region and the Mantle Edge Gland, (Fougerouse-Tsing and Herbaut, 1994).

6.4.6. As will be discussed more fully later, (Chapter 6.6.), data presented in this thesis supports the propositions that the Pleated Secretion of Groove F1F2. of the Pterioidea is not involved as a structural material in Shell Layer Formation, but acts as an extensible physical barrier between the products of the secretory tissues and glands lateral to it and those medial to it. Since what is thought to be forming Nacreous Organic Matrix has been observed under L.M and T.E.M in one species of *Pinctada* (*Pinctada sp. 2* Figures 5.135 and 5.136) covering the LatF1. in the folds of the Distal Folded Regions and the proximal part of F1., as well as between MedF1. and the Pleated Secretion of Groove F1F2., the tissue sources of the precursors of the Organic Matrices of the Outer Nacreous Layer are therefore likely to lie between the Distal Folded Region and Apical Groove F1F2..

6.4.7. As with the elongate columnar epithelium of the Isthmusistic epithelium, the Turquoise Glands and Granular Cytoplasm Secretory Glands of the Shoulder Region, those of the Pallial Gland Region, those of the Proximal, Middle and Distal Pallial Regions, the specialised structures of the Apical Groove F1.F2., and the Prismatic Layer Organic Matrix secretory structures of F2. and LatF3., the secretory structures of F1. are precise, and while recognisably similar throughout the *Pinctada* are yet specific to both a species and the position on the periphery of the Mantle Margin.

However whereas there is little doubt re the function at least in broad outline of other areas of the periphery and in most cases of the tissues on the various parts of the periphery, which of the tissues of F1. are engaged in Outer Nacreous Layer Shell Organic Matrices production are simply not known.

To recapitulate, the Proximal part of LatF1. in the *Pinctada* is a folded epithelium, with lower folds than the Distal Folded Epithelium and lacking its elaborate sinus system but with Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands in the subepithelium. The major histological feature of the LatF1. is the Mantle Edge Gland. This is an extensive tissue of elongate columnar epithelial cells which, via a slightly modified transitional tissue, are joined near the Anterior and Posterior Mantle Symphyses with the similarly elongate Isthmusistic Epithelium. However whereas the function of the latter is in very little doubt (the production of the Hinge), the function of the Mantle Edge Gland in pearl oyster is not known. Depending on the state of extension or retraction of the Mantle the Mantle Edge Gland may project beyond the shell periphery or lie medial to any part of the shell interior between this and the Pallial Inner Nacreous Layer. It is commonly seen in T.E.M to be covered with a surface secretion formed at least largely from the terminal microvillous vesicles of the apical membranes of the Mantle Edge Gland Cells. Whether this secretion takes part in the formation of the Outer Nacreous Layer or whether it contributes to the Inner Fibrous Sheath of the Prismatic Layer or has some other function is not known. As previously the Mantle Edge Gland has been named as the source tissue for all the precursors of the Organic Matrices of the Prismatic Layer. As will be discused further in Chapter 6.6., on morphological grounds alone, such a complicated function for this one cell - type, highly specialised tissue is untenable

Ubiquitous throughout the pearl oysters in the matrix of Terminal F1. and also seen here in other closely related superfamilies (e.g the Limopsoidea, Arcoidea and Ostreoidea) are a very specialised secretory cell with unusual staining affinities - the Ovoid Blue Glands. Again their function is unknown. Similarly the highly specialised Distal Diffuse Gland of the matrix of Terminal F1. in the *Pinctada* is of unknown function.

The subepithelial secretory glands of MedF1. - Turquoise Glands and Granular Cytoplasm Secretory Glands - are presumed to contribute at least some of the precursors of the Organic Matrices of the Outer Nacreous Layer since it was betweeen the MedF1. and the Pleated Secretion of Groove F1F2. that Nacreous Layer Organic Matrices were seen forming *(Pinctada sp. 2*, Figures 5.135 and 5.136). The Trabecular Turquoise Glands of this location are of the same staining affinities as those of the External Mantle and different from those of the F2. and LatF3..

However why there are such different arrays of secretory structures from place to place around the Mantle Margin in the Pterioidea is unknown. The most distinctly different array of subepithelial glands from those seen over the remaining four fifths (approx.) of the Mantle Margin, is at the Anterior periphery, from the Anterior Mantle Symphysis to ventral to the Byssal Embayment. The adjacent Outer Nacreous Layer is distinct from the Outer Nacreous Layer of the remainder of the shell periphery in that the medial surface shows a pattern of edges of the Nacre Sheets as in Figures 5.28 and 5.89, whereas the remainder of the periphery demonstrates parallel linear pattern of edges of Nacre Sheets. Again whether the alteration in subepithelial Glands of the MedF1. is related to the alteration in the pattern of edges of Nacre Sheets in the adjacent Outer Nacreous Layer is not known.

Ubiquitous throughout the *Pinctada* and nearly standard around the periphery of the Mantle Margin in radial section is the flat plateau - vee shaped gully - flat plateau structured Middle MedF1. epithelium but again its function is not known. If this is a mechanism to allow for greater extension of Med Fi. compared with the geometrically opposed part of LatF1. then presumably when this area is fully extended Distal MedF1. faces laterally, but to what purpose is unknown.

That the specialised structures of F1. may have functions unrelated to shell formation is shown by the formation of what appears to be a primitive eye on the Ancillary F1. of *Isognomon isognomon* The failure of predators and parasites to more readily attack the exposed F1. of open and poorly closing pearl oysters strongly suggests a chemical deterrent secreted by the Mantle Margin. Again which secretory structures may be involved in such a function is not known.

There is no evidence to suggest how F1. may produce the differences seen in the Outer Nacreous Layer compared with the Inner Nacreous Layer, except that Fougerouse - Tsing and Herbaut (1994) suggest that the colouration of the Outer Nacreous Layer of *Pinctada margaritifera* is produced by pigment glands in the surface epithelium of Proximal LatF1.. It is possible that the closely spaced parallel line patterns of edges of Nacre Sheets of the Outer Nacreous Layer may be as much due to physical causes as to chemical ones, (see Chapter 6.5.).

CHAPTER 6.5.

FORMATION OF THE ORGANIC MATRIX MORPHOLOGIES IN THE NACREOUS LAYERS OF PEARL OYSTERS.

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CHAPTER 6.5.

DISCUSSION (CONTD).

FORMATION OF ORGANIC MATRIX MORPHOLOGIES IN THE NACREOUS LAYERS OF PEARL SHELLS.

6.5.1. NAKAHARA'S (1991). DESCRIPTION OF NACRE FORMATION IN BIVALVES.

Nakahara, (1991) states that two distinct organic structures are involved in nacre formation in bivalves: An organic "sheet" which forms parallel compartments in the Extrapallial fluid and an envelope which intimately surrounds each growing crystal. He states that the sheets are composed of proteins rich in glycine and alanine while the envelope proteins are rich in aspartic acid.

Bivalve nacre as described by Nakahara, (1991) and seen in this study consists of flat sheets of aragonite Nacre Tiles enclosed in organic matrix. On a radial broken face the aragonite Nacre Tiles present as elongate bricks with the organic matrices surrounding each brick analogously to the cement of brickwork. In lateral or medial view the Nacre Sheets appear to be composed of similar sized irregular polygons which usually tend to irregular hexagons, diamond shapes or some other repeated geometric shape. Aragonite belongs to the Orthorhombic Crystal System and hence apparently hexagonal crystals of Aragonite are pseudohexagons resulting from twinning, (Rutley,1962).

Nakahara, (1991) in a comparison between Nacre formation in Bivalves and Gastropods has suggested that, in bivalves, a Nacre Sheet grows by the small free Nacre Tiles distal to the bound edge of a Nacre Sheet individually increasing in size until they coalesce as bound Nacre Tiles in a thus extended Nacre Sheet.

Before considering this description of Nacre formation, it is proposed to review the evidence seen in this work. The evidence seen in L.M. of pearl shell Nacreous Layers and of the remaining Nacreous Organic Matrices following acid decalcification, and S.E.M of pearl shells will be considered first, and then that from L.M and T.E.M of the External Mantles and Mantle Margins of pearl oysters.

6.5.2. L.M. OF DECALCIFIED NACREOUS LAYERS AND S.E.M. OF NACREOUS LAYERS OF PEARL SHELLS.

When shells of Pearl Oysters are viewed medially under L.M or S.E.M, the nacreous surface displays patterns like fingerprints - from strongly curved to nearly straight roughly parallel lines. These lines are the edges of successive Nacre Sheets.

These patterns are specific to certain regions on pearl oyster valves.

As above, (chapter 6. 3.), The Shoulder Regions of the *Pinctada* bear patterns composed of concentric circles, and less often spiral patterns, joined by curvilinear patterns and anastomosing line patterns, (Figures 5.13, a; 5.126, a).
The Pallial Gland Regions of the *Pinctada* display concentric ovoid and circular paterns and less significantly smallish spiral patterns, again joined by curvilinear and anastomosing linear patterns, (Figures 5.87, a; 5.126, b).

On all pearl oysters going ventrally and posteriorly from the Pallial Gland there is a gradual gradation from the patterns of the Pallial Gland Region to the patterns of the Distal Ventral and Distal Posterior Pallial Regions, where the Nacre Patterns tend towards those of evenly spaced parallel straight lines.
The Nacre Patterns displayed on the medial surface of the Proximal and Middle Anterior Pallial Regions are, by comparison with those of the Ventral and Posterior Pallial Regions, tortuous, with bent, curved and relatively straight lines which are commonly discontinuous. Nevertheless, the Distal Anterior Pallial Region again displays patterns which tend towards that of parallel straight lines.

The concentric circular patterns of the Shoulder Region and Pallial Gland Region represent circular stepped cones with the most medial Nacre Sheet in the centre. This most medial Nacre Sheet usually consists of about twenty or less Nacre Tiles and thus is about four or five Nacre Tiles across. Each more lateral Nacre Sheet is usually about seven Nacre Tiles wider than the one medial to it until the pattern becomes confused by the proximity of another pattern (e.g., another concentric circular pattern, a spiral pattern or a curvilinear pattern). The distances between the edges of the Nacre Sheets remain remarkably even between the circumferences of each two adjacent Nacre Sheets and between successive Nacre Sheets in the one pattern.

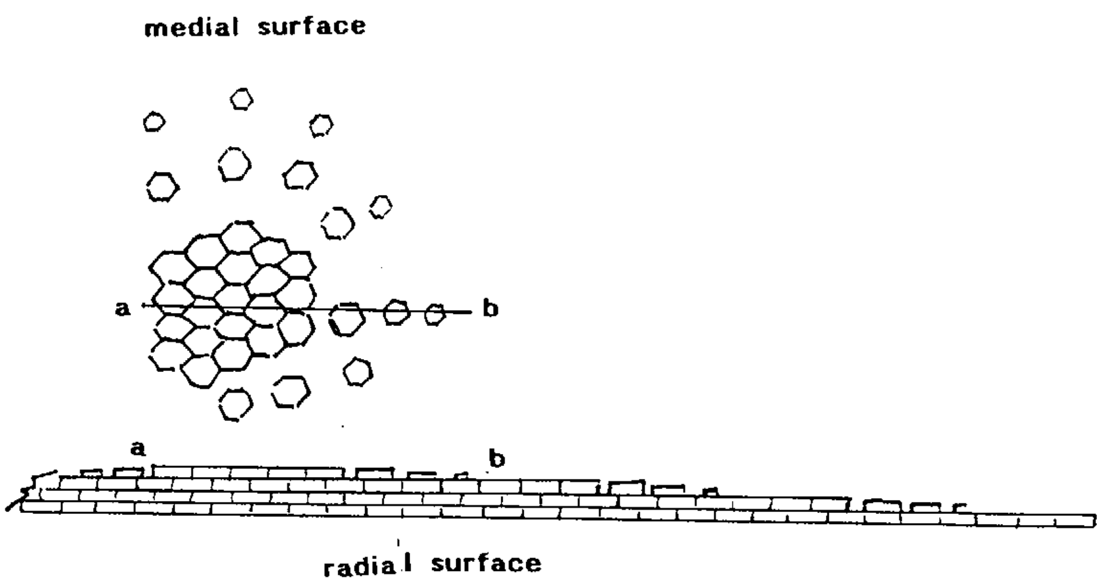


FIGURE 6.59. Part of the structure of a concentric circular pattern of edges of Nacre Sheets typical of the Shoulder Region near the Hinge in all members of the *Pinctada*. In section through the "centre" of the concentric pattern, (the twenty odd Nacre Tiles on the left above), the structure presents as a stepped cone of which the first four steps, (ie. the four most medial Nacre Sheets in such a structure), are illustrated above. The distance between edges of Nacre Sheets is remarkably constant for any one pattern of Nacre Sheet edges, (here depicted as steps seven Nacre Tiles wide) and as in every case in every pearl shell studied, the free Nacre Tiles decrease in size with distance from the edge of the Nacre Sheet and occur only on the proximal half of the exposed medial surface of each successively more lateral Nacre Sheet.

Spiral patterns are again representative of a stepped conical structure with a fundamentally different internal geometry. Again the surface distance between successive whorls of the spiral is remarkably constant. Outer whorls commonly terminate at a right angle joining the whorl to one of a set of parallel linear edges of Nacre Sheets.

Curivilinear and parallel straight line patterns, of their nature, represent flat step structures from a comparatively raised area on the medial surface to a lower, or vice versa. Going distally from the prominent thickening of the Pallial Gland Region to the Ventral or Posterior periphery is the "going down" direction of the wide flat "stairs" of the curvilinear and parallel straight line patterns of the Pallial Region.

Curvilinear patterns may contain anything from a series of tightly recurved nacre sheets to more gently curved ones. Linear Parallel patterns where extensive, usually again have remarkably even distances between any two edges of successive Nacre Sheets, and throughout the pattern.

It is invariably true that the partly bound Nacre Tiles approximate in surface dimensions the adjacent bound Nacre Tiles and the size of the free Nacre Tiles decreases with distance from the bound edge of the Nacre Sheet. Also the crystal habit of the free Nacre Tiles is a more perfect example of the habit to which the adjacent bound Nacre Tiles tend.

It is also almost invariably true that in any given area of medial surface of Nacreous Layer containing both Linear Parallel Patterns and Curvilinear patterns the distances between the successive curvilinear edges of Nacre Sheets is greater than the distance between successive edges of Nacre Sheets in the Linear Parallel Pattern. Further in any one region of the shell of a pearl oyster the tighter the recurving of the curved Nacre Sheets the greater is the distance between successive Nacre Sheets, and concomitantly, the greater the distance over which free Nacre Tiles spread from the bound edge of the Nacre Sheet.

At the peripheral Nacreo-prismatic Junction the Nacre Tiles in the peripheral Nacre Sheet maintain the average size of Outer Nacreous Layer Tiles to the edge of the Nacre Sheet. Distal to the edge of the peripheral Nacre Sheet, Nacre Tiles form a lacework in the grooves around the unsheathed medial ends of the Prismatic Layer Major Prisms. They are progressively smaller with the distance they are in the grooves from the bound edge of the Nacre Sheet - not with radial distance from the edge of the Nacre Sheet (Figure 6.58).

As far as can be clearly ascertained the small free Nacre Tiles distal to the edges of the Nacre Sheets whether on the medial surface of the Inner or the Outer Nacreous Layer invariably or almost invariably are situated at the junction of two underlying Nacre Tiles in the bound part of the next more lateral Nacre Sheet.

Nacre tiles whether they are diamond shaped, regular hexagonal, truncated diamond shaped, elongate hexagonal or truncated orthorhombic in any given location of the shell of a pearl oyster not only tend to the same crystal habit, but also tend to the same orientation of geometric axes whether they are bound or free, (Figure 6.56).

Bound Nacre Tiles in a mature Nacre Sheet while tending to the crystal habit of the adjacent free tiles are distorted into irregularly polygonal shapes with curved sides and often curved medial and lateral surfaces so that the one Nacre Tile may vary quite markedly in thickness from place to place.

Given the constraint that they occur astride the junction of underlying Nacre Tiles, free Nacre Tiles appear to be randomly scattered beyond the edge of the Nacre Sheet.

Partly-bound and bound Nacre Tiles commonly display on their lateral surface a shape which appears to be the enclosed shape of a previously free Nacre Tile. Less commonly Nacre Tiles may display on their lateral surface concave or convex circular structures (Figures 5.22, d and g; 5.43, b and c).

Despite the lack of regularity of the shape of single Nacre Tiles in a Nacre Sheet remarkably regular stair Patterns of Nacre Tiles are seen in successive Nacre

Sheets in radial sections. These are so common that they constitute the normal spatial arrangement of Nacre Tiles on a broken radial surface (Figure 5.29, b and d).

In the broken radial surface of a cultured pearl nacreous layer, Nacre Tiles Side Wall Organic Matrix can be seen to be coplanar through five successive Nacre Sheets (Figure 5.43, b).

6.5.3. EXTERNAL MANTLES AND MANTLE MARGINS OF PEARL OYSTERS.

The following qualifications apply to this assessment of the part played by secretory structures of the External Mantle and Mantle Margin of Pearl Oysters:

Firstly, the actual parts played by the secretions of any of the tissues in the formation of Nacreous Organic Matrices range between little known and unknown. The three major sources of secreted material seen in the Pallial Space of a pearl oyster are Trabecular Turquoise Glands, Granular Cytoplasm Secretory Glands and the budded-off vesicles from the swollen distal extremities of the microvilli of the surface epithelial apical membranes. No assessment of the comparative volumes contributed by the three sources is available from the evidence in this work.

Secondly, leaving aside the production of Hinge and the secretory activity of the Isthmus epithelium, it is assumed, because of the close proximity of the External Mantle to the nacreous surface, that the large molecule secretions of the tissues of the External Mantle are exclusively (or all but exclusively.) involved in the production of Nacreous Organic Matrix.

No such assumption can be made for the specialised tissues of the Mantle Margin where sensory and defensive mechanisms are likely as well as shell building secretory strucutres. A role in protecting the Nacreous Surface from sea water when the Mantle Margin is retracted is also envisaged for secretions of the Mantle Margin (Nakahara, 1991).

Thirdly, there is no known basis for the assumption that a particular type of gland will secrete Shell Organic Matrix precursor(s) at one site at the same rate as it does at another, nor that different species of secretory glands will provide Shell Organic Matrix precursors in the ratio of their volumetric abundance. Nor is there any basis for an assumption of constancy of rate of secretion of Shell Organic Matrix precursors over any time interval from any secretory structure or structures.

Necessarily this discussion is limited to presence, absence, or relative abundance of the various secretory glands and tissues in different loci.

Uniquely amongst those animals where the relevant tissues were studied, the pearl oysters and oysters display localised massive concentrations of Granular Cytoplasm Secretory Glands and Trabecular Turquoise Glands in the subepithelium of the Shoulder Region and the Pallial Gland Region. These concentrations of Subepithelial Glands in pearl oysters have a glandular volume which exceeds that of the entire remainder of the subepithelial glands of the External Mantle, yet the area of Nacre of the Shoulder Region plus that of the Pallial Gland Region, in pearl oysters of the Pinctada is little more than one fifth of the medial surface of the Inner Nacreous Layer.

6.5.4. If the secretory activity of the glands is standard throughout the External Mantle and all the secretions are used in the production of Nacreous Organic Matrix then either there must be a higher proportion of organic matrix to aragonite in the nacre opposed to the Shoulder Region and Pallial Gland Region of the Valve, or there must be relatively more volume of nacre produced in these Regions than the other Regions, or Shell Organic Matrix Precursors from these Regions must be transported to other Regions, or some combination of these.

No evidence is available from this study on relative concentrations of Nacreous Organic Matrices in the various regions of the valve - nor on whether over any given time scale at any stage in the shell's growth there is more nacre deposited in the Shoulder Region plus the Pallial Gland Region than over the remainder of the Valve.

Whether or not the evidence seen in this study supports the notion that Nacreous Organic Matrix produced in the Shoulder Region and Pallial Gland Region is transported to other regions of the Valve depends on the credence placed on Nakahara's. (1991) description of Bivalve nacre formation on one hand and the interpretations of the evidence presented here on the other.

Re Nakahara's (1991) description, it fails to account for most of the phenomena observed in this study. It explains neither the regularity of the positioning in successive Nacre Sheets of irregular Nacre Tiles; nor why the Nacre Patterns vary from place to place yet are broadly consistent in their distribution across numerous species.

In fact the only phenomenon seen in this study partially explained by Nakahara's description is the fact that whereas the "free" Nacre Tiles beyond the edge of a Nacre Sheet decrease in size with distance from the edge of the Nacre Sheet they are never seen as such (undersized Nacre Tiles) incorporated in mature Nacre Sheets. Nakahara suggests that it is the individual growth of each of these small Nacre Tiles which results in the growth of the Nacre Sheet. It is very difficult to see how such individual growth of small Nacre Tiles could result in either the regularity in size of the Bound Nacre Tiles nor the complex patterns of Nacre Sheet edges which are not only very similar from speciman to speciman in one species but closely approximated in many species. That the partly bound Nacre Tiles are full sized regular geometric shapes and the bound Nacre Tiles are invariably irregular shapes is strong evidence against Nakahara's description.

Precision in either criticism or suggestion is made difficult by lack of precision of data either from the literature or current observations on the Nacreous Organic Matrix. Nakahara. (1991) describes two Nacreous Organic Matrices - sheets of material which produce compartments parallel to the mantle (and nacreous) surfaces, and individual envelopes surrounding each Nacre Tile. He also talks of Organic Matrix inside the Nacre Tiles and illustrates an organic matrix envelope around the small "free" Nacre Tiles.

The evidence from this study suggests that all aragonite crystallisation takes place inside an organic matrix. That is that small "free" Nacre Tiles appear to be enclosed in Organic Matrix as well as the bound Nacre Tiles.

The remainder of this description of nacre formation depends to a great extent on what interpretation is to be placed on the many layers thick (up to approximately 40) of Nacre Tile sized "brickwork" seen on the surface of the Distal Folded Region, LatF1. and MedF1. of one pearl oyster.

Nakahara, (1991) says that the T.E.M. staining method employed in this study dissolves aragonite and so it might be argued that this material resulted from decalcified nacre which had somehow lodged in this position as an artifact. A close examination of the edges of the material makes this suggestion very unlikely. The open ends of the brickwork appear to be forming from a secreted material which is more electron dense than the interior of formed "bricks" and both granules and cilia appear to be associated with the formation. From both L.M and T.E.M it would appear that the cytoplasmic "sheets" of material seen in Trabecular Turquoise Glands are a major source of the material from which Nacreous Organic Matrix is formed.

Further, while it may be conceivable that decalcified Shell Organic Matrix may have somehow reached this position, it is not concievable that Shell Organic Matrix could have lodged in the secretions of MedF1. underneath the Pleated Secretion, where similar Nacreous Organic Matrix like brickwork is seen apparently forming from the secretion in which it is emmeshed, (Figure 5.136).

A study of both lots of Nacreous Organic Matrix "brickwork" utterly fails to reveal support for the notion that the Nacreous Organic Matrix consists of organic sheets enclosing "envelopes" (Nakahara, 1991). On the contrary, the part of the Organic Matrix analogous to the vertical sheets of mortar in brickwork appear to join the "horizontal sheets" and no sign of them can be seen lining the horizontal sheets as would be expected if they were in fact sections of hexagonal prismatic envelopes.

There are given in the Results, (Figures 5.134, b, c and d; 5.140, c; 5.148, a, b and d; 5.149, a and b; 5.150, b, c and d; and 5.154,a), numerous examples of parallel sheets of material on locations on the External Mantle and Mantle Margin of a large number of species of Pearl Oysters which have spatial arrangements and intervening connections which mimic the "brickwork" strucutre of nacre. If the interpretation that these are forming Nacreous Organic Matrices is correct and if it is accepted that the multiple layers of preformed Nacreous Organic Matrices "brickwork" seen on T.E.M is in fact forming nacre prior to calcification then Nakahara's, (1991) description is unacceptable.

All the phenomenon seen in this work fit the concept that nacre is laid down in layers many Nacre Sheets thick as uncalcified preformed sheets of "bricks" which are constrained into sizes and shapes by the chemistry of the scleroprotein precursors secreted by the Turquoise Glands, Granular Cytoplasm Secretory Glands and perhaps also the vesicular bodies from the mantle epithelium apical membrane microvilli termini.

The "free" Nacre Tiles of the Nacre Sheet edges of the Inner and Outer Nacreous Layers are in no way diffently related to the adjacent edge of a Nacre Sheet or the "bound" Nacre Tiles of that Nacre Sheet than are the "free" Nacre Tiles seen surrounding the periphery of the medial end of Major Prisms to the most peripheral Nacre Sheet of the Outer Nacreous Layer. In every case observed the free Nacre Tiles of the Nacreo-prismatic Junction tend to the same crystal habit as do the adjacent bound and partially bound Nacre Tiles of the peripheral Outer Nacreous Layer Nacre Sheet. As with the edges of Nacre Sheets, the free Nacre Tiles of the Nacreo-prismatic Junction decrease in size with distance from the bound edge of the Nacre Sheet - but in this case this distance is the distance along the grooves surrounding the Major Prisms.

Pressure between the External Mantle and the Medial surface of the valve on Nacreous Organic Matrix "brickwork" prior to calcification, coupled with progressively decreasing amounts of secretion of Organic Matrix precursors may explain the flattened conical step pyramid structures which appear as concentric circles in Shoulder Region and Pallial Gland Region patterns of edges of Nacre Sheets. Similarly pressure on the supposed greater amounts of organic matrix precursors produced by the glands of the Shoulder Region and the Pallial Gland Region would explain the tongues of curvilinear patterns projected distally from the Pallial Gland Region and Anteriorly from the Shoulder Region.

Pressure between the distal part of the Mantle - whether Distal Folded Region or LatF1. - and the medial surface of the Prismatic Layer would explain why the small "free" Nacre Tiles of the Nacreo-prismatic Junction are forced along the grooves in between Major Prisms. In the same way in every case which could be observed, the free small Nacre Tiles most distant from the bound edge of a Nacre Sheet of Inner Nacreous Layer were located on the slight groove at the junction of underlying (more lateral) Nacre Tiles.

That bound Nacre Tiles are superimposed on smaller "free" Nacre Tiles from a previous deposition of nacre is in accord with the presence of what appear to be the outlines of enclosed small Nacre Tiles in numerous examples of nacre from all areas of all pearl oyster shells studied. Since the small free Nacre Tiles are invariably positioned on the junction of underlying Nacre Tiles and the Nacre Tiles of any species in any area tend to similar sizes this also explains the "stair" patterns seen on the radial broken surface of successive Nacre Sheets.

That small "free" Nacre Tiles, larger "free" Nacre Tiles, "partially bound" Nacre Tiles and Bound Nacre Tiles in each location on the medial surface of both the Inner and Outer Nacreous Layers of all species of pearl oysters' shells tend to a single crystal habit can be explained either by Nakahara's contention that the full sized nacre crystals have grown by accretion onto the small free tiles or that the Organic Matrices of that region constrain the crystallising calcium carbonate to a certain habit.

This study has provided no evidence as to why the orientation with respect to the edge of the related Nacre Sheet of the geometric axes of the Nacre Tiles (bound and free) in a given area should all (or mostly) be the same. It would seem very likely that the chemistry of the Organic Matrices on the medial surface of the Nacre Sheets on which the Free Nacre Tiles form constrains them to orientations with similarly directed geometric axes.

CHAPTER 6.6.

DISCUSSION (Continued).

PRISMATIC LAYER ORGANIC MATRIX MORPHOLOGY AND THE MICROANATOMY AND HISTOLOGY OF FOLDS F2. AND LATF3. IN PEARL OYSTERS.

INDEX

CHAPTER 6.6.

DISCUSSION (Continued).
PRISMATIC LAYER ORGANIC MATRIX MORPHOLOGY AND THE MICROANATOMY AND HISTOLOGY OF FOLDS F2. AND LATF3. IN PEARL OYSTERS.

6.6.1. PRISMATIC LAYER ORGANIC MATRIX COMPONENTS.

As previously, the outer Shell Layer of the Pterioidea is a calcitic Prismatic Layer - consisting of medially superimposed Growth Scales. The Organic Matrices of the Prismatic Layer are those of the Outer Fibrous Sheath, the Inner Fibrous Sheath and the Prisms. The Prisms, irregularly polygonal tubes standing normal to the shell surface, have organic matrix components of Outer Prismatic End Plate, Inner Lateral Structure, Transverse Parallel Prismatic Side Walls, Linear Structure Interprismatic Organic Matrix, Reteform Interprismatic Organic Matrix, Intraprismatic Organic Matrix and Inner Prismatic End Plates.

6.6.2. UNCERTAINTIES IN THE DATA.

This work has left uncertainties regarding these structures as previously noted. Those germane to this discussion are the following:

6.6.2.1. It is not known why some Growth Scales, in species where these are usually separated at least by Inner and Outer Prismatic End Plates, have, in some areas, no physical division between them and the adjoining Growth Scale or Growth Scales.

6.6.2.2. The physical and functional as well as the chemical relationships of the Linear Structure Interprismatic Organic Matrix, Reteform Interprismatic Organic Matrix, Transverse Parallel Prismatic Side Walls and the Intraprismatic Organic Matrix remain to some extent obscure.

In the only instance where Intraprismatic Organic Matrix was observed free from the enclosed calcite tablets, it appeared virtually indistinguishable morphologically from the Reteform Interprismatic Organic Matrix (Figure 5. 128). In numerous instances where the Intraprismatic Organic Matrix has been broken out of Prisms along with the calcite tablets, what appear to be twin transverse parallel remnants of Intraprismatic Organic Matrix can be observed on the side walls joining with, and morphologically very similar to, Reteform Interprismatic Organic Matrix. (Figure 5.96, e). In numerous instances it has appeared that Reteform Interprismatic Organic Matrix forms a sheath covering the exposed side walls of Major Prisms on a radial broken surface (Figure 5.356, a), and appears in at least one instance to be clearly physically associated with an Inner Prismatic End Plate (Figure 5.145, c). However Linear Structure Interprismatic Organic Matrix, (Figure 5.39), may be an external Sheath associated with the Reteform Interprismatic Organic Matrix.

The relationship of Intraprismatic Organic Matrix, Reteform Interprismatic Organic Matrix and Linear Structure Interprismatic Organic Matrix to each other is confused by the fact that two of them, the Reteform and Linear Structure Interprismatic Organic Matrices (as far as is known) never have been observed in decalcified shell and the Intraprismatic Organic Matrix in this situation is invariably largely destroyed. In the acid decalcified shell the Transverse Parallel Prismatic Side Walls are quite obviously common to adjoining prisms. However these last, by far the most dominant feature in acid decalcified pearl shell Prismatic Layer, are almost always largely obscured under S.E.M of non-decalcified shell. An explanation of why Reteform Interprismatic Organic Matrix is not seen in acid decalcified shell and remnants of Intraprismatic Organic Matrix are, if they are the same material, might be that the acid strength was higher or remained relatively high for longer in the vicinity of the former during decalcification than between the calcite tablets.

Perhaps the description which best fits the observations is that the Major Prisms and Growth Process Prisms have Transverse Parallel Side Walls which are shared between adjacent Prisms. Inside the Transverse Parallel Side Walls is an

OrganicMatrix structure which presents as a continuous sheet with a pattern of linear ridges parallel to the long axes of the Prisms, on the surface opposed to the Transverse Parallel Side Walls. However, viewed from inside the Prisms, this sheet of Organic Matrix is lined with a three dimensional network of frothy material (the Reteform Interprismatic Organic Matrix). This reteform (or frothy) material extends transversely across the Prisms in twin lamina, (Intraprismatic Organic Matrix), thus dividing the Prism into numerous compartments in which are the pseudohexagonal, thickish calcite tablets.

6.6.2.3. Similarly it is uncertain whether the Outer Prismatic End Plate and the Inner Lateral Structure are related chemically and come from the same source. This appears likely as where the former stains aberrantly blue (e.g., with Azan) the physically associated Inner Lateral Structure invariably reflects the same aberrant staining affinity.

6.6.2.4. To these uncertainties must be added doubts as to the overall method of production of the Organic Matrices of the Growth Scale. That is, it is not known whether it is produced in segments which are then moved more distally for production of the next segment, or all produced in one piece.

6.6.3.ANATOMY OF THE MANTLE MARGIN AND GROWTH SCALE SYNTHESIS.

The geometry of the Mantle Margin is instructive. In *Pinctada maxima* the contracted lengths of F1. and F2. after fixation are usually about the same - in the vicinity of 2.0mm. The greatest attainable length of each of these two Mantle Margins is not known but given the structure of their radial Fibrous Connective Tissue it is probably not much more than their length in a contracted state. Since all pearl oysters can join their F3. Mantle Margins when fully gaped, the extended length of F3. in an adult *Pinctada maxima* is in the order of 25 - 30mm.

Previously, (Discussion, Chapter 6.1..) a mechanism was described whereby, by partial withdrawal of the previously touching F3. peripheries, the Growth Processes could be produced in the secreted Prismatic Layer Organic Matrices on LatF3.. The parallelogram shapes of the Outer Prismatic End Plates constrained into rows near the edges of the Growth Processes suggests that at least the Outer Fibrous Sheath, Outer Prismatic End Plates, and the Prism Side Walls and Inter- and Intra-prismatic Organic Matrices are secreted, fabricated and in a perhaps partially sclerified form by the time the F3. peripheries have parted.

It appears from the histology of LatF3. and MedF2. that these tissues are, on their own, little more capable of synthesizing a scleroprotein Structure of the complexity of the Prismatic Layer of a pearl shell than is LatF1.. Tissues of sufficient complexity to synthesise such a structure are the Proximal and Middle LatF2.. However as above, their radial length in an extended state is probably in the order of about 3.0mm in a large *Pinctada maxima* whereas the radial length of a Growth Scale in such an animal is in the order of 30mm. This is the same order of magnitude as the radial length of the fully extended LatF3.. Hence while the tissue surface on which the Growth Scale is formed and the Growth Processes are shaped is of the same order of magnitude as the Growth Scale on the shell, the underlying tissues are probably incapable of forming it; and those tissues which are capable of forming it from the point of view of their complexity are only about 10% or less in radial length of the radial length of the Growth Scale.

6.6.4. PRISMATIC LAYER ANOMALIES.

Further confusion is added to the picture by the fact that the Prismatic Layer is not consistent in the relationship of its parts. For example the Lateral surface of a Growth Scale may be defined for a large part of its proximal length by Outer Prismatic End Plates, which may simply not exist distally with the prisms of the medial Growth Scale simply merging with those of the overlying Growth Scale.

6.6.5. RELATIVE POSITION OF SHELL AND MANTLE MARGIN DURING LOCATION OF A NEW GROWTH SCALE.

The Proximal extremities of the Growth Scales are usually interleaved with the distal extremities of the successive depositions of Outer Nacreous Layer. These Proximal extremities of Growth Scales present, in radial section, as sometimes very elongate extensions of the proximal pointed end of the distorted spindle shape which the Growth Scale has in radial section.

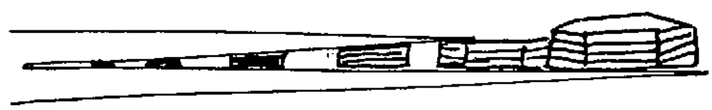


FIGURE 6.60. Diagram of radial section of proximal extremity of a Growth Scale between distal extremities of successive depositions of Outer Nacreous Layer.

Commonly, this elongate proximal extension of the Growth Scale ceases to stain red with Azan proximally where Transverse Parallel Side Wall Organic Matrix can no longer be accurately distinguished under L.M., and appears as a thin line of blue foam like material between two lots of nacre. While between the two lots of nacre neither Outer Fibrous Sheath, Outer Prismatic End Plates, Inner Prismatic End Plates nor Inner Fibrous Sheath can be distinguished. The latter is not surprising as Inner Fibrous Sheath has never been seen in medial view of peripheral Nacreo-prismatic Junction anywhere in any species of the Pterioidea with S.E.M.

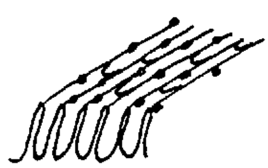
This lack of not only an Outer Fibrous Sheath but also Outer Prismatic End Plates lateral to the diminuitive underlying pieces of Inner Lateral Structure and, more distally, incipient prisms, suggests that at the time of deposition of the Growth Scale the tissue responsible for Inner Lateral Structure secretion and more distally the tissue responsible for prism Transverse Lateral Side Wall Organic Matrix secretion are immediately opposed to the medial surface of the distal extremity of the last laid down part of the Outer Nacreous Layer.

The various discrete entities which constitute the organic matrices of the Prismatic Layers of pearl shells and the probable sources of their precursors will now be considered.

6.6.6.OUTER FIBROUS SHEATH.

Structure.

The Outer Fibrous Sheath in Lateral View shows coarse more or less parallel fibres. The "Fibres" are about 0.8μm apart centre to centre on a young adult *Pinctada maxima*. Where the fibres are eroded they can be seen to continue in a plane normal to the surface of the Prismatic Layer which suggests that they are either thin slats which present edge on to the lateral surface, or are in fact pleats. There are fairly frequent anastomoses between the "fibres" (or pleats) which are recurved (Fig. 5.33, d) with the rounded ends of the anastomoses mostly pointing in the same direction. The surface of the fibres (or pleats) bears small nodules about the size and frequency of the Black Granules which can be seen on the Pleated Secretion, the detrital secretion on the medial surface of the distal Growth Scale, and on the Vesiculate Secretion of Groove F1F2..



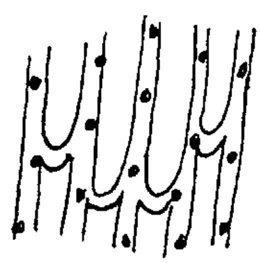


FIGURE 6.61. The Outer Fibrous Sheath of the Prismatic Layer in the Pterioidea.

In *Pinctada sp.1.* (Figure 5.137, b and c) is shown the developing Vesiculate Secretion of Groove F1F2.. For reasons already given this is thought to be the most likely source of the Outer Fibrous Sheath of the Prismatic Layer.
As seen in *Pinctada maxima* (Figure 5.67), it is secreted by the subepithelial secretory glands of Proximal LatF2..
This layer is modified in harmony with the subsequent positioning of the underlying Outer Prismatic End Plates. In some species the fibres of the Outer Fibrous Sheath are farther apart over the central area of the Outer Prismatic End Plates and more closely opposed over the peripheries. (Figure 5.33, b; 5.93, d). In *Pinctada fucata* the fibres over the central area of the Outer Prismatic End Plate run normal to the general direction of the fibres of the Outer Fibrous Sheath (Figure 5.127, e).

6.6.7. The Outer Prismatic End Plates, Inner Lateral Structure and Proximal LatF2..

The Outer Prismatic End Plates cap the Outer Surface of the Major Prisms and the Growth Process Prisms. As such they are either irregularly polygonal on the Growth Scale or the middle of the Growth Processes or rectanular on the edge of the Growth Processes.

They are of at least several different constructions having a concentric ring structure in some species and hexagonal "spider web" construction in others.

FIGURE 6.62. Lateral view, morphology of Outer Prismatic End Plates, (repeat of Figure 2.11).

As above they are accurately positioned under local variation in direction of the surface constituents of the Outer Fibrous Sheath and therefore presumably their

Organic Matrix Precursors are secreted in situ by secretory structures in the same locus as those which secrete the Outer Fibrous Sheath - Proximal LatF2., or alternatively, they are in some way positioned by the variations in the Outer Fibrous Sheath.

The Inner Lateral Structure joins the medial surface of the Outer Prismatic End Plate and invariably parallels the Outer Prismatic End Plate to which it is attached in staining affinities. What appears to be forming Inner Lateral Structure has been observed forming from Vesiculate Secretion at Proximal LatF2.

6.6.8. The Transverse Parallel Prismatic Side Walls and Middle LatF2..

The Transverse Parallel Prismatic Side Walls are the most obvious and voluminous feature of the Prismatic Layer Organic Matrix in acid decalcified shells.

Shape and Size

Junctions

Resistance to Boring Sponge.

They are composed of flattened slats of scleroprotein which stretch from corner to corner of the prisms. The individual slats in *Pinclada maxima* are commonly 35 μm long, 1.5 μm wide and 1.5 μm thick. There is a thickening at the corner of the prisms, suggesting that the prism increases in length by addition of one slat to each side of the prism at the same time. For the greater part of most prisms of most Prismatic Layers they stain amber with Mallorys, yellow/gold with M.S.B and A.B./M.S.B and dark red with Azan. However there are some parts of most prism layers which stain purple with Mallorys, grey/blue with M.S.B and A.B./M.S.B and blue with Azan. Usually these latter stainings occur in a fairly discrete band or bands parallel to and either at or just medial to the lateral surface of the Growth Scale in which they occur. In *Pinclada fucata* in radial section of decalcified shell they are the only indication (proximal to the Growth Processes) of the shape of the Growth Scales. These alteration in staining affinities occur abruptly - one wall of a prism may stain e.g., dark red with Azan and the adjacent walls on either side may stain dark blue with this stain. The slats of

Parallel Transverse Side Wall material which stain bluish colours with the Trichrome used are invariably partly destroyed in the process of acid decalcification. Where with Azan e.g., a blue staining slat joins red staining slats at the corner of a prism instead of a smooth swelling as seen between red slats joining, the blue staining slat appear to be "butted into" the junction of the red slats. The staining affinities and degree of sclerification of the parts of the Prismatic Layer will be discussed further later.

Forming Prismatic Layer Parallel Transverse Prismatic Side Wall Organic Matrix has been observed at Middle LatF2. so that it appears likely that the specialised secretory structures of this location are responsible for producing at least some of the precursors of its Organic Matrices.

6.6.9.Linear Structure Interprismatic Organic Matrix, Reteform Interprismatic Organic Matrix and Intraprismatic Organic Matrix andMiddle LatF2..

As above, it is here assumed that these three structures, from consideration of acid solubility, position, tissue of formation and geometrical relationships may well be fairly closely related in chemistry and origin, and two of them may be exactly the same substance in a different place. They will thus be considered together.

As above, Linear Structure and Reteform Interprismatic Organic Matrices have never been observed in acid decalcified pearl shells, but they invariably invest and obscure Parallel Transverse Interprismatic Side Wall Organic Matrices on the radial broken surface of pearl shells.

The colour reactions suggest that the Light Blue Glands of Middle LatF2. is the source of one of the precursors of the Intraprismatic Organic Matrix and possibly therefore also the Reteform and Linear Pattern Interprismatic Organic Matrices but this is uncertain. It is tempting to suggest that the semicircular infoldings of the Middle LatF2. function as moulds for the formation of the Parallel Transverse

Interprismatic Side Wall Organic Matrices and that all four side wall and Intraprismatic Organic Matrices are generated in this area but there is little direct evidence for this.

6.6.10.Inner Prismatic End Plates.

Radial section of decalcified pearl shell Prismatic Layer commonly shows a thickened layer of Organic Matrix on the medial end of a Major Prism or Growth Process Prism although this is sometimes lacking in some areas. On the Growth Processes and distal medial surface of Growth Scales these Inner Prismatic End Plates are covered by the Inner Fibrous Sheath. With S.E.M it is often difficult to ascertain whether the surface observed is organic or inorganic. Where the inorganic calcite can be unequivocally observed the microcrystalline structure is commonly oriented parallel to one side of the polygon. In an adjoining prism the microcrystaline structure may be oriented parallel to a side of the polygon which joins the other at about 120°. This indicates, a., that the orientation of the calcite crystals is determined individually in each prism, and b., that the determinant is probably the Reteform Interprismatic Organic Matrix - this being the Organic Matrix forming the Inner Surface of the polygonal prism.

There is no evidence in this study to suggest a tissue source of the Inner Prismatic End Plates except that having the same staining affinities as the other constituents of the Prismatic Layer other than the Intraprismatic Organic Matrices, and having regard to their location on the medial surface of the Growth Scale, their secreted precursors are presumably produced by the subepithelial Granular Cytoplasm Secretory Glands of F2, and perhaps LatF3..

6.6.11.Inner Fibrous Sheath.

The Inner Fibrous Sheath, as above, clothes the medial surface of the Growth Processes and the distal medial Growth Scale. It is always lacking for some distance distal to the peripheral Nacreo-prismatic Junction. It is of far finer construction than the Outer Fibrous Sheath and totally lacks the small protrusions thought to be deposited "Black Granules" of the latter.

Because of its distribution it may be a product of the precursor secretions of Distal LatF3. but alternatively may be added by Distal LatF1. after the newly formed Growth Scale is placed in position.

No vestige of any of these structures has been seen in the cultured pearl examined - there the Radial Layer appeared very similar to the aragonite Prismatic Layer of *Velesumio ambiguus*.

6.6.12.Functions of the Prismatic Layer in Pearl Oysters and its Relationship to Shell Layers in Other Bivalves.

Starfish predators of Bivalves attach suckers to the outer surface of the valves to pull them apart - thus a major function of the Growth Processes may be that they fall off under this circumstance and thus protectect the oyster from predation.

A major threat to pearl oysters is shell destruction by boring sponges. As shown here (Figure 5.44), boring sponges commonly penetrate a pearl shell either through the exposed Nacreous Layer near the hinge or the sponge may gain access down a Major Prism and then produce cavernous destruction in the underlying nacre. From this it is apparent that a freely projecting Growth Process which is destined to be broken off acts as a protection for the pearl oyster as does the relatively resilient side walls of the Prismatic Layer - especially in young pearl oysters where the Nacreous Layers are thin, and towards the periphery of older oysters.

It is of interest that the Organic Matrices of the Prismatic Layer of Pearl Oysters mimic the staining affinities of a non-calcareous outer Shell Layer scleroprotein sheet which is ubiquitous in those Bivalvia which do not have an outer calcitic Prismatic Layer - the martius yellow positive scleroprotein produced by the Mantle Margins of members of these superfamilies. That the Prismatic Layer of pearl oysters and some other Bivalves is calcitic may be a secondary result of the development of a scleroprotein which is resistant to acid hydrolysis and more specifically, resistant to destruction by a boring sponge. For these reasons and by comparison of the geometrical relationships and staining reactions of the Organic Matrices of the calcitic Prismatic Layer of Pearl Oysters - especially the yellow/gold staining with M.S.B. and A.B./MS.B.. it is here held that the outer Shell Layers (Prismatic Layers), of the Pinnoidea, Pterioidea, Mytiloidea, Ostreoidea, Nuculoidea and Tellinoidea and some Veneroidea are all ornate derivitives of at least part of the outer non-calcareous trilaminate outer fibrous Shell Layer of the primitive Bivlave *Velesunio ambiguus* and have no relationship at all to the simple aragonite Prismatic Layer of that primitive Bivalve.

As a corollary, where the Bivalves in this study possessed a calcitic outer calcareous Shell Layer they lacked a separate non-calcareous outer Shell Layer. Conversely, every Bivalve studied whose calcareous Outer Shell Layer was aragonite possessed a separate Non-calcareous Outer Shell Layer, in part at least comprised of a martius yellow positive scleroprotrein lamina.

REFERENCES.

Dix, T.G. 1972. Histochemistry of mantle and pearl sac secretory cells in *Pinctada maxima* (Lamellibranchia). *Australian J. Zoology* 20 : 359-368.

Dix, T.G. 1973. Histology of the mantle and pearl sac of the pearl oyster *Pinctada maxima* (Lamellibranchia). *Journal of the malacological society of Australia* 2 : 365-375.

Drury, R.A.B. and Wallington, E.H. 1980. *Carleton's Histological Technique*. 5th. edition, Oxford University Press.

Fougerouse-Tsing, A. and Herbaut, C. 1994. *Atlas Anatomique de l'huitre perliere Pinctada Margaritifera*. Establissement pour la Valorisation des Activites Aquacoles et Maratimes. Papeete - Tahiti.

Gabe, M. 1976. *Histological Techniques* Translation by Bleckith, R. E. and Masson, A. K., Springer- Verlag, of *Techniques Histologiques* (1968), Masson et C. Paris.

Garcia-Gasca, A. Ochoa-Baez, R. I. and Betancourt, M. 1994. Microscopic anatomy of the pearl oyster *Pinctada mazatlanica* (Hanley, 1856). *J. of Shellfish Res.* 13 (1) : 85-91.

Gervis, M.H. and Sims, N.A. 1992. *The biology and culture of pearl oysters (Bivalvia : Pteriidae)*. ICLARM Contribution No. 837, ICLARM Stud. Rev. 21, 49p.

Hynd, J. S. 1955. A revision of the Australian pearl-shells, Genus *Pinctada* (Lamellibranchia). *Aust. J. Mar. Freshwater Res.* 6 (1) : 98-137.

Jabbour-Zahab, R., Chagot, D., Blanc, F. and Grizel, H. 1992. Mantle histology, histochemistry and ultrastructure of the pearl oyster *Pinctada margaritifera* (L.). *Aquat. Living Resour.*, 5 : 287-298.

Joose, J. and Geraerts, W. P. M. 1983. Endocrinology. In K. M. Wilbur and A. M. S. Saleuddin (eds.), *The Mollusca*, 4, part 1, Ch. 8 : 318-390. Academic Press, New York.

Moore, R.C. (ed.) 1971. *Treatise on Invertebrate Paleontology*. Part N, Vol. 3, Mollusca 6, Bivalvia. The Geological Society of America Inc. and the University of Kansas.

Nakahara, H. 1991. Nacre formation in bivalve and gastropod molluscs. In S. Suga and H. Nakahara, (eds.), *Mechanisms and phylogeny of mineralisation in biological systems*. Sixth international symposium on biomineralisation. Springer-Verlag, Tokyo.

Nakahara, H. and Bevelander, G. 1971. The formation and growth of the prismatic layer of *Pinctada radiata*. *Calc. Tiss. Res.* 7 : 31-45.

Nakahara, H., Kakei, M. and Bevelander, G. 1980. Fine structure and amino acid composition of the organic "envelope" in the Prismatic Layer of some bivalve shells. *Venus* 39 (3) : 167-177.

Revarthy, B., Bharathi, M. S. and Ramalingam, K. 1989. The amino acid composition of the periostracum and organic matrix of the shells in three marine bivalves. *Proc Indian natn. Sci. Acad.* B55 No. 2 : 107-110.

Read, H. H. 1962. *Rutley's Elements of Mineralogy*. 25th. edition, Thomas Murby & Co. London.

Saleuddin A. S. M. and Petit, H. P. 1983. The mode of formation and the structure of the periostracum. In K. M. Wilbur and A. S. M. Saleuddin (eds.), *The Mollusca* 4, part 1. Ch. 5 : 199-234. Academic Press, New York.

Spurr, A. R. 1969. A low viscosity epoxy resin embedding medium for electron microscopy. *Ultrastructure Research* 26 : 31-43.

Shadwick, R. E. and Gosline, J. M. 1983. Molecular biomechanics of protein rubbers in molluscs. In K. M. Wilbur and A. S. M. Saleuddin, (eds.), *The Mollusca*, 1, Ch.9, 399-427. Academic Press, New York.

Stasek, C. R. and McWilliams, W. R. The comparative morphology and evolution of the molluscan mantle edge. *The Veliger* 16 (1) : 1-19

Steedman, H. F. 1970. A one solution triacid general stain which differentiates oxygenated from non-oxygenated red blood corpuscles. *Stain Technology* 45 (6): 247-253.

Taylor, J. D. 1973. The structural evolution of the bivalve shell. *Palaeontology* 16 Part 3 : 519-534.

Taylor, J. D., Kennedy, W. J. and Hall, A. 1969. The shell structure and mineralogy of the Bivalvia : Introduction : Nuculacea to Trigonacea. Bull. Br. Mus. nat. Hist. Zool. suppl. 3 : 1- 125.

Taylor, J. D., Kennedy, W. J. and Hall, A. 1973. The shell structure and mineralogy of the Bivalvia. Lucinacea - Clavagellacea. Conclusions. Bull. Br. Mus. nat. Hist. Zool. 22 : 253-294.

Tsujii, T. 1960. Studies on the mechanism of shell and pearl formation in Mollusca. *Journal of the Faculty of Fisheries Prefectural University of Mie* 5 :

Vokes, H.E. 1980. *Genera of the Bivalvia.* The Paleontolgical Res. Inst. Ithaca New York. pp307.

Wada, K. 1981. Colour and fine structure of pearls. *Kobutsugaku Zasshi* 15 (3): 143-150.

Waite, J. H. 1983. Quinone tanned scleroproteins. In K. M. Wilbur and A. S. M. Saleuddin, (eds.), *The Mollusca.* 1 Ch. 11 : 467-496. Academic Press, New York.

Waller, T. R. 1978. Morphology, morphoclines and a new classification of the Pteriomorphia (Mollusca: Bivalvia). *Phil. Trans. R. Soc. Lond.* B 284 : 345-365.

Watabe, N. 1984. Shell, in Bereiter - Hahn, Jurgen, Matoltsy, A. G. and Richards, K. S. eds., *Biology of the Integument.* Springer - Verlag, Berlin. Ch. 25 : 448-485.

Weiner, S., Talmon, Y. and Traub, W. 1983. Electron diffraction of mollusc shell organic matrices and their relationship to the mineral phase. *Int. J. Biol. Macromol.* 5 : 325-

Weiner, S. and Traub, W. Macromolecules in mollusc shells and their functions in biomineralisation. *Phil. Trans. R. Soc. Lond.* B 304 : 425- 434.

Wilbur, K. M. and Saleuddin, A. S. M. 1983. Shell formation. In K. M. Wilbur and A. S. M. Saleuddin, (eds.), *The Mollusca* 4, part 1, Ch. 6 : 236-279. Academic Press, New York.

Winsor, L. 1984. *Manual of Basic Zoological Microtechniques.* Printed James Cook University, Townsville.

Yamaguchi, K. 1994. Shell structure and behaviour related to cementation in oysters. *Marine Biology* 118 : 89-100.

APPENDIX 1.

GLOSSARY OF FORMAL TERMS USED.

Excluded from this Glossary are:

1. Normal self explanatory descriptive terms which are written in lower case;
2. Formal histological or anatomical terms in normal usage unless more accurately defined;
3. Taxonomic names.

A.

Adductor Muscle Scar.

Of pearl oysters, the single area of aragonite Myostracal Prisms on the medial surface of the Valve of a pearl shell where the Adductor Muscle adheres to the Valve.

Ancillary F1.

The second most lateral of the four Marginal Mantle Folds of the Genera *Pteria, Isognomon* and *Malleus*

Anterior.

Of a pearl oyster or bivalve, refers to the aspect or border associated with the Byssus, or in its absence the Oral Cavity.

Anterior Mantle Symphysis.

The part of the Mantle at the antero-dorsal periphery where the Left and Right Mantle Margins fuse.

Apical Channel.

In pearl oysters, the narrow channel bounded by the cells of the Omega Gland laterally and the Dactylocytes medially wherein lies the forming Pleated Secretion of Groove F1F2.. In other bivalves, e.g. the Mytilidae, an analogous feature.

Apical Groove F1 Ancillary F1..

The most proximal part of the Marginal Mantle Groove F1 Ancillary F1. in the Genera *Pteria*, *Isognomon* and *Malleus*.

Apical Groove F1F2..

The proximal Marginal Mantle Groove F1F2. and adjacent structures:- Omega Gland, Omega Gland Glands, Sub-Omega Gland Connective Tissue, Dactylocytes, Black Granule Secretory Cells, Circum-pallial Nerve, Circum-pallial Sinus and associated secretions.

Apical Groove F2F3..

The proximal part of Marginal Mantle Groove F2F3. and associated epithelia and subepithelial glands and structures.

B.

Bag of Marbles Turquoise Gland.

A large unicellular secretory gland which stains turquoise with A.B./M.S.B stain and is refractory to all other stains used here. Hence it is probably acid-mucopolysaccharide secreting. Under T.E.M it is distinguished from Trabecular Turquoise Glands by its distinctive cytoplasmic structure implied by the name.

Black Granule Secretory Cells.

Highly specialised epithelial cells which occur on the most proximal part of Proximal LatF2. of pearl oysters immediately adjoining the most distal Dactylocyte where the Apical Channel opens into Groove F1F2.. The apical cytoplasm stains a distinctive purple with Azan and green with A.B./M.S.B. These cells synthesise and secrete the electron dense granules seen with T.E.M. on the medial surface of the Pleated Secretion of Groove F1F2. and the lateral surface of the Vesiculate Secretion of Groove F1F2..

C.

Celtic Scroll Cell.

Cells which comprise the outer layer of the Omega Gland Proximal Part which secrete large vesicles into the proximal part of Groove F1F2..

Circum-pallial Nerve.

A large nerve trunk which runs just proximal to the apex of Groove F1F2. for the entire periphery of both left and right Mantles, and supplies major rami at intervals to all Marginal Mantle Folds.

Circum-pallial Sinus.

A large blood sinus which runs beside the Circum-pallial Nerve around the entire periphery of both left and right Mantles and is connected directly to at least some of the sinus of F1. In *P.maxima* and the *Ostreacea* it commonly contains numerous haemocytes.

D.

Dactylocytes.

Elongate columnar epithelial cells with prominent central nuclei whose apical surfaces line the medial surface of the Apical Channel. As such their microvillous apical membrane touches against the forming Pleated Secretion of Groove F1F2. which separates them from the Omega Gland.

Distal.

According to the sense used, distal can mean away from the hinge, or more usually, with reference to the valve or a radial section of the Mantle, distal can mean away from the Adductor Muscle Scar or Adductor Muscle respectively.

Distal Diffuse Glands.

Specialised large secretory unicellular glands with indistinct nuclei and cell membranes whose cell bodies lie adjacent to sinus in the Distal F1. subepithelium, and which secrete into the distal Groove F1F2.. The glands have unusual colour reactions to the stains used and these colour reactions are not standard in different species. They stain khaki with M.S.B. and A.B./M.S.B in *P.margaritifera* and *P.maxima* but stain various pink colourings in some of the smaller *Pinctada* with these stains.

Distal Folded Epithelium.

The elongate columnar epithelium covering the Distal Folded Region. The apical membrane bears numerous microvilli which have club-like vesiculate ends which bud-off to form an acid-mucopolysaccharide rich surface secretion.

Distal Folded Region.

The region of the External Mantle lying between the Distal Pallial Region and the Proximal part of LatF1., lateral to the Circum-pallial Nerve and Sinus. The region is characterised by prominent outfoldings with a subepithelium of radiating sinus, fibrous connective tissue and musculature. In the subepithelium are numerous glandular structures some of which are believed to secrete into the radiating sinus.

Distal LatF1..

The most distal of the three regions into which the lateral surface of Marginal Mantle FoldF1. is divided, covered by the Terminal Epithelium.

Distal LatF2..

The most distal of the three histological regions of LatF2..

Distal LatF3..

The more distal of the two histological regions into which LatF3. is divided.

Distal MedF1..

The most distal of the three regions into which MedF1. is divided characterised by the Ovoid Blue Glands, Distal Diffuse Glands and other Granular Cytoplasm Secretory Glands in the subepithelium.

Distal MedF2..

The more distal of the two regions into which MedF2. is divided.

Distal Pallial Region.

In pearl oysters, the most distal of the three regions into which the External Pallial Region is divided just proximal to the Distal Folded Region. There is an increase in both subepithelial secretory glands and subepithelial musculature of the External Mantle of this region compared with that of the Middle Pallial Region.

Dorsal.

With reference to all bivalves by convention the Dorsal border is the border near the hinge.

E.

Extra-pallial Space.

The space (largely a potential space) between the lateral surface of F1. and the overlying medial surface of the Valve. In pearl oysters its proximal extent may be defined by the contact of the folds of the Distal Folded Epithelium with the Mantle. In Bivalves where there is junction of the External Mantle to the medial surface of the Valve at the Pallial Line, this delimits the space proximally. In those bivalves where there is physical continuity between the secretions of the Mantle Margin and the outer surface of the Valve, this delimits the space distally.

F.

F1..

Symbol used here for the lateral Marginal Mantle Fold.

F1. Rami of Circum-pallial Nerve.

Branches of these rami innervate the secretory structures and muscles on both the medial and lateral sides of F1..

F2.

The second most lateral of the two Marginal Mantle Folds in those species with two or three Marginal Mantle Folds, and the third most lateral Marginal Mantle Fold in those species with four Marginal Mantle Folds including an Ancillary F1. Marginal Mantle Fold, i.e., species of the genera *Pteria*, *Isognomon* and *Malleus*. This fold is terminally bifurcate in some of the *Ostreoidea* and trilobate in the *Anomioidea* and *Pectinoidea*. It bears the developing and developed eyes in these superfamilies.

F2. Rami of Circum-pallial Nerve.

These branches of the circum-pallial nerve in the *Pterioidea* alternately leave the distal surface of the parent nerve trunk and then branch into the F1. Rami and the F2. Rami which then pass under the Apex of Groove F1F2., or leave the proximal surface of the parent nerve trunk. In either case they then enter F2. and supply laterals to the glandular and muscular structures of the subepithelium of Proximal LatF2. before innervating the glands and muscles of the Middle LatF2.. Terminal branches supply the secretory and muscular structures of Distal LatF2.. Collateral branches traverse the parenchyma of F2. to supply MedF2..

F3..

The most medial of the three Marginal Mantle Folds of the *Pinctada* and other Bivalves with three Marginal Mantle Folds and of the four Marginal Mantle Folds of *Pteria*, *Isognomon* and *Malleus*.

G.

Granular Cytoplasm Secretory Glands.

All those unicellular secretory glands with granular cytoplasm which occur intraepithelially or with their cell bodies largely in the subepithelium which secrete through the External Mantle epithelium or that of the Mantle Margin.

Excluded are various specialised glands described elsewhere.

Groove F1. Ancillory F1..
The Marginal Mantle Groove between F1. and Ancillary F1..

Groove F1F2..
The Marginal Mantle Groove between F1. and F2..

Groove F2F3..
The Marginal Mantle Groove between F2. and F3..

Growth Processes.
In pearl oysters and some closely related bivalves, protrusions which occur at fairly regular intervals on the periphery of each Growth Scale of the Prismatic Layer. Lines of these Growth Processes of successive Growth Scales radiate from the Antero-dorsal corners of the Valve on the lateral surface.

Growth Scale.
In medial or lateral view, the flattened distorted horse-shoe shaped layer which probably represents a single deposition of Prismatic Layer in pearl oysters. The greater part of the medial surface of the last deposited Growth Scale is visible as the Prismatic Layer forming the peripheral part of the medial surface of a pearl shell Valve. The proximal border usually commences between sheets of Outer Nacreous Layer. On the radial surface, the Growth Scale thickens distally from the pointed proximal border in the medio-lateral dimension, and then decreases again towards its distal extremity so that the radial surface of the entire Growth Scale forms a somewhat distorted spindle. In those species where there is no delimitation of the lateral surface of the Growth Scale and the Major Prisms run directly through the full width of the Prismatic Layer, (eg. *Pinctada fucata*), the outer limits of each Growth Scale are commonly indicated by variations in staining affinities between the outer surface of one growth Scale and the inner limit of its predecessor.

H.

Hinge.

The protein rubber junction between the two Valves just inside the Dorsal Border.

I.

Inner Fibrous Sheath.

Fibrous covering of medial surface of the Prismatic Layer whether of Major Prism Layer or Growth Process. The Inner Fibrous Sheath is lacking just distal to the Nacreo-prismatic Junction.

Inner Lateral Structure.

A layer of smaller structures enclosed in organic matrix in the lateral end of Major Prisms beneath the Outer Prismatic End Plates. While these structures are very commonly seen with L.M. of decalcified shells they are very rarely observed with S.E.M of radial broken surface of Prismatic Layer.

Inner Prismatic End Plate.

Thickened covering of the medial end of some Major Prisms and Growth Process Prism.

Intraprismatic Organic Matrix.

Reteform transverse organic matrix dividers in prisms of the Prismatic Layer which delineate medially and laterally the thickened Calcite Tablets of the prisms.

Interprismatic Organic Matrix.

In light microscopy the acid decalcified walls of the prisms consist of parallel slats of transverse fibres. With S.E.M of radial broken prismatic layer three distinct types of structures are revealed:

(a). Reteform Interprismatic Organic Matrix appears similar in composition to Intraprismatic Organic Matrix and is the most commonly seen phenomenon between prisms.

(b). Linear Interprismatic Organic Matrix consists of parallel ridges usually running parallel to the long axis of the prism on which they occur.

(c). Transverse parallel Interprismatic Organic Matrix may be related to that seen with L.M of decalcified shell.

Isthmus.

The raised ridge of tissue in the dorsal midline which runs from the Anterior Mantle Symphysis to the Posterior Mantle Symphysis.

Isthmusistic Epithelium.

In pearl oysters and some other bivalves, (eg. the *Arcacea*), greatly elongate columnar epithelium which covers the dorsal surface of the isthmus and expands down the lateral surfaces of the isthmus in parallel with the overlying expansion of the enlargement of the hinge. It is bounded on either side for its entire length by a deep sulcus into which Granular Cytoplasm Secretory Glands discharge.

L.

Light Blue Glands.

In pearl oysters of the Genus Pinctada, specialised Granular Cytoplasm Secretory Glands which are restricted in their occurrence to the Middle LatF2., where they are found in all species of the *Pinctada* investigated. They are associated in this location with an unusual type of Turquoise Gland. They are fairly refractory to all stains used with which they stain a faint blue.

Linear Pattern Interprismatic Organic Matrix.

See Interprismatic Organic Matrix.

M.

Major Prism.

A single polygonal prismatic structure of a Growth Scale of a pearl oyster Prismatic Layer. Dependant on species and position on the Growth Scale, terminated medially by either the bare medial surface of the Prismatic Layer, an Inner Prismatic End Plate or the latter covered by Inner Fibrous Sheath, and usually terminated laterally by Outer Prismatic End Plate covered by Outer Fibrous Sheath. Major Prisms are distinguished from Minor Prisms and from the Prisms of Growth Processes.

Major Prism Layer.

A layer of the Prismatic Layer formed of either a single Growth Scale, or, where Growth Scales lack physical lateral and medial boundaries, successive fused Growth Scales excluding Growth Processes.

Mantle Edge Gland.

The tissue composed of elongate columnar epithelial cells which covers usually the greater part of LatF1, and is all but ubiquitous throughout the Bivalvia. It is particularly well developed in the *Pterioidea* and even more so in the *Arcoidea*. It has features in common with the Isthmusistic epithelium which it joins near the Anterior and Posterior Mantle Symphyses.

Marginal Mantle Fold.

One of between two and four (dependant on species) folds of tissue which form the distal edge of the Mantle. The Marginal Mantle Folds of the Left and Right Mantles are joined at the Anterior Mantle Symphysis and Posterior Mantle Symphysis

Marginal Mantle Grooves.

The Grooves between the Marginal Mantle Folds into which the products of the adjacent secretory structures are discharged.

Microgranular Glands.

Specialised unicellular glands which occur in the Parenchyma of F2. and F3.

especially seen in *Pinctada maxima*. With Mallorys their cytoplasm stains amber and with Azan bright red. With T.E.M they are large cells with cytoplasm composed largely of relatively very small electron dense granules. They appear to secrete into the sinus of the Fold parenchyma and seem likely to have the function of increasing turgidity of the Folds by the osmotic activity of their secretory products.

Middle LatF2..

Specialised tissue comprising the Middle third of LatF2.. The epithelium in fixed specimens invariably displays plateaux separated by semicircular infoldings in Radial Section. The surface epithelium is distinguished by a dense mat of long cilia and microvilli. The subepithelium is host to Light Blue Glands and specialised Turquoise Glands. Because of location and staining affinities, a role in production of Intraprismatic Organic Matrix and perhaps also Reteform Interprismatic Organic Matrix is suggested.

Middle MedF1..

Specialised tissues comprising the middle third of MedF1. characterised by a distinctive epithelium and subepithelial arrangement of fibrous connective tissue, muscles, sinus and secretory glands.

Middle Pallial Region.

The Middle of the three Regions into which the Pallial part of the External Mantle of Pearl Oysters is divided on histological grounds.

Multivesiculate Cell.

A cell type of the inner layer of epithelial cells of the Omega Gland which may contribute both vesicles and Amorphous Secretion to the formation of the nascent Pleated Secretion in the Apical Channel of Groove F1F2..

Myostracal Prisms.

The aragonite prisms lying in the Inner Nacreous layer whose medial surface is or was the place of attachment of the Adductor Muscle in the *Pterioidea*.

N.

Nacre.

The material of which pearls and mother of pearl is composed in pearl oysters and similar shell layers in other Bivalves consisting of twinned orthorhombic crystals of arogonite which assume a flattened pseudohexagonal or related habit, enclosed in Organic Matrix. Gastropod nacre is differently structured.

Nacreous Layer.

Refers to a shell layer of discrete origin usually comprising many Nacre Sheets.

Nacreous Organic Matrix.

The organic part of Nacre secreted e.g., by the External Mantle of a pearl oyster.

Nacre Sheet.

In pearl oysters and some other bivalves nacre is commonly composed of multiple layers of Nacre Tiles which in radial section look like courses of brickwork. A Nacre Sheet is analogous to one course of brickwork.

Nacre Tile.

The aragonite and Organic Matrix entity usually tending to a pseudohexagon which consists of twinned orthorhombic aragonite crystals and has a flat tile habit. Nacre Tiles are classified as bound if they are fully incorporated in a Nacre Sheet, partially bound if partly loose at the edge of the Nacre Sheet and "free" if they are situate on the lateral surface of the underlying Nacre Sheet, isolated from the bound and partially bound Nacre Tiles which form the edge of their own Nacre Sheet.

O.

Omega Gland.

A specialised epithelial ridge which lies in the lateral aspect of the Apical Groove F1F2. continuously from the Anterior Mantle Symphysis to the Posterior Mantle Symphysis of both Mantles of pearl oysters and some related Genera. It is ubiquitous in those species investigated which have no junction of Valve with External Mantle or Mantle Margin distal to the Adductor Muscle and which have a calcitic Simple Prismatic Outer Shell Layer outside Inner and Outer Nacreous Layers as do pearl oysters. It forms the lateral side of the Apical Channel of Groove F1F2. and its deeper cells are responsible, along with the opposed Dactylocytes for secretion of the nascent Pleated Secretion of Groove F1F2.. Its superficial cells, the Celtic Scroll Cells, secrete large vesicles into proximal Groove F1F2. lateral to the Pleated Secretion.

Organic Matrix.

The Organic constituent of a calcified Shell Layer.

Organic Matrix Precursors.

The specific chemicals secreted by discrete glands and tissues of the External Mantle and Mantle Margin which combine chemically to form Organic Matrix extracellularly. Also the enzymes secreted to catalyse these reactions.

Outer Fibrous Sheath.

The fibrous or pleated superficial sheath which covers the lateral surface of the Prismatic Layer or an equivalent material on the lateral surface of a Prismatic Layer Growth Scale.

Outer Prismatic End Plate.

The thickened plate of Organic Matrix which caps the outer end of a prism whether Major Prism or Growth Process Prism. It is overlain by Outer Fibrous Sheath and thickened internally where it joins the organic matricial walls of the Inner Lateral Structure.

Ovoid Blue Glands.

Specialised unicellular glands which occur ubiquitously in Distal F1. of pearl oysters and some other Genera of Bivalves. They are unusual in that they stain a variety of blue and purplish colours with all other stains used and also stain a strong greenish turquoise with A.B./M.S.B. With this stain they have a small dark red staining spherical nucleus placed at one end of their ovoid cell body. They discharge into distal Groove F1F2.. Their function is unknown.

P.

Pallial Gland.

A dense mass of glandular tissue consisting of the bodies of Trabecular Turquoise Glands and several types of Granular Cytoplasm Secretory Glands lying under the external mantle just distal to the Adductor Muscle.

Pallial Gland Region.

The Pallial Gland, surface epithelium above the Pallial Gland, subepithelial tissues near the Pallial Gland and the opposed Inner Nacreous Layer of the Valve.

Pallial Line.

The junction of External Mantle and medial surface of the Valve in some bivalves which distally limits and encloses the Pallial Space.

Pallial Region.

The tissues and shell structures associated with the Pallial Space.

Pallial Space.

In Bivalves with a junction of external mantle and Medial Valve surface at the Pallial Line, the space enclosed by this junction. In pearl oysters, the Pallial Space is less definitely defined peripherally by the Distal Folded Epithelium. Here, where the Shoulder Region and Pallial Gland Region are distinguished on histological grounds, the term Pallial is reserved for that region between these regions and the Distal Folded Region.

Parallel Transverse Interprismatic Organic Matrix.
See Interprismatic Organic Matrix.

Pleated Secretion of Groove F1F2..
A continuous sheet of secreted material which lies in Groove F1F2. from the Anterior Mantle Symphysis to the Posterior Mantle Symphysis of each mantle in all pearl oysters and some related Genera studied. It is formed in the Apical Channel of Groove F1F2. by the joint actions of the deeper cells of the Omega Gland, the Multivesiculate Cells and the Dactylocytes. Its medial side receives Black Granules from the Black Granule Secretory Cells. It acts as a partition between the secretions of the secretory structures lateral to it from those of those medial to it. Its pleated form allows for maintenance of this partition despite the state of distension or contraction of the Marginal Mantle Folds. It is not thought to have a structural role in pearl shell formation.

Posterior.
With reference to pearl oysters and other Bivalves the border opposite to anterior i.e., aboral or anal.

Posterior Mantle Symphysis.
The part of the Mantle at the Postero-dorsal periphery where the Left and Right Mantle Margins fuse.

Prismatic Layer.
Generally to describe calcified prismatic Shell Layers other than the Nacreous Layers in the Bivalvia. In *Pterioidea* and *Pinnoidea* restricted to the shell layer lateral and distal to the Nacreous Layers. In these Superfamilies the "prisms" of the prismatic layer are irregular polygonal (but tending to hexagonal) structures

with long axes normal to the shell surface. Each Prism is enclosed in Organic Matrix. and in pearl oysters Organic Matrix internal dividers delimit the spaces occupied by the Calcite Tablets.

Proximal LatF1..

The region of the Lateral part of F1. between the Distal Folded Region and the Mantle Edge Gland.

Proximal LatF2..

The region of the proximal third of LatF2. characterised by Black Granule Secretory Cells and Receptacle Glands in some species of Pearl Oyster.

Proximal LatF3..

A region comprising a histologically fairly homogenous region which comprises by far the greater part of the LatF3..

Proximal MedF1..

The most proximal of the three distinct histological regions on MedF1. distal to the Omega Gland.

Proximal MedF2..

The more proximal of the two distinct histological regions on MedF2..

Proximal Pallial Region.

The most proximal of the three histological regions of the Pallial Mantle and the opposing Inner Nacreous Layer of the Valve.

R.

Receptacle Glands.

Specialised apparent indentations in the epithelium of Proximal LatF2. of many species of *Pinctada* and Middle LatF2. of *Pteria* into which subepithelial secretory glands discharge. Presumed to play a role in production of scleroprotein components of the Prismatic Layers of these animals.

Reteform Interprismatic Organic Matrix.
See Interprismatic Organic Matrix.

S.

Shell Layers.
The more or less discrete entities which form the valves of a Bivalve being distinguished from each other either by structure or origin and usually both e.g., the Prismatic Layer of calcite prisms on the lateral surface of a pearl oyster and the Outer Nacreous Layer of aragonite prisms medial to it. The Outer and Inner Nacreous Layers of the *Pterioidea* are very unusual in that the transition from one to the other is indistinct.

Shell Organic Matrix(ices).
See Organic Matrix(ices).

Shoulder Region.
That area bounded by the Isthmus dorsally, the Adductor Muscle ventrally, a line from the anterior margin of the Adductor Muscle normal to the Hinge anteriorly, and from the postero-dorsal margin of the Adductor Muscle normal to the Hinge posteriorly. The term includes the External Mantle epithelium, the subepithelial Shoulder Gland and associated tissues and the opposed Inner Nacreous Layer of the Valve.

Sinus
Usually a blood vessel or fluid filled space in tissues especially the sinus beneath the External Mantle, the Circum-pallial Sinus, and those in the parenchyma of the Marginal Mantle Folds.

Subepithelium.

When used formally this term refers to all those structures beneath a certain specified epithelium whose physiological functioning is connected with that epithelium, or the production or discharge of substances secreted by or through that epithelium.

Sulcus.

A more or less deep groove e.g., in the External Mantle constituting a specific histological entity.

T.

Terminal Epithelium.

The columnar epithelium on the LatF1. distal to the Mantle Edge Gland.

Turquoise Gland.

-Trabecular Turquoise Gland.

A large unicellular secretory gland whose cell body is usually largely subepithelial, which has a prominent centrally placed nucleus and cytoplasmic compartments divided by partitions (trabeculae). The compartments are filled with acid - mucopolysaccharide secretory material.

U.

Unicellular Glands of the Omega Gland.

The group of secretory glands almost invariably comprising both Trabecular Turquoise Glands and Granular Cytoplasm Secretory Glands which discharge into the proximal lateral region of Groove F1F2., between the proximal and distal parts of the Omega Gland.

V.

Ventral.

(border) opposite the Dorsal (border) or in that direction.

Vesicular Microvilli.

Almost the entire external Mantle and Mantle Margin of pearl oysters is covered with an epithelium which forms vesicles on the extremities of a dense mat of apical microvilli which are then released as small secretory vesicles. The function of the resultant secretion is unknown.

Vesiculate Secretion of Groove F1F2..

In pearl oysters this secretion, while not as obvious as the Pleated Secretion, usually lines LatF2, and is secreted by its structures. It is possible that it has different constituents at different times if, as appears likely, there is a batch process formation of the Prismatic Layer. Its lateral surface bears electron dense particles secreted by the Black Granule Secretory Cells.

APPENDIX 2. SUGGESTED USE OF HISTOLOGY OF THE MANTLE AND MANTLE MARGIN OF PEARL OYSTERS AND OTHER BIVALVES IN THEIR HIGHER ORDER CLASSIFICATION.

A2.1 REALIGNMENTS AMONGST THE PEARL OYSTER GENERA *PINCTADA, PTERIA, ISOGNOMON* AND *MALLEUS*.

A.2.1.1. In Vokes, (1980), the genera *Pinctada* and *Pteria* are included in the Family Pteriidae and the genera *Isognomon* and *Malleus* in the Family Isognomonidae. Species of all these Genera have a calcitic Prismatic Layer outside two aragonitic Nacreous Layers.

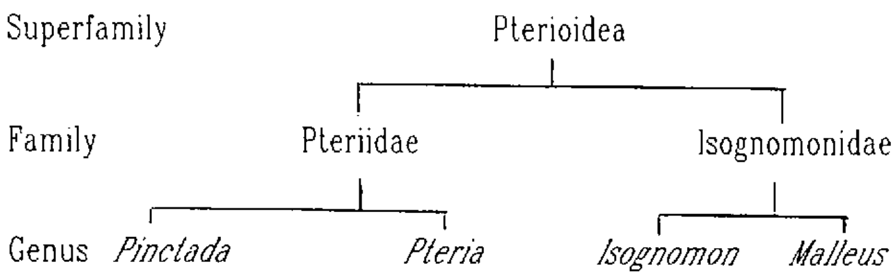


Figure A2.1. Vokes's (1980) classification of the Pearl Oyster Genera.

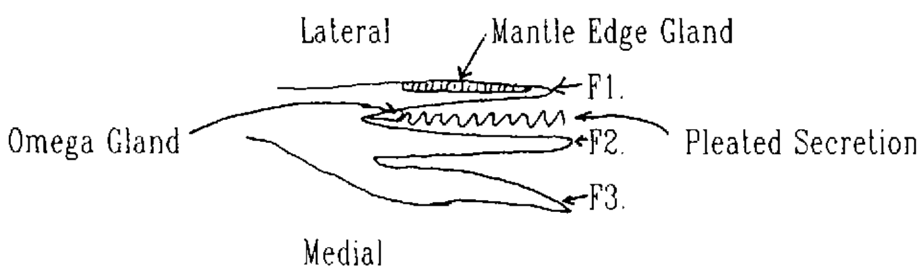
A.2.1.2. Bases for Realignments from Shell Micromorphology, Anatomy and Histology.

While there are some important micromorphological differences between the Prismatic Layers of different species of *Pinctada*, the various parts of the decalcified Prismatic Layer Organic Matrices display remarkably similar staining affinities to a variety of staining procedures.

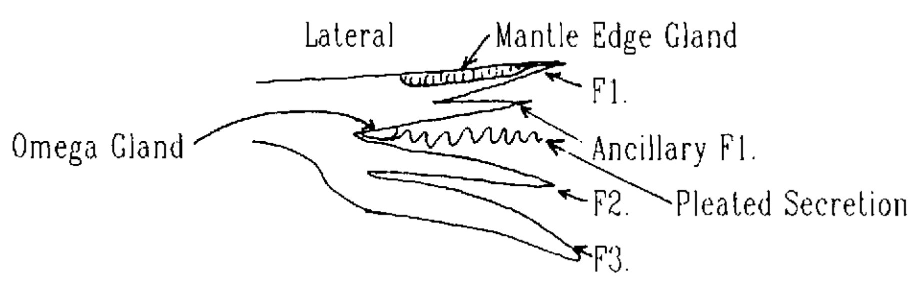
The organic matrices of the decalcified Prismatic Layer of *Pteria penguin* grossly stain similarly to those of the Pinctada with these same stains. However, each part of the organic matrices of the Prismatic Layer of this species with all of these stains displays a brownish tinge not present with any species of the *Pinctada* studied. Further, there are many more Growth Scales of Prismatic Layer superimposed on each other in *Pteria penguin* than in any of the *Pinctada*, and the prisms are smaller in all their dimensions in *Pteria penguin* than in any of the Pinctada in valves of a similar size. The same brownish alteration in colour reactions to the various stains used is seen in the soft tissues of these same animals. While this alteration in staining may well be an artifact resultant from difference in affinity for Bouin's Fixative it is none the less a consistent difference between these genera. All these differences are true not only for other members of the Genus *Pteria* but also for members of the Genus *Isognomon* studied, as for *Malleus alba* The nacre of members of the *Pinctada* studied is much softer than the nacre of the members of the genera *Pteria* and *Isognomon*.

The microanatomy of the Mantle Margin of the members of the Genus *Pinctada* shows three simple Marginal Mantle Folds. Histology reveals the Mantle Edge Gland on the LatF1. and a Pleated Secretion emanating from the Apical Channel of Groove F1F2. between the Omega Gland and the Dactylocytes.

In *Pteria*, *Isognomon* and *Malleus* there are four Marginal Mantle Folds. The Mantle Edge Gland is on the lateral surface of Fl. (LatF1.). The Omega Gland and Dactylocytes together with their Pleated Secretion are between the second most lateral and the third most lateral Marginal Mantle Folds. From a consideration of the histology between the Mantle Edge Gland and the Omega Gland it is concluded that the two outer Marginal Mantle Folds of *Pteria*, *Isognomon* and *Mallens* can be properly considered as Fl. and Ancillary Fl..



Genus *Pinctada*.



Genera *Pteria*, *Isognomon* and *Malleus*.

Figure A2.2. The Mantle Margins of *Pinctada*, *Pteria*, *Isognomon* and *Malleus*.

Pteria and *Isognomon* appear to have quite similar Mantle Margin histologies. *Malleus alba* has a quite different histology in that there are secretory glands in the Mantle Margin which secrete into the Circumpallial Sinus. This in turn lies against the tissues which secrete the Pleated Secretion of Groove FIF2. in this species.

A2.1.3. Suggested Realignment of Pearl Oyster Genera.

On these bases, the Genus *Pteria* is split from the *Pinctada* and included with the Genus *Isognomon* in the Subfamily Pteriinae and the Genus *Malleus* is placed in the subfamily Malleinae. These two subfamilies are linked in the family Pteriidae and the Family Isognomonidae is abolished. The genus *Pinctada* is then placed in the Family Pinctadidae and the Pinctadidae and the Pteriidae become families of the Superfamily Pterioidea.

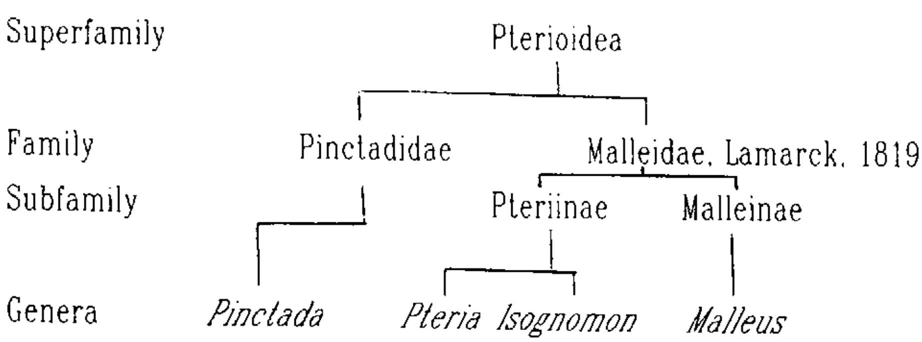


Figure A2.3 Suggested new alignments of the pearl oyster genera.

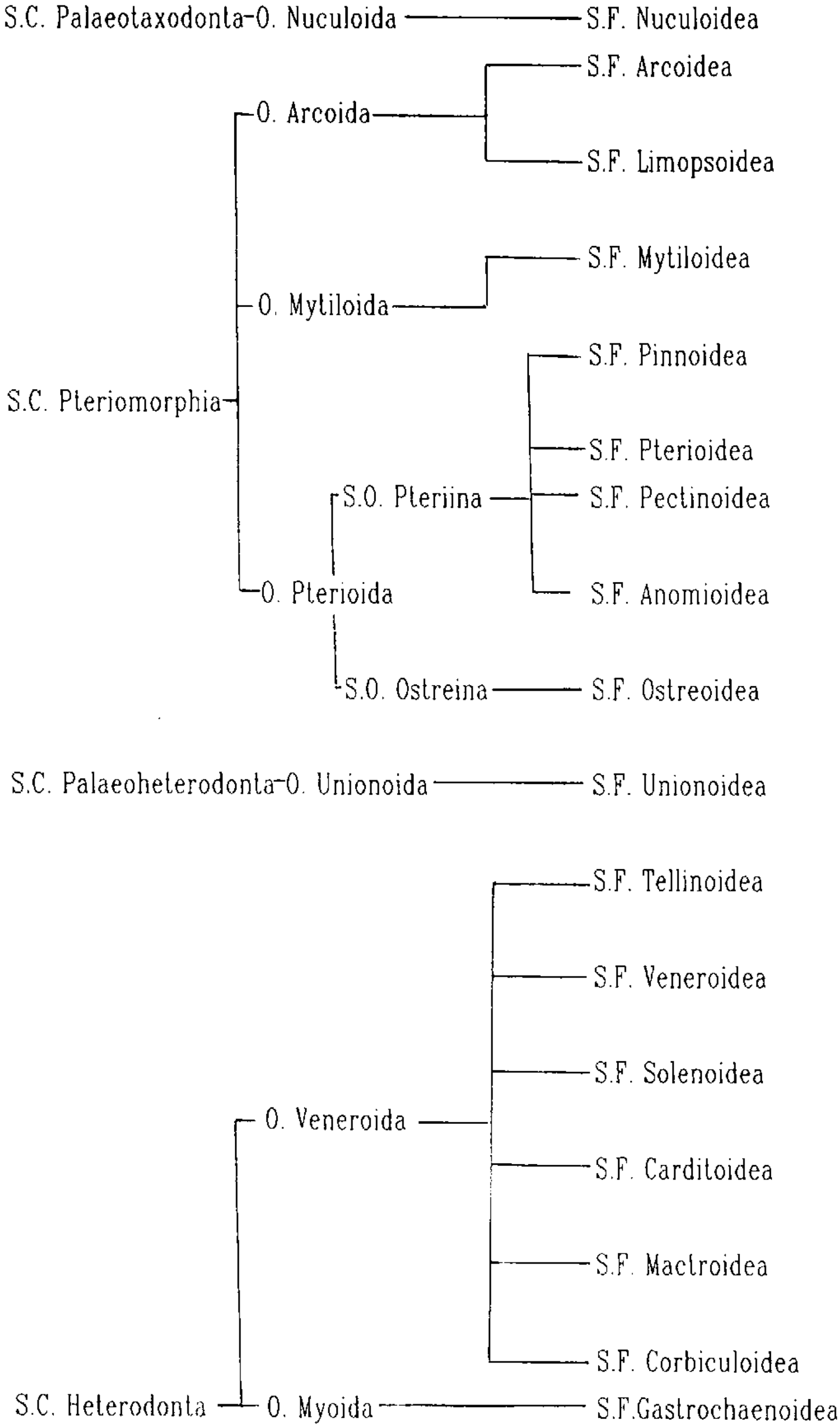


Figure A2.4 Vokes's (1980) classification of relevant superfamilies of Class *Bivalvia.* S.C. = Sub-class; 0. = Order; S.O. = Sub-order; S.F. = Super - family.

A.2.2. ALTERATION IN THE HIGHER ORDER CALSSIFICATION OF SOME BIVALVES SO THAT THEIR TAXONOMIC RELATIONSHIPS REFLECT THEIR DEGREE OF HISTOLOGICAL SIMILARITY.

Basically, the Bivalve classification used for this work is that of Vokes (1980), altered where necessary to accommodate the re-classifiction of the *Pteriomorphia* by Waller (1978). Figure A2.4. shows Vokes's classification of the Superfamilies to which the species used in this study belong. Waller used the Shell Layers described by Taylor (1973) in the construction of a cladogram used in his reclassification of the Pteriomorphia. The following further alterations are suggested by this study of the histology of their Mantles and Mantle Margins.

A2.2.1. Classification from Vokes, (1980) and Waller, (1978) of the Pinnoidea, Pterioidea, Ostreoidea and Pectinoidea.

Waller (1978) groups the Suborder Ostreina and Suborder Pectinina in the Order Ostreoida. He then groups Suborder Pinnina with Suborder Pteriina in the Order Pterioida. The amalgamated Order Limoida and Order Ostreoida is joined with Order Pterioida in the Superorder Pteriomorphia (Figures A2.5 and A2. 9).

Superorder			Pteriomorphia	
Order	Pterioida		Ostreoida	Limoida
Suborder	Pinnina	Pteriina	Ostreina	Pectinina..

Figure A2.5. Waller's (1978) classification of the Suborders Pinnina, Pteriina, Ostreina and Pectinina.

In the oysters, the similarity of the histology of the Genera Ostrea, and Saccostrea sees them grouped in the subfamily Ostreinae, Family Ostreidae.

The dissimilarity of Hyotissa from these sees it placed in subfamily Pycnodonteinae, Family Gryphaeidae (Moore,1971). These families are then placed in the Superfamily Ostreacea.

Superfamily		Ostreoidea	
Family	Ostreidae		Gryphaeidae
Subfamily	Ostreinae		Pycnodonteinae
Genus Ostrea	Saccostrea		Hyotissa

Figure A2.6. Classification of Genera Ostrea, Saccostrea and Hyotissa, (current work and Moore 1971).

Members of the Superfamilies Dimyoidea and Plicatuloidea were not used in this work. The author therefore follows Waller (1978) in placing Ostreoidea, Dimyoidea and Plicatuloidea in the Suborder Ostreina, (Figure A2.7). The histologies of the External Mantle and Mantle Margin of Anomioidea and Pectinoidea accords with Waller's joining of these Superfamilies in the Sub-order Pectinina.

Suborder		Ostreina	
Superfamily	Ostreoidea	Dimyoidea	Plicatuloidea

a.

Suborder	Pectinina	
Superfamily	Anomioidea	Pectinoidea

b

Figure A2.7. Waller's classification of the Superfamilies Ostreoidea, Dimyoidea and Plicatuloidea, in the suborder Ostreina (a), and the Superfamilies Anomioidea and Pectinoidea in the Suborder Pectinina (b).

On the results of this study two major diversions from the cladogram of Waller (1978) are proposed on the basis of "shared derived character states" (Waller, 1978), of the histologies of the Mantles and Mantle Margins.

A2..2..2. Proposed amalgamation of Pinnoidea and Pterioidea in Sub-order Pteriina.

In Waller (1978) the Suborder Pinnina is separated from the Suborder Pteriina on the basis that the Pteriina possess the derived character state of byssal notch and the Suborder Pinnina have the primitive condition of no byssal notch. This is disputed. The byssal notch of the Pinnina is not as dramatic as that of the Pinctada but is none the less present in an attenuated form. In common with the Genera Pinctada, Pteria, Isognomon and Malleus, Pinna possesses a similar Mantle Edge Gland, three Marginal Mantle Folds, a somewhat different but still present Omega Gland producing a Pleated Secretion of Groove F1F2, and the close involvement with the secretory tissues of Groove FIF2. of the Circumpallial Nerve and its branches.

Moreover the secretions of the very similar Mantle Margins in the Pinnoidea and the Pterioidea produce very similar shell outer calcitic Prismatic Layer Organic Matrices.
Because of these strong similarities and disagreement with the basis for separation, it is proposed that the superfamilies Pinnoidea and Pterioidea be joined in the suborder Pinnina.

Suborder	Pinnina
Superfamily	Pinnoidea Pterioidea

Figure A2.8 Suggested alteration to Waller's classification of the Superfamilies Pinnoidea and Pterioidea.

A2..2.3. Proposal to Split Ostreina from Pectinina and Join it with Pinnina.

As described in Taylor (1973) most oysters have an outer layer of calcitic prisms on the upper Valve, and the remainder of the upper Valve and the lower Valve is "foliated calcitic" Shell Layers. When a comparison is made between the secretory tissues and glands which produce the Valve with outer calcitic Prismatic Layer and those which produce the Valve allegedly lacking a calcitic Prismatic Layer either in the same animal or in different species, the latter are found to be more complicated histologically than the former - not the reverse (Chapter 5). It now appears that some species of oyster have calcitic Prismatic Layers on both Valves (Chapter 5. Yamaguchi. 1994). The major morphological differences between the secretory structures of the Mantle Margins of the pearl oysters of Genus *Pinctada* and the oyster is that the oysters have a bifurcate terminal region on F2. (Chapter 5). The Circum-Pallial Nerve and Circum-pallial Sinus may be even more closely involved in the secretion of the shell Organic Matrices of the Ostreoidea than in the Pterioidea or Pinnoidea (Chapter 5).

In Waller (1978) the Suborder Ostreina is joined with the Suborder Pectinina in the Order Ostreoida. The claimed shared derived character states on which this junction is based are two:- Firstly the Ostreina and Pectinina are said to share the derived character state of "specialised, greatly extensible tentacles, differentiated in function and position". The Pinnoidea and Pterioidea, in Waller (1978) joined in the Order Pterioida, (here proposed to be grouped in the Suborder Pinnina), are said (Waller 1978), to share the derived character state of "branching, moderately, extensible, generalised tentacles". While indubitably the species of the Pectinina have "specialised greatly extensible tentacles differentiated in function and position", the tentacles of the Ostreina are closer to the description of "branching moderately extensible generalised tentacles". It is herein held that this is no basis for separation of the Ostreina and Pteriina - rather the reverse.

Similarly the Ostreina and Pectinina are joined by Waller (1978) on the basis of their supposed similar "calcitic shell ultrastructure". They are both said to have "foliated structure" whereas the Pinnoidea and Pterioidea are said to have "simple prisms, no foliated structure".

Most species of the Ostreina in fact have at least on one valve an outer calcitic Prismatic Layer as do all the members of the Pinnoidea and Pterioidea (Taylor 1973). According to Taylor these and the Propeamussium are the only bivalves which have an outer layer of calcitic prisms. Further, the Organic Matrices of the outer Shell Layer of Amusium, if it is "foliated structure", behave very differently under acid hydrolysis from the "foliated structure" Organic Matrices of the shell of Saccostrea, (Chapter 1).

However, far more cogent bases for these amendments to classifcation are the unmistakable similarities in the arrangement of, and types of histological species of the Mantles and Mantle Margins of the pearl oysters and the oysters; and the marked lack of similarities of Mantle and Mantle Margin histologies in either of these groups to those of the Pectens and Spondylids.

In short a consideration of both the secretory structures which produce the shell Organic Matrices precursors and the Shell Layers themselves point to a grouping of the Ostreoidea with the Pterioidea - Pinnoidea, and a separation of all these from the Anomioidea - Pectinoidea.

The Ostreina and the Pinnina are therefore placed in the Order Pterioida and the Order Ostreoida is eliminated, as in Vokes (1980). The Orders Pectinoida, and Pteroida are then grouped in the Superorder Pteriomorphia (Figure A2.10).

A2.2.4. Classification of the Limopsoidea and Arcoidea.

In agreement with Waller (1978) the super families Limopsoidea and

Arcoidea are placed in the Order Arcoida, Super-order Prionodonta. Taylor (1973) suggested that on the basis of Shell Layers these Superfamilies should be classified in the Heterodonta. Similarities in certain aspects of their Mantle Margin histology and the sources of the precursors of the Organic Matrices of their Shell Layers place them close to the Pterioidea, (Figures 5.167 and 5.168).

A2.2.5. Classification of the Mytiloidea.

Similarly the genera Lithophaga, Botulopa and Trichomya are placed in Superfamily Mytiloidea, Order Mytiloida, Superorder Isofilibranchia (Figures A2.9 and A2.10).

These three superorders, Pteriomorphia, Prionodonta and Isofilibranchia are then placed within the subclass Autobranchia (Figures A2.9 and A2.10).

As previously, apart from the features referred to above, the classification used here is that of Vokes (1980) amended in some ways to accord with Waller (1978).

A2.2.6.

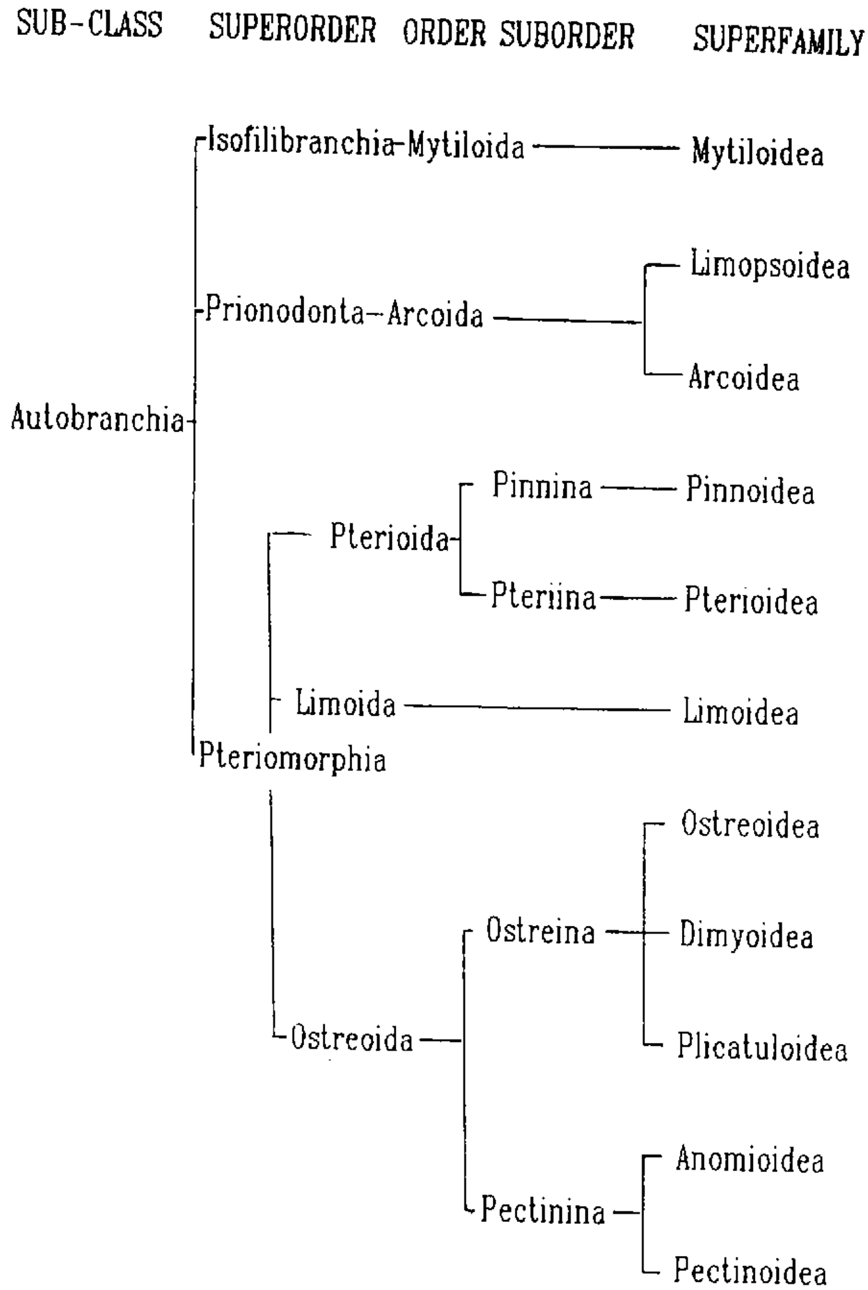


Figure A2.9. Waller's (1978) classification of the *Autobranchia.*

A2.2.7.

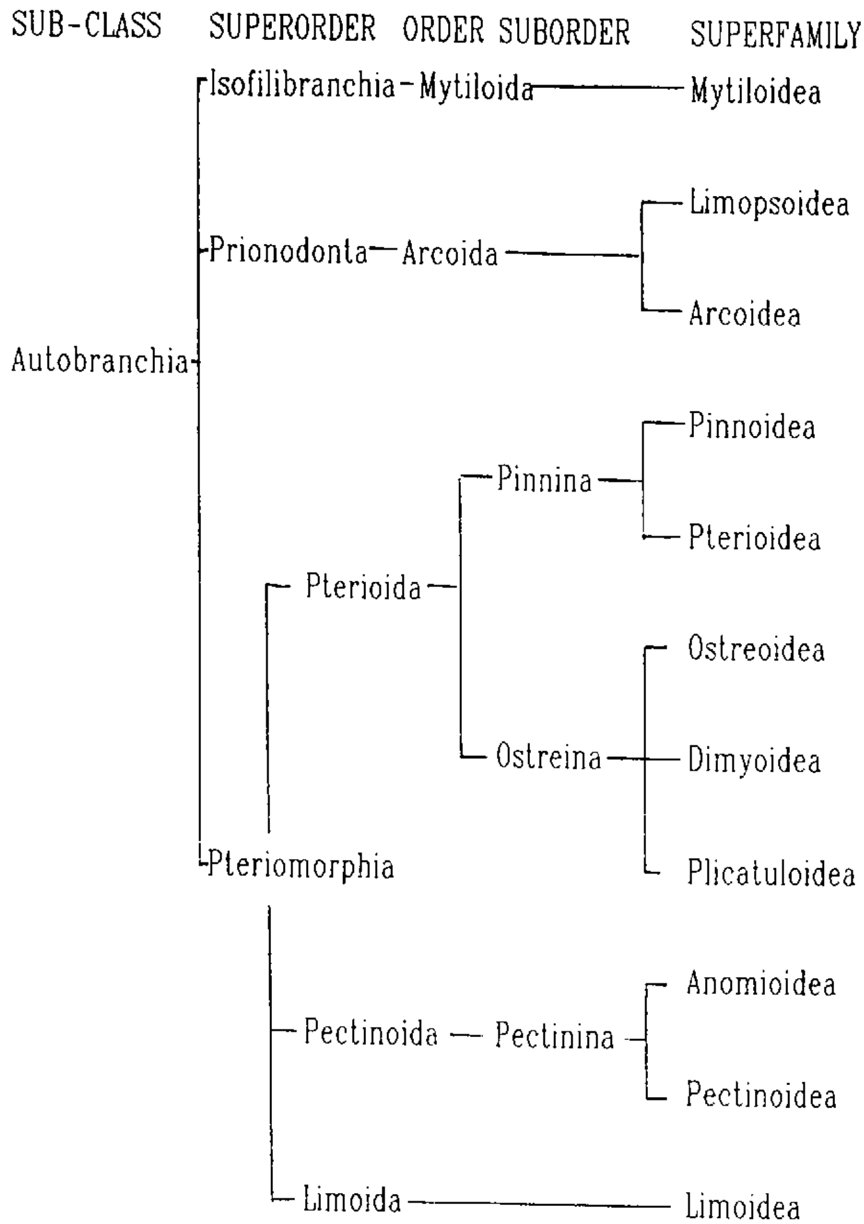


Figure A2.10. Classification of the Autobranchia based on Waller (1978) with two alterations. Firstly, the Pinnoidea and Pterioidea are joined in the Suborder Pinnina. Secondly, the Pectinina and Ostreina are joined in Order Pterioida to accord with histological similarities in the External Mantles and Mantle Margins of these taxa, and differences of the histology of the Mantle and Mantle Margin between the Ostreina and Pectinina.