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RESEARCH

Evaluating the palatability of Tasmania's native sea grapes (*Caulerpa* spp.): a potential new seafood

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Abstract

We assessed the palatability of four Tasmanian *Caulerpa* sea grape species: *C. geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula*, and quantitatively evaluated the palatability profiles of these species with commercially available *C. lentillifera*. Initial palatability was assessed through a hedonic human sensory evaluation, followed by quantitative instrumental sensory evaluation of tastes and volatile organic compounds using an electronic tongue (e-tongue) and headspace solid-phase microextraction gas chromatography–mass spectrometry (GCMS). The hedonic human sensory evaluation found three species to have a positive overall liking of approximately seven out of nine, while the fourth species, *C. hodgkinsoniae*, was significantly less liked overall. This initial evaluation found that the attributes of taste and texture were most influential in driving the separation in overall liking between *C. hodgkinsoniae* and the other, more palatable Tasmanian species. However, the e-tongue found few differences among the tastes of the five species, with most of the significant differences being small in magnitude, and as such the non-volatile component was unlikely to be the reason why *C. hodgkinsoniae* was disliked. Analysis with GCMS, however, found significant differences among the five species, indicating that the aroma profiles of these seaweeds play an important role in their palatability and hence potential culinary acceptance. Overall, our results highlight the potential for the Tasmanian sea grape species *C. geminata*, *C. sedoides*, and *C. simpliciuscula* as comparable to the benchmark for edible species of *Caulerpa*, *C. lentillifera*, given their positive hedonic human sensory evaluation in combination with comparable flavour profiles determined instrumentally by e-tongue and GCMS.

Keywords Seaweed palatability · Sea grapes · *Caulerpa* · Chlorophyceae · Electronic tongue · GCMS

Introduction

Seaweeds (macroalgae) are often touted as a key ingredient in humanity's work to achieve food security and combat diet-related diseases, due to their high nutritional value and ability to be cultivated without taking up significant quantities of land or freshwater (Godfray et al. 2010; Brownlee et al. 2012; Déléris et al. 2016). Globally, seaweeds produced from aquaculture have more than tripled from 2000 to 2022, with 36.5 million tonnes wet weight cultivated in 2022 (FAO 2024). Over 97% of seaweed cultivation occurs in Asia, with over 80% destined for human consumption as foods or phyco-colloids (Loureiro et al. 2015; Cai and Galli 2021). Similarly, interest in seaweeds as a food source is growing in mainstream western cultures, where its consumption has historically been limited to indigenous, traditional cultures (Brownlee et al. 2012; Thurstan et al. 2018; Figueroa et al. 2021).

Several species of commonly consumed *Caulerpa* (Chlorophyta, Bryopsidales) are cultivated and wild harvested across south-east Asia and the Pacific, particularly *C. lentillifera* and *C. racemosa* (de Gaillande et al. 2017; Zubia et al. 2020). These edible *Caulerpa* are commonly known

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in English as “sea grapes”, “green caviar”, or “bubble Caulerpa”, which derive from the distinctive, vesiculate ramuli morphology of the algae along with its unique texture and sea flavour (Paul et al. 2014; Zubia et al. 2020; Amin et al. 2021). Fresh sea grapes are typically consumed accompanied by lime, soy sauce, or coconut milk, or in a fresh salad. They offer consumers high amounts of minerals, vitamins, and dietary fibre, in addition to other less concentrated nutrients such as omega 3 and 6 fatty acids and proteins (de Gaillande et al. 2017). These nutritive properties vary significantly among individuals and species (de Gaillande et al. 2017; Zubia et al. 2020).

Comparisons of red (phylum Rhodophyta), brown (phylum Ochrophyta), and green native Australian seaweeds with red and brown Japanese seaweeds has shown that Australian seaweeds provide similar culinary experiences and nutritional benefits to the Japanese species, though no *Caulerpa* species were involved in this comparison (Skrzypczyk et al. 2019). However, while overall flavour is a primary driver of a food’s acceptance, the modern western diet’s unfamiliarity with the unique aromas, tastes, and textures of seaweeds will likely determine the extent to which they gain acceptance as a food (Chapman et al. 2015; Skrzypczyk et al. 2019; Jensen et al. 2022). Nevertheless, seaweeds are strong candidates for improving food flavour profiles with their relatively high levels of sodium and potassium salts and the sought-after umami compound glutamate (Mouritsen et al. 2019; Figueroa et al. 2021; Jensen et al. 2022).

In terms of food flavour profiles, the electronic tongue (e-tongue) and headspace solid-phase microextraction gas chromatography – mass spectrometry (GCMS) are used to quantitatively analyse the flavour profiles of foods. These instruments respectively measure the non-volatile compounds (taste) and volatile organic compounds (VOCs; smells) of foods and can be used to identify differences in samples to a high sensitivity correlated to palatability (Kobayashi et al. 2010; López-Pérez et al. 2017). The production of taste and aroma related compounds is influenced by the organism’s abiotic and biotic environment, and plays a functional role providing chemical communication, including pheromones for reproduction, grazing deterrents or attractants, immune responses, and competition inhibitors, and some VOCs in turn affect climate functioning (Amsler and Fairhead 2005; Akakabe and Kajiwara 2009; Paul and Pohnert 2011; Garcia-Jimenez et al. 2013). The analysis of seaweeds with e-tongue and GCMS for palatability research is a relatively new field of study, which has potential to uncover new, flavourful species of seaweeds for eating, in addition to quantifying how their palatability profiles might change under certain environmental conditions or preparations (Jensen et al. 2022).

The distinctive grape-like morphology occurs in many species of *Caulerpa* inhabiting tropical and temperate regions of the Pacific Ocean, with south-eastern Australia harbouring an especially high diversity (Womersley 2003; Zubia et al. 2020). Of the thirteen *Caulerpa* species native to Tasmania, southern Australia, six species have vesiculate ramuli: *C. geminata*, *C. simpliciuscula*, *C. vesiculifera*, *C. sedoides*, *C. cactoides*, and *C. hodgkinsoniae*, but none of these are commonly eaten (Scott 2018). Of these six sea grape species native to Tasmania, Australia, the four species *C. geminata*, *C. simpliciuscula*, *C. sedoides*, and *C. hodgkinsoniae* were chosen for this study because these four species occurred in high enough abundance to be included in the various experiments, while the other two species were uncommon. *C. geminata* and *C. sedoides* have medium sized ramuli (3–7 mm) compared to the large *C. hodgkinsoniae* (> 10 mm) and the miniscule *C. simpliciuscula* (< 1 mm) ramuli (Government of South Australia 2010; Scott 2018).

This paper assesses the palatability of four Tasmanian sea grape species *C. geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula* (Fig. 1) and quantitatively compares the palatability profiles of these species with a commercially sourced tropical sea grape *C. lentillifera* cultivated in Vietnam. None of the Tasmanian species had previously been investigated for their palatability. Assessment of palatability was achieved through a hedonic human sensory evaluation followed by instrumental sensory analysis of tastes and VOCs using an e-tongue and GCMS.

Materials and methods

Sample collection

Samples for each species were collected by snorkellers from the following sites in southeastern Tasmania based on their abundance: *Caulerpa geminata* (Charlotte Point: −43.2727° N, 147.1456° E), *Caulerpa simpliciuscula* and *Caulerpa sedoides* (Spectacle Head: −42.8692° N, 147.6076° E), and *Caulerpa hodgkinsoniae* (Fortescue Bay: −43.1260° N, 147.9587° E). Approximate depths for each site were four, one, and two metres respectively. Snorkellers collected a total of 800 g of each species by hand from at least three different patches, with each patch isolated by at least one metre, in order to harvest a representative set of samples for each site. Individuals of each species were pooled together in large, sealed zip-lock plastic bags with natural seawater from the collection site and placed inside insulated cooler boxes containing ice.

Human sensory evaluation

Samples for the human sensory evaluation were collected on 2 February 2023. They were served to participants the following day. Participants ($n = 58$) of mixed gender (39

Fig. 1 The four native Tasmanian *Caulerpa* species investigated: *C. geminata* (A), *C. hodgkinsoniae* (B), *C. sedoides* (C), and *C. simpliciuscula* (D). Scale bar is approximate. Photographs by Micah Landon-Lane

women, 16 men, one non-binary, two undisclosed) and age (21–79 years, median age 30 years) were recruited using email, social media, and word of mouth from the local Hobart area, with screening for age (at least 18 years) and previous consumption of seaweed (at least twice per year). Participants were seated in isolation at the Institute for Marine and Antarctic Studies, University of Tasmania Salamanca building in controlled lighting at room temperature of 20 °C. The University of Tasmania Human Research Ethics Committee approved this project.

The samples were served raw, as this is how sea grapes are commonly eaten (de Gaillande et al. 2017; Zubia et al. 2020). Pieces 3–6 cm (1–2 g) in size were randomly taken from the frond tips of the algae and shaken to remove excess seawater. These samples were taken directly from their ziplock bag containers filled with natural sea water prior to serving. Discoloured or unhealthy fronds or fronds with epiphytic growth were excluded. All species were served on a white plate, together in fixed positions, with a code representing each species to anonymise species identity. Participants could eat each species in whichever order they wished and could ask for more samples. A survey sheet was provided in which participants evaluated each species against a set of palatability attributes on a hedonic scale of 1–9 (1 = dislike extremely, 9 = like extremely) for eight specific palatability attributes: appearance, smell, colour, texture, taste, sweetness, saltiness, and aftertaste, as well as overall liking. Taste in this evaluation was equivalent to overall flavour and therefore included sample aroma. Water and rice crackers (Sakata®) were available, and participants were encouraged to eat these to help reset palates between servings.

Sample preparation for instrumental sensory analysis

Instrumental sensory analysis was conducted using an e-tongue (Insent TS-5000Z, Japan) and GCMS (GCMS-QP2020 with AOC-6000 autosampler, Shimadzu, Japan) to investigate the tastes and VOCs of the four Tasmanian *Caulerpa* species in addition to the commercially grown sea grape *Caulerpa lentillifera* (Amin et al. 2021; Vilar et al.

2021; Jensen et al. 2022). The *C. lentillifera* was imported from Vietnam by Sydney Sea Grapes (https://sydneyseagrapes.com.au/). These brine-dehydrated sea grapes were rehydrated by rinsing and then soaking in distilled water for three minutes.

Samples used in the instrumental sensory analysis were collected on two occasions. The first set of samples were obtained by setting aside approximately 50 g of each species from the human sensory evaluation. These samples were frozen on 6 February 2023, at −18 °C and subsequently used in the instrumental sensory analyses approximately five months later. The second collection of samples took place on 22 June 2023, from the same sites and depths as previously. After collection, these samples were stored in sterile aerated seawater at 12 °C for four days until transportation to the sensory equipment in sealed bags of sterile seawater on ice in insulated cooler boxes, where half were subsequently refrigerated at 1 °C until use the following day, and the other half were frozen. Fresh and frozen samples of the four Tasmanian *Caulerpa* species *C. geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula* were analysed in order to test whether the freezing process significantly altered flavour profiles and to compare with the frozen samples from the human sensory evaluation. Both types of frozen samples were thawed at room temperature prior to sample preparation and analysis by the e-tongue. The instrumental sensory analyses took place five months after the human sensory evaluation; these samples had therefore been frozen for five months by the time they were thawed and prepared for analysis.

Three replicate samples were run in the e-tongue and GCMS, except for the human sensory evaluation samples, where there was only enough seaweed for one replicate, and *C. lentillifera*, which, due to its role as a reference sample, was replicated four times in the e-tongue and three times in the GCMS. Thirty grams of each sample were shaken to remove excess liquid, combined with 150 mL of distilled water, and blended in a 900 Series Nutri-Bullet for 10 s. Each sample was then centrifuged for 10 min at 4,400 rpm before being filtered through a paper filter (Kimtech) to remove solid pieces of *Caulerpa*. For the e-tongue, 70 mL of this liquid was put into 2 × 50 mL plastic cups. For the GCMS, an 8 mL aliquot of this liquid was placed into 20 mL glass vials with air-tight PTFE-faced screw cap lids and frozen at −18 °C until analysis in the GCMS. Air blank (i.e., control samples; $n = 3$) were also analysed in the GCMS to ascertain any background levels of VOCs.

Electronic tongue

An e-tongue is an instrument comprising individual sensors designed to quantify the various tastes experienced by a human tongue, in order to measure differences between foods and drinks. The general operation of the e-tongue has been thoroughly described previously (Hayashi et al. 2013). In brief, the e-tongue measured nine tastes for each sample: sourness, sweetness, saltiness, richness, umami, bitterness, astringency, and bitter and astringent aftertastes using artificial lipid membrane sensors (Zhu et al. 2020). The tastes were quantified by measuring the potentiometric difference between each sensor and a reference electrode. Richness, and aftertastes of bitterness and astringency were derived by measuring the remaining potentiometric difference on the umami, bitterness, and astringency sensors, respectively, after three seconds of rinsing. Sensors were washed between sample measurements. Samples were analysed in a random order with nine samples measured per batch.

Headspace solid-phase microextraction gas chromatography-mass spectrometry

Samples were analysed in the GCMS following the methods of López-Pérez et al. (2017), with the following modifications: samples were equilibrated at 50 °C for 15 min, after which time the solid phase microextraction fibre (SPME; RESTEK SPME arrow 1.10 mm wide, phase thickness 120 µm, phase length 20 mm, carbon WR/PDMS, Cat # 27,487, Switzerland) was inserted to the headspace of the vial. Compound extraction from the headspace occurred at 50 °C for 30 min. Compounds were desorbed from the fibre for 2 min at 250 °C in the injector port of the GCMS (splitless). The GC-QP2020 was fitted with a single quadrupole mass spectrometer (Shimadzu, Japan) and a Shimadzu SH-Rxi-5-Sil MS column (USA; 30 m length, 0.25 µm thickness, 0.25 mm diameter). The oven temperature protocol began at 50 °C for 2.5 min, increased to 90 °C at 3 °C min^{-1}, to 140 °C at 6 °C min^{-1}, to 180 °C at 2 °C min^{-1}, to 230 °C at 20 °C min^{-1}, and held at 230 °C for 15 min. The interface temperature was 250 °C, the solvent cut time was 2 min, and the scan range was between 35–450 m/z. VOCs were carried by helium with a constant flow rate of 1.4 mL min^{-1}. Total run time was 62 min.

Analysis of the raw GCMS data was conducted in the program GCMS Postrun Analysis (Shimadzu), where VOCs were isolated, and their peak area (total ion count) was used as a semiquantitative measure of the concentration of each compound. VOCs were identified by matching the compounds with the reference mass spectra and retention times of the NIST 17 and FFNSC 3 mass spectral libraries. Values for the peak areas of any compounds of interest detected in the air blank samples were averaged and subtracted from the values for the associated compounds of each species. Abundance of VOCs was calculated by averaging the peak area of each VOC across fresh samples, ignoring instances of VOC absence. Presence/absence results for each VOC in each species was done by pooling across all treatments

(Supplementary Material I). Description of VOC aromas was first searched for in published papers on seafood aromas, but where no description could be found this way, the VOC was searched for in the PubChem (2024) database or lastly in The Good Scents Company (2021). As the aroma descriptions in these latter two databases are taken from isolated VOCs and VOC aromas often differ depending on their concentration and delivery medium (The Good Scents Company 2021), aroma information from these sources is marked with an asterisk to differentiate it from the primary sources.

Statistical Analysis

For the human sensory evaluation, differences in overall liking scores among species were tested using one-factor ANOVA followed by Tukey's HSD post-hoc tests to determine pairwise differences. For overall liking, a transformation ($x^{1.5}$) was done to meet ANOVA assumptions of homogeneity of variances. The degree to which participants liked the combined palatability attributes of each species were visualised with non-metric multidimensional scaling (nMDS), which was created using untransformed data for the eight specific palatability attributes, excluding overall liking. An analysis of multivariate homogeneity of group dispersions (variances) test found that untransformed palatability attribute scores had significant heterogeneity of variances (BETADISPER: $F_{3,228} = 5.759$, $P < 0.001$). As such, data were transformed ($x^{1.5}$), which homogenised the between-group variances (BETADISPER: $F_{3,228} = 1.455$, $P = 0.227$). Following this, the palatability attribute scores were analysed with permutational multivariate analysis of variance based on Euclidean distances (PERMANOVA; permutations = 999) and a SIMPER analysis to see how much each variable contributed to the separation of each species. Finally, the specific palatability attributes were tested post-hoc with Tukey's HSD to determine where species differentiation existed among the attributes (significance level $P < 0.05$).

Initially we tested for differences in taste based on the instrumental sensory analyses of the e-tongue and GCMS multivariate data among treatments (fresh and frozen tissue) and species (*C. geminata*, *C. simpliciuscula*, *C. sedoides*, and *C. hodgkinsoniae*) using a two-factor PERMANOVA, followed by nMDS and SIMPER (permutations = 999). Both the e-tongue (BETADISPER: $F_{7,16} = 0.729$, $P = 0.651$) and the GCMS multivariate data (BETADISPER: $F_{7,16} = 1.358$, $P = 0.288$) were not transformed prior to analysis by PERMANOVA as group variances were not significantly different. Second, because *Caulerpa* is consumed fresh, we also tested for differences in individual taste attributes of fresh tissue among the four Tasmanian species and *C. lentillifera* using 1-factor ANOVAs followed by Tukey's tests and visualised separation among these species with nMDS. Finally, we ran a third nMDS visualising separation among fresh, frozen, and human sensory evaluation tissue pooled across the Tasmanian species. We pooled across species for each treatment as we only had a single replicate for each species for the human sensory evaluation samples. For the GCMS data, VOCs significantly ($P < 0.05$) associated with a particular species were ascertained by visual inspection of the vectors correlated with a species in nMDS plots. Significance level for vectors in nMDS plots were set individually in order to maximise the number of vectors without cluttering the plot. Post-hoc Tukey's HSD analyses were conducted on e-tongue data for fresh samples only, including *C. lentillifera*, and fresh and frozen treatment types, excluding *C. lentillifera*.

All statistical analyses were done in RStudio (version 4.1.2).

Results

Human sensory evaluation

For overall liking, *C. hodgkinsoniae* scored significantly lower (mean score of 4.46; ANOVA: $F_{3,228} = 49.15$, $P < 0.001$; Fig. 2) compared to the species *C. geminata*, *C. sedoides*, and *C. simpliciuscula*, which all scored similarly for overall liking (mean scores of 7.14, 7.19, and 7.21 respectively, Tukey's HSD: all $P > 0.992$).

There were significant differences in palatability attributes of the human sensory evaluation among the species (pseudo-$F_{3,228} = 16.800$, P(perm) = 0.001; Fig. 3 and Fig. 4). *Caulerpa hodgkinsoniae* scored significantly lower than *C. geminata, C. sedoides,* and *C. simpliciuscula* for

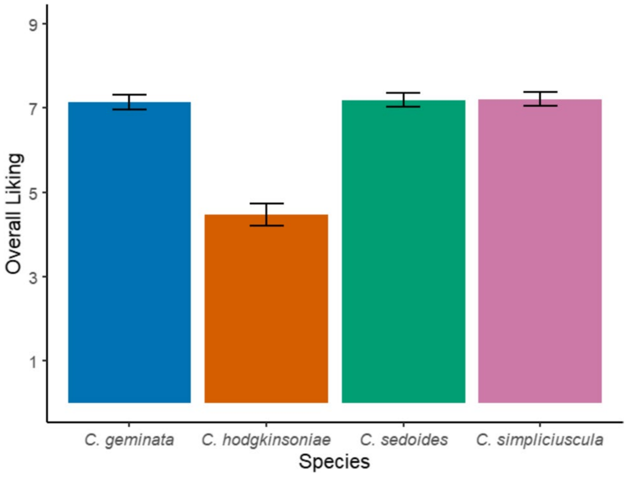

Fig. 2 Mean ± SE scores for overall liking of Tasmanian sea grape species *Caulerpa geminata, C. hodgkinsoniae, C. sedoides,* and *C. simpliciuscula* ($n = 58$). Overall liking was rated on a nine-point hedonic scale from dislike extremely (1) to like extremely (9)

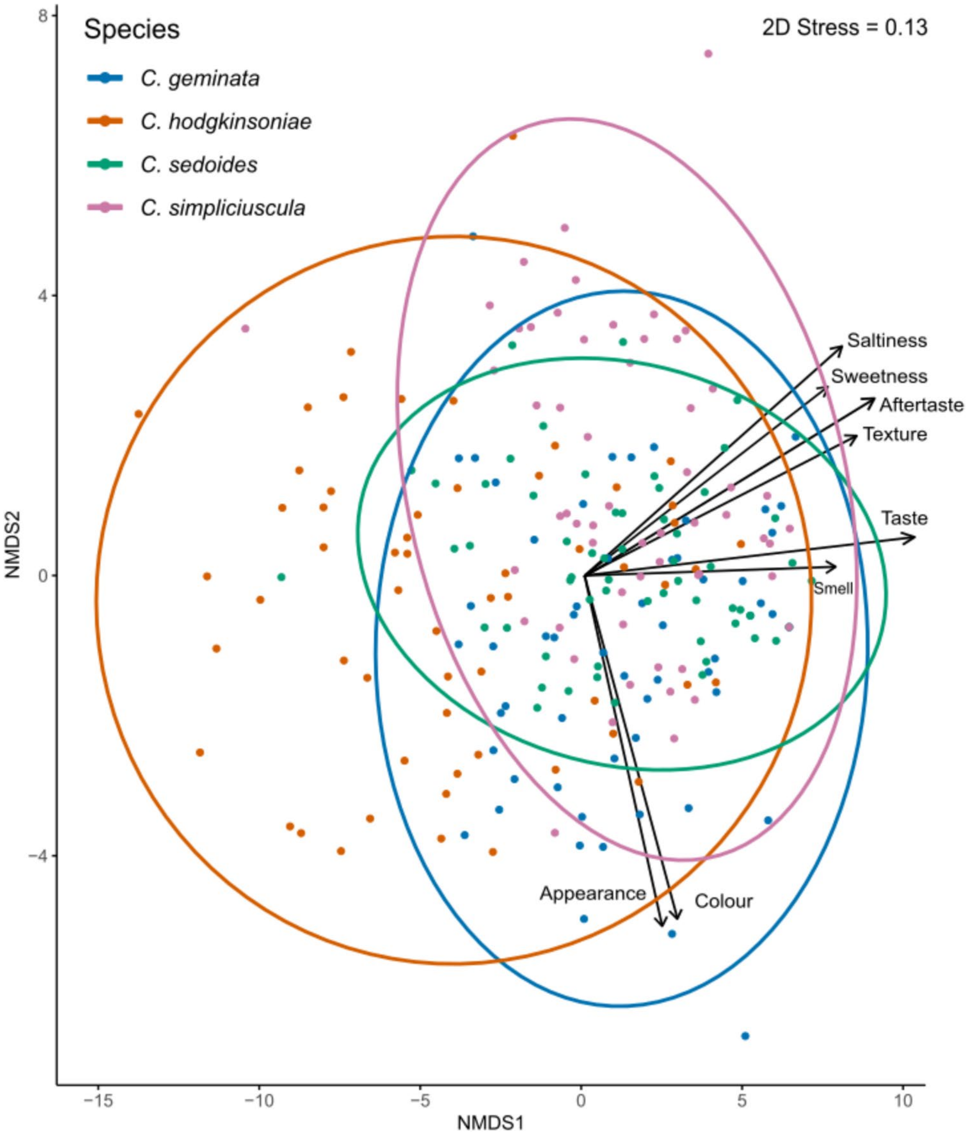

Fig. 3 nMDS plot of hedonic palatability attribute scores from the sensory evaluation for species *Caulerpa geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula*, based on Euclidean distance ($n = 58$). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Arrows indicate direction of increasing palatability attribute score

the taste and aroma related palatability attributes (Tukey's HSD: all $P < 0.001$). Texture, followed by taste, were the biggest contributors to the differences observed between *C. geminata, C. sedoides,* and *C. simpliciuscula* versus *C. hodgkinsoniae*, with mean scores for texture of 6.85, 7.22, 7.55, and 4.21 respectively (ANOVA: $F_{3,228} = 39.230$, $P < 0.001$), contributing to 16.11, 17.01, and 17.53% respectively for differences between *C. hodgkinsoniae* and the other three species (SIMPER). Mean scores for taste were 6.90, 6.98, 7.14, and 4.28 respectively (ANOVA: $F_{3,228} = 40.100$, $P < 0.001$), with the metric separating *C. hodgkinsoniae* by 15.56, 15.93, and 15.55% respective to the other three species (SIMPER). Conversely, texture and taste varied little among the higher scoring species *C. geminata, C. sedoides,* and *C. simpliciuscula* (Tukey's HSD: texture all $P > 0.17$, taste all $P > 0.86$). Finally, the visual attributes varied little between *C. geminata, C. sedoides,* and *C. simpliciuscula* versus *C. hodgkinsoniae*, contributing just 9.46, 9.20, and 10.26% respectively to the differences for appearance, and 9.24, 8.53, and 8.76% to the differences between these species for colour (SIMPER). These differences were only significant between *C. geminata* and *C. hodgkinsoniae*, where *C. geminata* scored higher in these attributes (Tukey's HSD: appearance $P < 0.001$, colour $P = 0.016$).

E-tongue

Both treatment (fresh vs. frozen, pseudo-$F_{1,23} = 4.582$, P(perm) = 0.006) and species (pseudo-$F_{3,23} = 2.606$, P(perm)

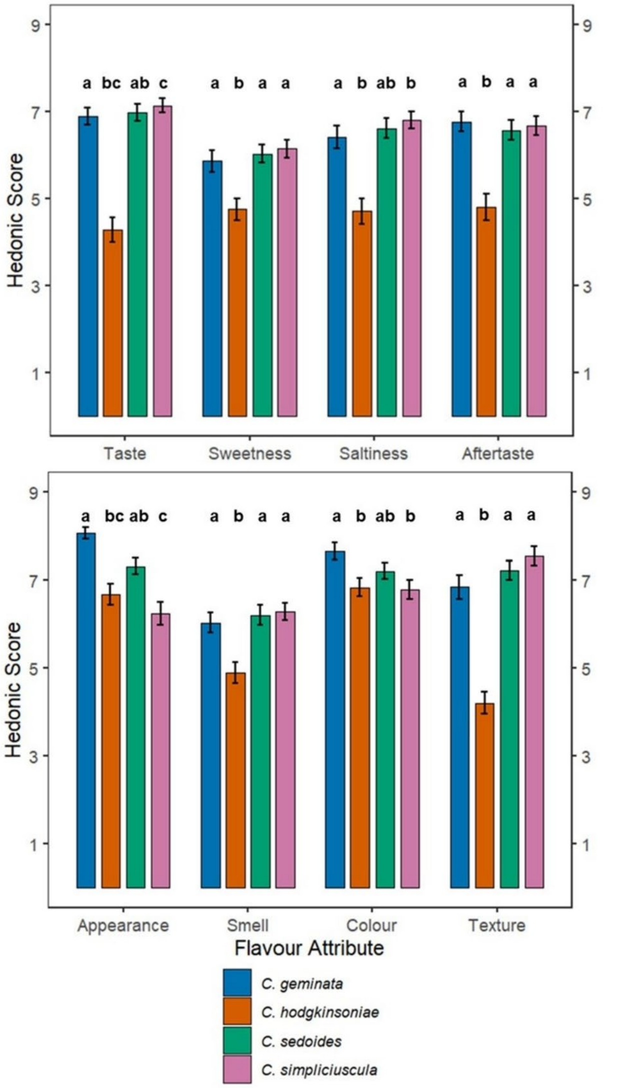

Fig. 4 Mean ± SE scores for each palatability attribute of Tasmanian sea grape species *Caulerpa geminata, C. hodgkinsoniae, C. sedoides,* and C. *simpliciuscula* (n = 58). Palatability attributes are rated on a nine-point hedonic scale from dislike extremely (1) to like extremely (9)

= 0.013) significantly affected e-tongue taste scores with no interactive effect of treatment and species (pseudo-$F_{3,23}$ = 1.591, P(perm) = 0.176). Generally, this analysis and nMDS indicated frozen samples had higher scores for richness and umami but lower scores for aftertastes of bitterness and astringency (Tukey's HSD: all $P < 0.008$; Fig. 5). The nMDS comparing fresh, frozen and human sensory evaluation samples (pooled across species) highlighted a similar separation between frozen and fresh samples (Fig. 6).

The nMDS for fresh samples only highlighted the general similarity among each species' taste profile, i.e., the differences among each species were approximately equal (Fig. 7). Umami (ANOVA: $F_{4,11}$ = 13.410, $P < 0.010$) and sourness (ANOVA: $F_{4,11}$ = 21.160, $P < 0.035$) were the only tastes where any species (*C. lentillifera* in both cases) was significantly different from all other species (Fig. 8). The Tasmanian species were alike in every taste except sourness, where *C. sedoides* was less sour than *C. geminata* (Tukey's HSD: $P < 0.005$).

Saltiness was found to be the largest contributor to separation between the Tasmanian species and *C. lentillifera* (SIMPER). However, the results for saltiness (Figs. 7 and 8) should be treated with caution with regards to *C. lentillifera*, as this species was dehydrated in brine and then rehydrated in fresh water prior to analysis, which almost certainly distorts the results for saltiness. Following saltiness, the second greatest separators between the Tasmanian species and *C. lentillifera* were sourness for *C. geminata* and *C. hodgkinsoniae*, bitterness for *C. sedoides*, and sweetness for *C. simpliciuscula* (SIMPER: 17.78, 15.20, 21.45, and 24.57% respectively). When comparing among Tasmanian species, saltiness was also the greatest contributor to differences between *C. geminata, C. sedoides,* and C. *simpliciuscula* versus *C. hodgkinsoniae* (SIMPER: 34.69, 37.33, and 34.03% respectively).

Gas chromatography – mass spectrometry

As with the e-tongue, both treatment (fresh vs. frozen, pseudo-$F_{1,23}$ = 4.407, P(perm) = 0.017) and species (pseudo-$F_{3,23}$ = 6.394, P(perm) < 0.001) significantly affected the identity and quantity of VOCs, with no interactive effect of treatment and species (pseudo-$F_{3,23}$ = 1.591, P(perm) = 0.176). Generally, this analysis shows greater variability in fresh than frozen samples (Fig. 9), yet the nMDS depicting treatment groups (pooled across species) shows a broad similarity between the three treatments (Fig. 10). Finally, the nMDS of fresh samples shows an association of particularly *C. geminata* and *C. sedoides* but also *C. simpliciuscula*, with *C. hodgkinsoniae* and *C. lentillifera* separating from that grouping (Fig. 11). An aroma description of the VOC vectors in Fig. 11 is given in Table 1 and highlight that the majority of these VOCs are aldehydes, with four of these aldehydes correlating stronger with *C. hodgkinsoniae* and *C. lentillifera*.

A total of fifty VOCs of interest were identified in the five species (Supplementary material I). There were 30 compounds found in *C. geminata*, thirty-eight in *C. hodgkinsoniae*, 33 in *C. sedoides*, 36 in *C. simpliciuscula*, and 27 in *C. lentillifera*. The VOCs identified here can be sub-classed into aldehydes (22), ketones (9), alcohols (6), carboxylic acids (5), esters (4), furans (2), phenols (1), and sulphur-based compounds (1). Five VOCs were found exclusively in *C. hodgkinsoniae* and one VOC,

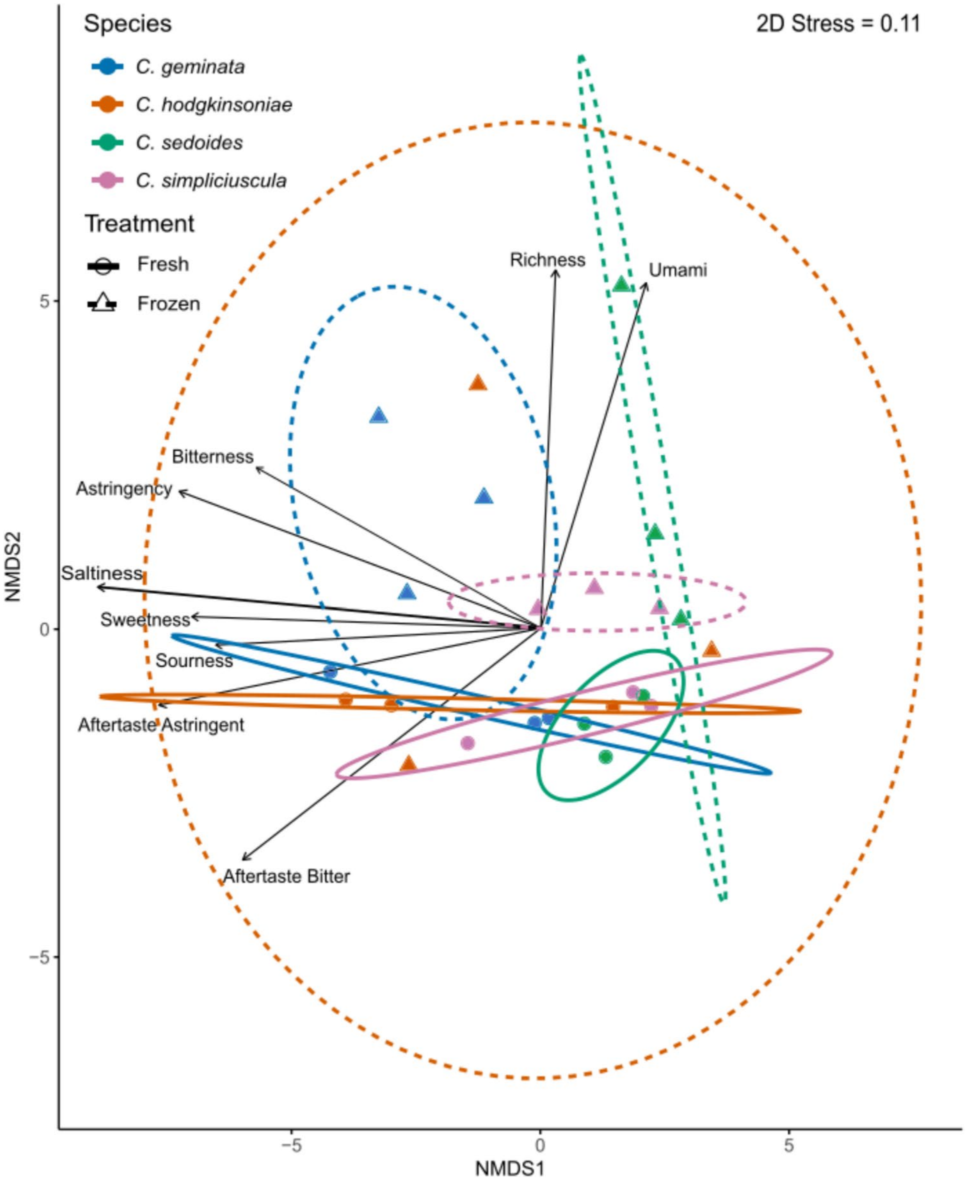

Fig. 5 nMDS plot of two-factor taste scores from the e-tongue for species *Caulerpa geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula*, and treatments fresh and frozen, based on Euclidean distance ($n = 3$). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Arrows indicate direction of increasing taste score

(*E,E*)−3,5-octadien-2-one, was found in all species except *C. hodgkinsoniae* (Table 2). VOCs associated with *C. hodgkinsoniae* and *C. lentillifera* are presented in Table 3 and 4 respectively and again highlight a large presence of aldehyde VOCs.

Discussion

The comparison of four native Tasmanian *Caulerpa* sea grape species found that *C. hodgkinsoniae* is significantly less palatable than *C. geminata*, *C. sedoides*, and *C. simpliciuscula*. This lower palatability of *C. hodgkinsoniae* was primarily driven by lower scores in texture and taste, while the relatively similar visual attribute scores among all Tasmanian species had minimal influence on the overall liking. When considering the instrumental sensory analyses, however, the clear separation of *C. hodgkinsoniae* from the other Tasmanian species was absent when using the e-tongue, and the similarities among the Tasmanian species were generally shared with the tropical sea grape *C. lentillifera*. The GCMS, on the other hand, indicated the lower palatability of *C. hodgkinsoniae* in the human sensory evaluation was, in addition to its unpalatable texture, linked to the VOCs associated with this species.

Taste and texture were the two leading palatability attributes that separated *C. hodgkinsoniae* from the other Tasmanian species in the human sensory evaluation.

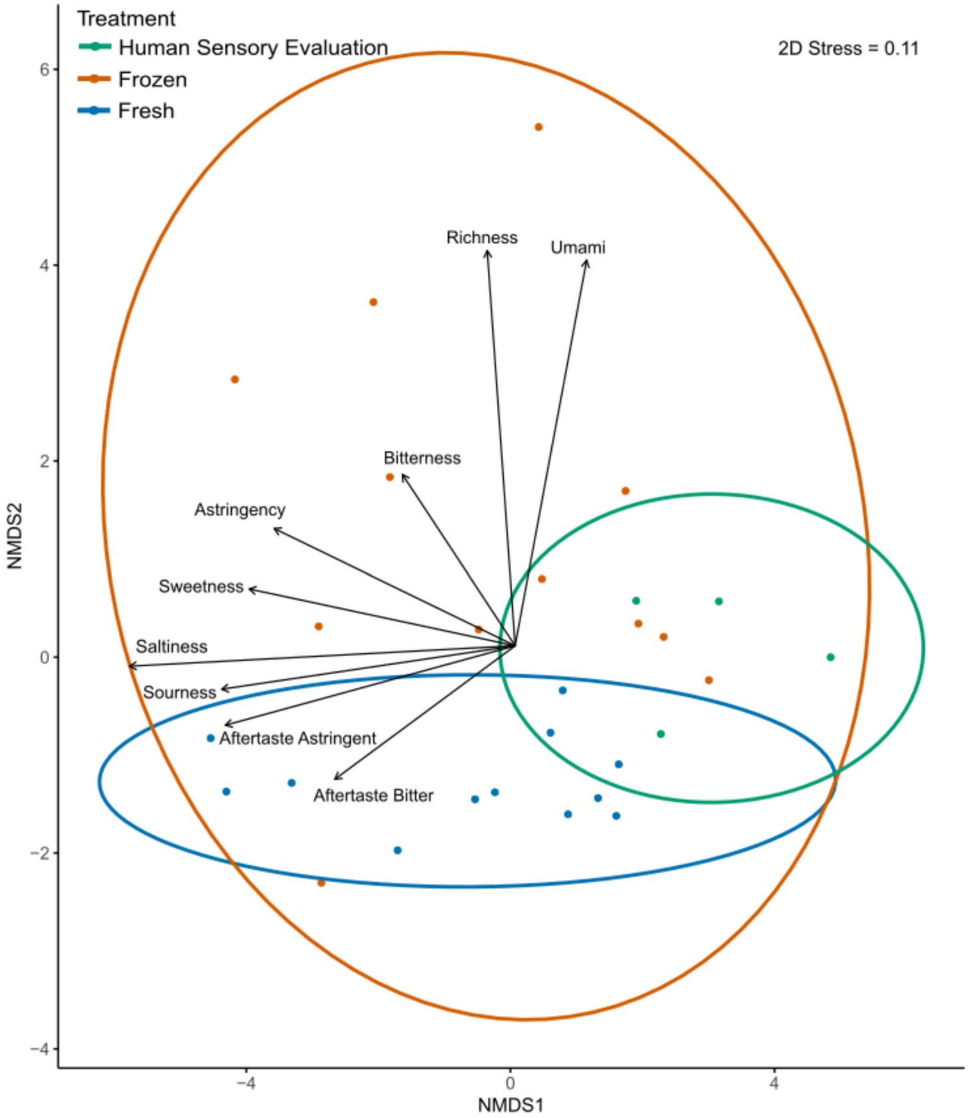

Fig. 6 nMDS plot of taste scores from the e-tongue for the treatment types fresh, frozen, and human sensory evaluation, based on Euclidean distance ($n = 12$ for fresh and frozen, $n = 4$ for human sensory evaluation). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Arrows indicate direction of increasing taste score

An explanation of poor taste is likely nuanced, with the unpleasant taste in *C. hodgkinsoniae* possibly due to the presence of secondary metabolites, such as caulerpenyne, a sesquiterpene, that protect against fouling and grazing (Tejada Gavela et al. 2016; Zubia et al. 2020; Kumar and Sharma 2021). Interestingly, terpenes are important odour and taste compounds, which, in addition to poorer hedonic scoring for taste, could also explain the lower hedonic scoring for smell in *C. hodgkinsoniae* (Cox-Georgian et al. 2019; Garicano Vilar et al. 2020; Moreira et al. 2022). However, while we detected smaller terpenes, such as the monoterpenes dendrolasin, alpha- and beta-ionone, and trans-geranylacetone, these compounds were found in all species, except for dendrolasin, which was only found in *C. geminata* and *C. sedoides*. There is otherwise no information on the presence of terpenes in the Tasmanian species. Regarding texture, participants commented that *C. hodgkinsoniae* had a thicker "skin" than the other species, which left a tough residue in the mouth, whereas the other three species' "skins" mostly disintegrated in the mouth. This is likely related to the larger sized ramuli of *C. hodgkinsoniae*, implying that smaller sized ramuli offer preferable textures. Indeed, in all three palatability analyses, particularly the human sensory evaluation and GCMS, the nMDS consistently showed a closer association between *C. geminata* and *C. sedoides* than with the third palatable Tasmanian species *C. simpliciuscula*. This is despite *C. sedoides* and *C. simpliciuscula* being collected from the same location but emphasises the importance of morphology: *C. geminata* and *C. sedoides* share a similar morphology to *C. lentillifera*. Although the instrumental sensory analyses did not show *C. geminata* and *C.*

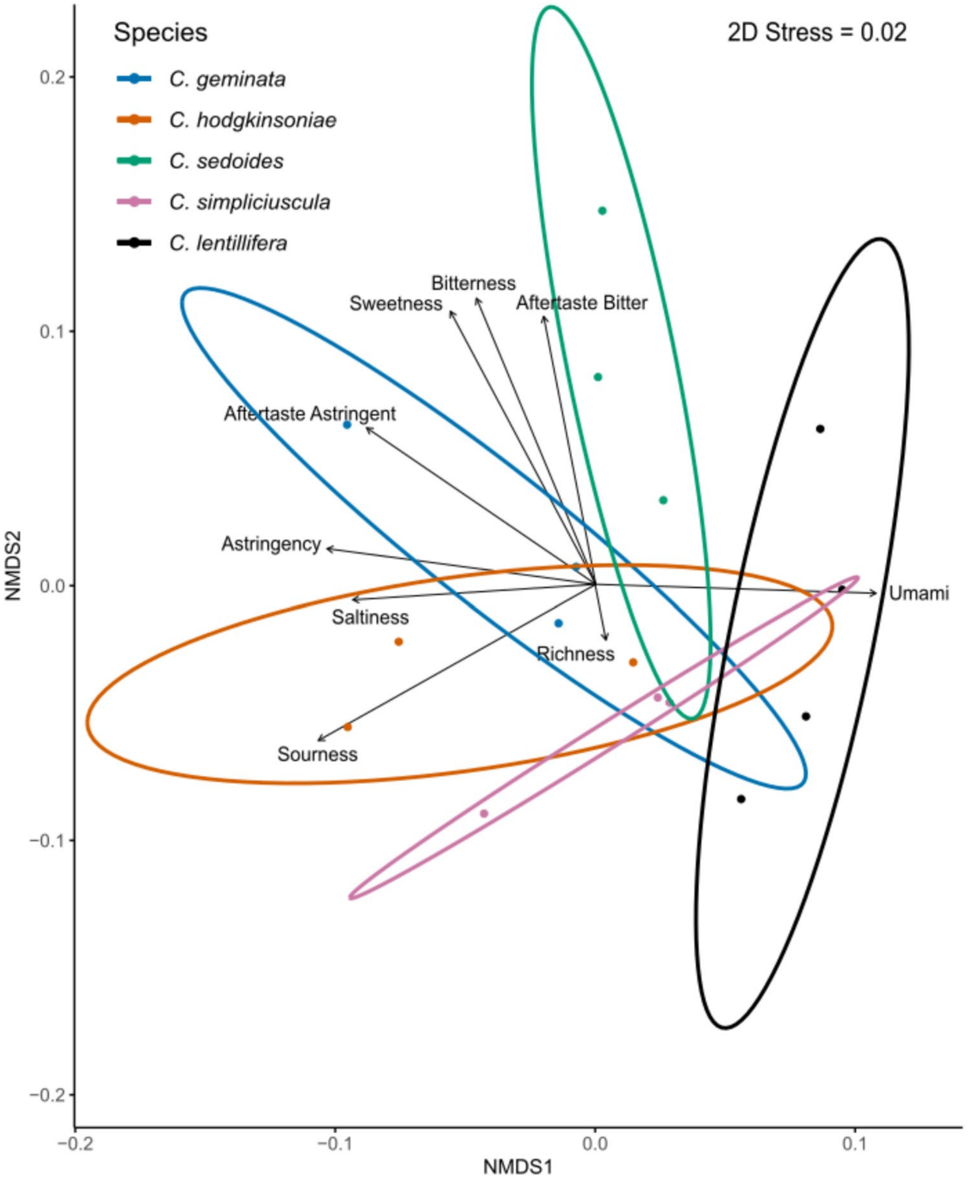

Fig. 7 nMDS plot of taste scores from the e-tongue of the fresh treatment group for species *Caulerpa geminata*, *C. hodgkinsoniae*, *C. sedoides*, *C. simpliciuscula*, and *C. lentillifera* based on Euclidean distance ($n = 3$ for all except *C. lentillifera*, for which $n = 4$). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Arrows indicate direction of increasing taste score

sedoides to be more closely aligned with *C. lentillifera* than the other Tasmanian species, comparisons of fresh and brine dehydrated sea grapes are limited by the effects brine dehydration has on sea grape palatability (Stuthmann et al. 2023).

The results from the e-tongue illustrate few differences in taste profile between the Tasmanian sea grape species and the tropical sea grape *C. lentillifera*, implying Tasmanian sea grapes are a novel marketable seafood and could make ideal substitutes for imported tropical sea grapes from a taste perspective, a result that agrees with a previous palatability study comparing Australian and Japanese seaweeds (Skrzypczyk et al. 2019). Indeed, the few significant differences among the species tested here were often of such a small degree that a human tongue may not notice any difference among the species. For example, comparing mean scores of *C. geminata* and *C. lentillifera* for umami and sourness, the two tastes where *C. lentillifera* was significantly different to all Tasmanian species and most different to *C. geminata*, was just 1.6 and 2.32 respectively (Fig. 8). It has previously been reported that for Insent TS-5000 e-tongues (the model used in this study), a difference in scores of one point is the minimum difference perceivable to a highly sensitive human tongue (Kobayashi et al. 2010). As such, differences of this magnitude are likely indistinguishable to most people. This grouping of taste profiles is particularly apparent among the Tasmanian species, where virtually no significant differences

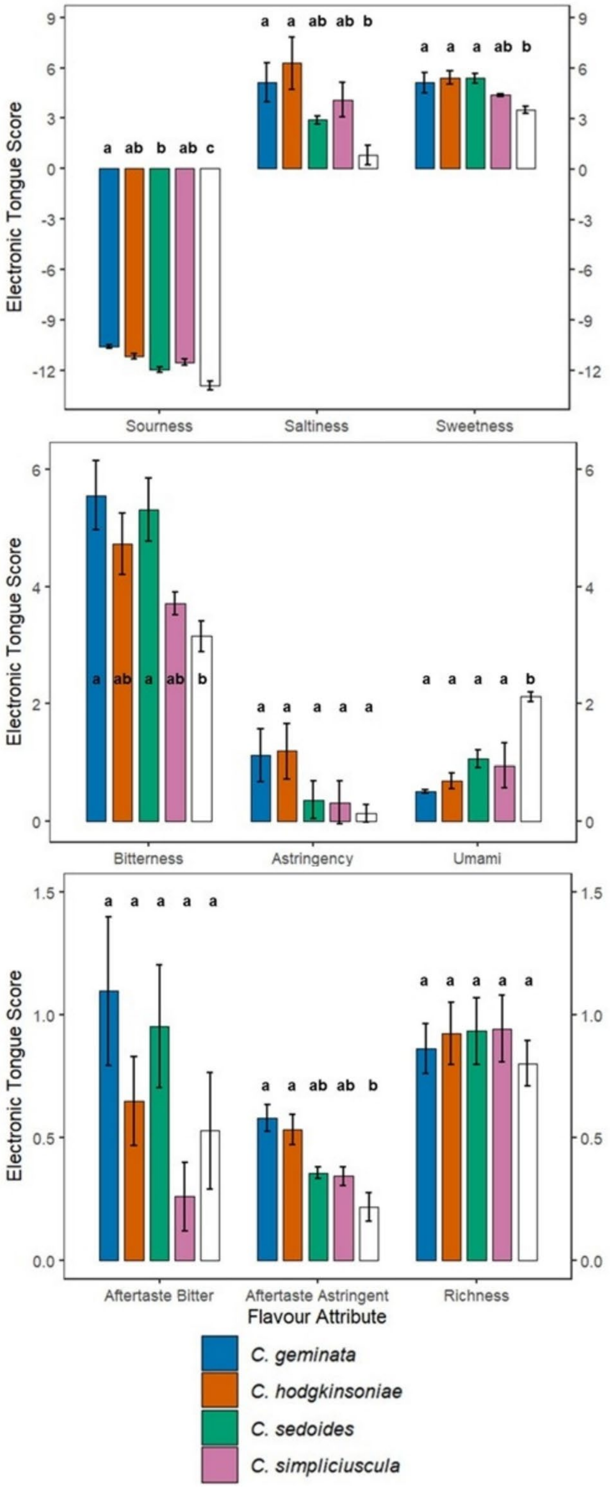

Fig. 8 Mean ± SE taste scores for fresh *Caulerpa geminata, C. hodgkinsoniae, C. sedoides, C. simpliciuscula*, and *C. lentillifera* ($n = 3$ for all except *C. lentillifera*, for which $n = 4$). Shared letters denote significant groupings of species within taste attributes ($P < 0.05$)

of tastes were observed, and notably does not isolate the less palatable *C. hodgkinsoniae*, as would have been predicted after the human sensory evaluation results. Finally, it is worth highlighting that the sensory and instrumental analyses used samples collected five months apart, and that sea grape palatability may vary seasonally (López-Pérez et al. 2017). This could contribute to the low hedonic taste scores for *C. hodgkinsoniae* compared to the similar e-tongue taste profiles for the five species.

The separation in palatability of *C. hodgkinsoniae* was exposed more clearly in the GCMS, where this species was found to contain more VOCs than any of the other species. Many of the VOCs identified in *C. hodgkinsoniae* were aldehydes, which are often pungent aroma compounds that generally offer pleasant odours with longer chain aldehydes and unpleasant odours with shorter chain aldehydes (Garicano Vilar et al. 2020). One of these short chain aldehydes found exclusively in *C. hodgkinsoniae*, for example, was 3-methyl-2-butenal, which has previously been identified contributing to the aroma of dried fish bladders (Table 2; Li et al. 2024). Additionally, *C. hodgkinsoniae* recorded more VOCs than any other species in this study, of which several VOCs were uniquely present or absent. As such, any of the unique VOCs in *C. hodgkinsoniae* could be used as flavour markers, indicating the presence of potentially undesirable flavours. Nonetheless, as the other Tasmanian species differed significantly in their aroma profile but were all considered equally liked for their smell, there does not appear to be a specific formula for a palatable sea grape aroma. Indeed, more research needs to be done before it is possible to relate flavours in seaweeds with specific VOCs (Garicano Vilar et al. 2020).

The total number of VOCs reported here are similar to several other studies of VOCs in non-*Caulerpa* seaweeds, but are noticeably lower than the numbers found in dehydrated seaweeds (Peinado et al. 2014; Yamamoto et al. 2014; Balbas et al. 2015; López-Pérez et al. 2017; Zhu et al. 2021). This is likely due to the types of seaweeds studied and their differing processing methods. However, the types of VOCs identified here were broadly similar to those found in those other studies, such as β-cyclocitral, an oxidative product of the pigment β-carotene, but with the notable absence of any pyrazines, which have never been found in fresh seaweeds, and pyridines and amines, which have rarely been recorded in any seaweeds (López-Pérez et al. 2017; Wang et al. 2022).

The process of freezing samples prior to preparation for the e-tongue and GCMS did alter the aroma and taste profiles of the seaweeds. This suggests that future studies should ideally use fresh *Caulerpa* samples for these

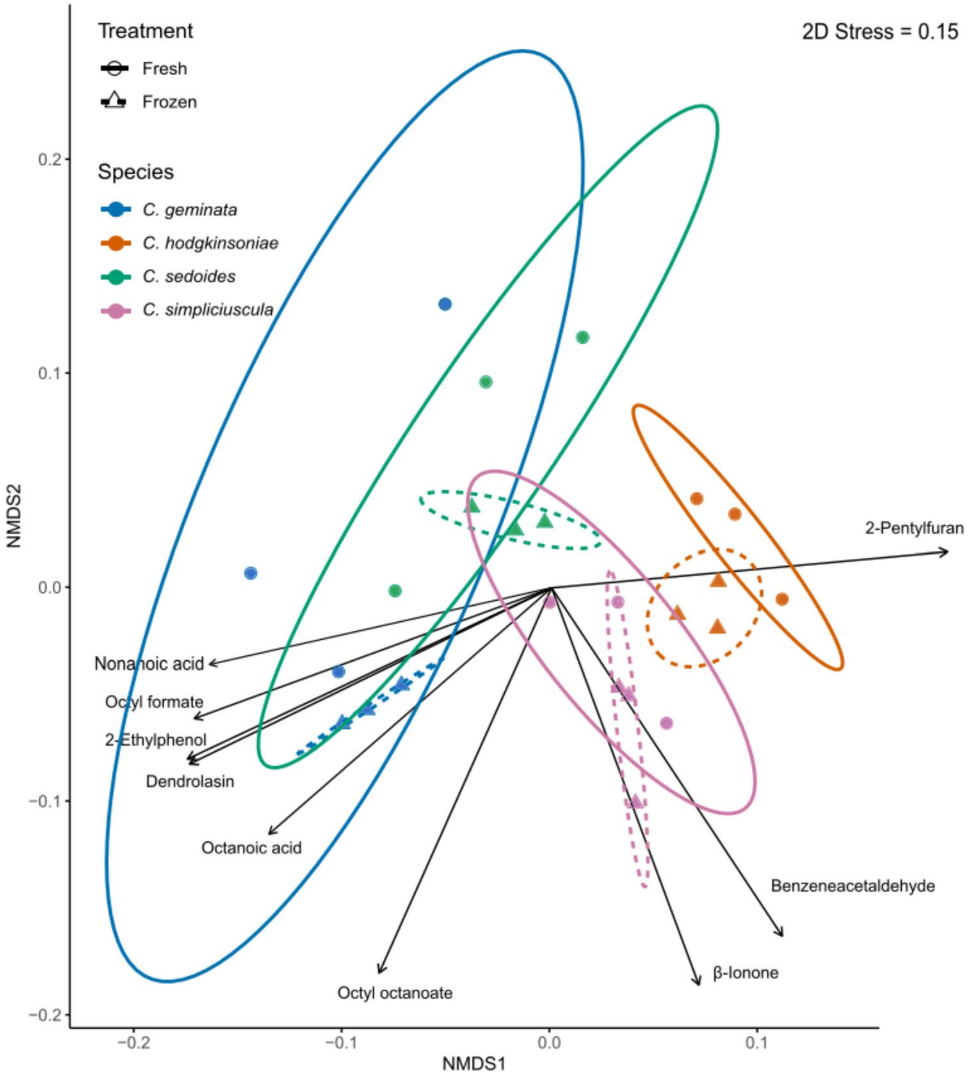

Fig. 9 nMDS plot of two-factor GCMS VOC data for species *Caulerpa geminata*, *C. hodgkinsoniae*, *C. sedoides*, and *C. simpliciuscula*, and treatments fresh and frozen, based on Euclidean distance ($n = 3$). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. VOC vectors shown are to $P < 0.005$ significance. Arrows indicate direction of increasing VOC abundance

analyses, considering tropical sea grapes are mostly recommended to be served fresh (Zubia et al. 2020). Importantly, however, no interactive effect between species and treatment was observed in either e-tongue or GCMS, meaning the freezing process affected palatability profiles for each species similarly; that is, the differences in palatability profiles observed among fresh samples of these species are comparable to the differences observed among frozen samples of these species. Freezing likely affects sea grape palatability by compromising the cell structure and the subsequent leaking of the cytoplasm during the thawing process. This would result in a higher proportion of the solid elements of the cell, such as the cell wall, in the post-processing sample solution, compared to fresh samples. This may be responsible for the increased scores for richness and umami in the frozen samples. Human sensory evaluation samples largely aligned with the frozen samples, particularly with regards to VOC profiles, but the low replication hinders our ability to confidently say if these groups differed significantly. Additionally, given the five month gap between sample collections for the human and instrumental sensory analyses, variations between the two frozen groups could be attributed to seasonal shifts in palatability (López-Pérez et al. 2017). However, though factors such as depth and site are known to influence seaweed palatability for grazing animals, gastronomic research of intraspecific seasonal or spatial variations in palatability of seaweeds is nonexistent (Taylor et al. 2003; Keeley et al. 2015).

Secondary metabolite compounds, sesquiterpenes, and caulerpenyne in particular, are present in many *Caulerpa*, including the widely consumed *C. lentillifera*, and can become toxic when highly concentrated (Nagappan and Vairappan 2014; Tejada Gavela et al. 2016; Kumar

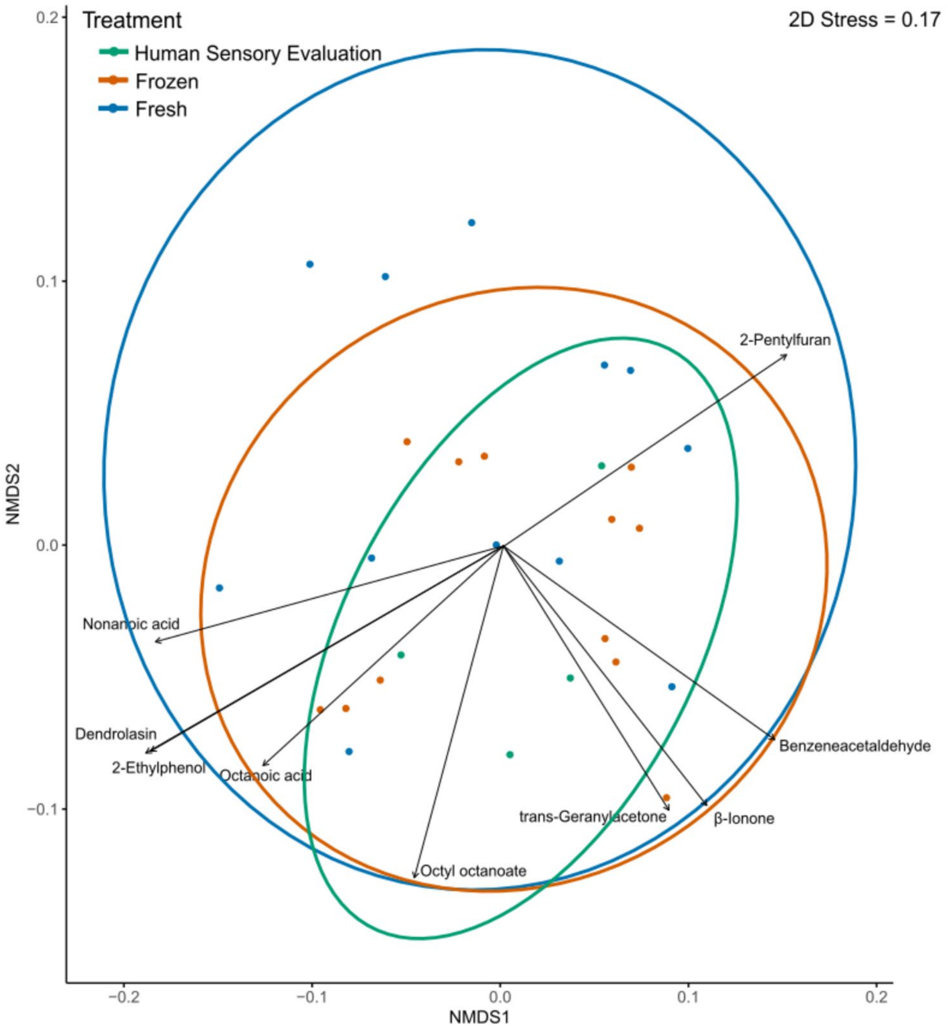

Fig. 10 nMDS plot of GCMS compound data of the three treatment types fresh, frozen, and human sensory evaluation, based on Euclidean distance ($n = 12$ for fresh and frozen, $n = 4$ for human sensory evaluation). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Vector arrows show compounds with a statistical significance of $P < 0.005$. Arrows indicate direction of increasing VOC abundance

and Sharma 2021). There is currently no prescribed safe intake limit for sesquiterpenes (Durán et al. 2021). Despite the defensive biological role of terpenes generally, small doses are unlikely to result in toxicity, evidenced by the long-standing traditional consumption of *C. lentillifera* and other seaweeds (Zubia et al. 2020; Durán et al. 2021; Kumar and Sharma 2021). Indeed, terpenes are well established in traditional and modern medicines (Cox-Georgian et al. 2019). Still, the safety of any novel seaweed should certainly be established prior to its commercialisation as a food. Furthermore, from a culinary perspective, the importance of taste and texture in this human sensory evaluation indicates that it would be prudent to ensure these attributes are prioritised when investigating seaweeds for their potential as novel seafoods.

Overall, the higher sensory scores for *C. geminata*, *C. sedoides*, and *C. simpliciuscula* from the human sensory evaluation indicate that eating these seaweeds was a positive experience. Indeed, given sea grapes are commonly consumed with accompaniments such as lime and soy sauce or in a salad, the palatability scores of these species, which were tasted in isolation, could be improved when combined into a dish. These sensory scores, however, may have been influenced by the fact that only seaweed-eating participants were involved in the experiment; these scores may be different if surveyed by non-seaweed eating participants. Interestingly, the visual attributes of appearance and colour had little influence on each species' overall liking scores; *C. simpliciuscula* had the lowest mean score for both visual attributes but had the highest mean score for both taste and overall liking, though not significantly higher than *C. geminata* and *C. sedoides*. Nevertheless, *C. geminata* attained the highest mean score of any species in any attribute with 8.07 for its appearance, an attribute which is prized in sea grapes and would certainly be selected for, alongside taste and texture, by prospective chefs looking to utilise these novel seafoods (Paul et al. 2014).

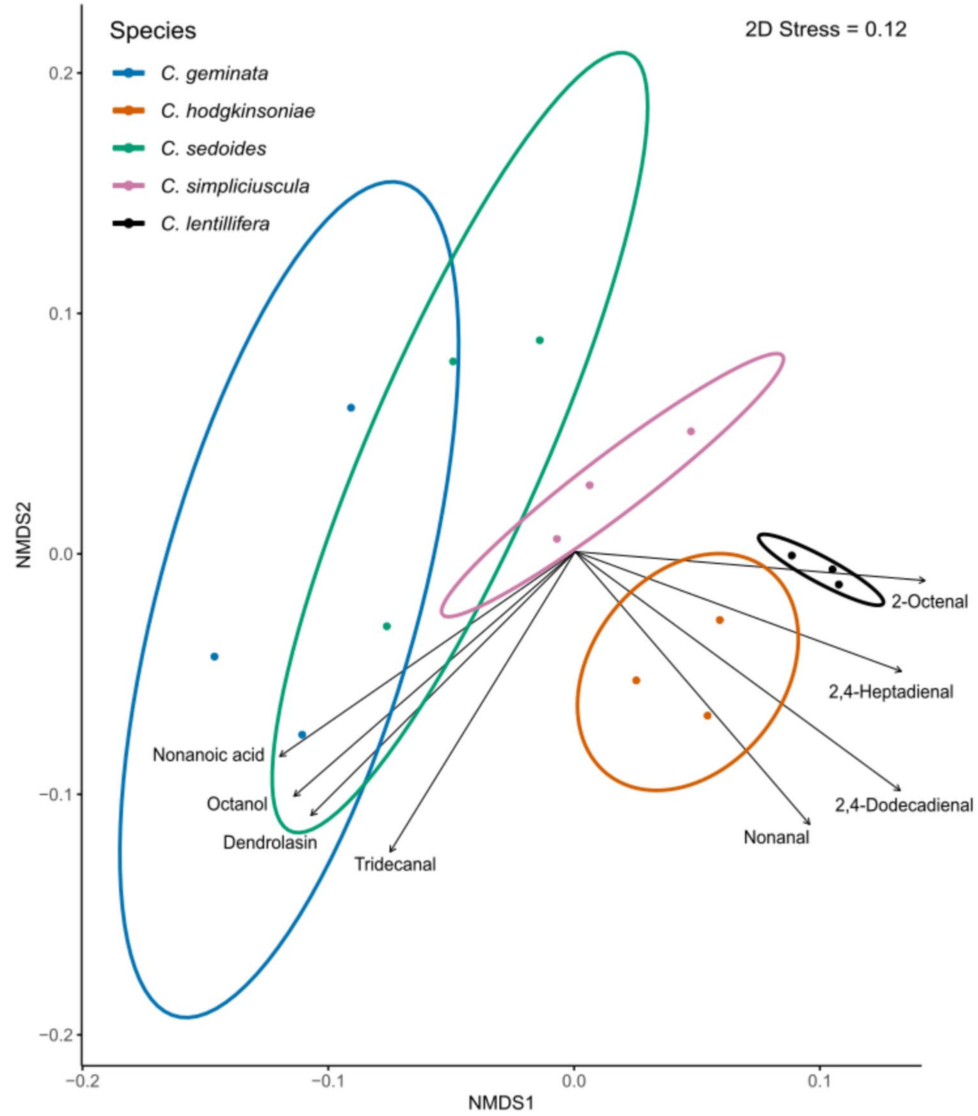

Fig. 11 nMDS plot of GCMS VOCs of the fresh treatment group for species *Caulerpa geminata*, *C. hodgkinsoniae*, *C. sedoides*, *C. simpliciuscula*, and *C. lentillifera* based on Euclidean distance ($n = 3$ for all except *C. lentillifera*, for which $n = 4$). Ellipses depicting group variance are calculated based on the covariance of the data with 95% confidence intervals. Vector arrows show VOCs with a statistical significance of $P < 0.01$. A description of the aroma of each VOC is provided in Table 1. Arrows indicate direction of increasing VOC abundance

Table 1 Aroma description of volatile organic compounds (VOCs) with significant contributions ($P < 0.01$) to separation among five *Caulerpa* species of the fresh treatment, as shown in Fig. 11

Volatile organic compound	Aroma descriptor
Nonanoic acid	Waxy, dirty, cheesy with a cultured dairy nuance*
Octanol	Waxy, citrus*
Dendrolasin	Floral*
Tridecanal	Waxy, citrus, with a hint of grapefruit peel*
Nonanal	Fishy, seafood
(*E*,*E*)−2,4-Dodecadienal	Oily, cucumber, melon, citrus, pumpkin, nut meat*
(*E*,*E*)−2,4-Heptadienal	Fishy, seafood
(*E*)−2-Octenal	Green, spicy, vegetable*

Aroma descriptions for the VOCs come from Garicano Vilar et al. (2020) The Good Scents Company (2021) PubChem (2024) Asterisks indicate aroma descriptors originating from isolated VOCs from PubChem (2024) and The Good Scents Company (2021) databases

Table 2 Volatile organic compounds (VOCs) uniquely present in or absent from *Caulerpa hodgkinsoniae*

Volatile organic compound	Aroma descriptor
3-Methyl-2-butenal	Grassy, found in microalgae
Hexanol	Sweet, green, herbaceous, woody*
2-Pentylfuran	Associated with crab meat and fish soup aroma
4-Methyl-benzaldehyde	Floral*
(*E,E*)−3,5-Octadien-2-one	Pungent herbaceous*
6-Methyl-3,5-heptadien-2-one	Seafood aroma, makes overall seafood smell more pleasant

Aroma descriptions for the VOCs come from (Gu et al. 2013; Garicano Vilar et al. 2020; The Good Scents Company 2021; Moran et al. 2022; Zhang et al. 2022; PubChem 2024). Asterisks indicate aroma descriptors originating from isolated VOCs from PubChem (2024) and The Good Scents Company (2021) databases

Table 3 Aroma description of volatile organic compounds (VOCs) associated ($P <$ 0.05) with fresh *Caulerpa hodgkinsoniae*

Volatile organic compound	Aroma descriptor
Dodecanal	Soapy, waxy, aldehydic, citrus, orange rind with floral nuances*
Hexanal	Fishy, seafood aroma
Nonanal	Fishy, seafood aroma
(*E*)−2-Octenal	Green, spicy, vegetable*
2-Pentylfuran	Associated with crab meat and fish soup aroma
(*E,E*)−2,4-Dodecadienal	Oily, fatty, dairy with a hint of citrus*
3-Octenone	Mushroom, earthy*

Aroma descriptions come from Gu et al. (2013) The Good Scents Company (2021) PubChem (2024). Asterisks indicate aroma descriptors originating from isolated VOCs from PubChem (2024) and The Good Scents Company (2021) databases

Future research should be directed towards the potential for cultivation of the palatable *C. geminata*, *C. sedoides*, and *C. simpliciuscula* species, to see if these species are similarly conducive to propagation from fragments as the tropical sea grape *C. lentillifera*, and what conditions optimise their growth (Paul et al. 2014; Guo et al. 2015). Furthermore, the six VOCs uniquely identified in *C. hodgkinsoniae* should also be investigated as potential markers for palatability, such as to inform harvesting schedules to optimise for aroma (Table 2). Although we solely ascribe the variations in palatability reported here to the factor of species and freezing treatment, there are other factors affecting seaweeds' palatability profiles, such as its geographic origin, time of collection, and method of palatability analysis (López-Pérez et al. 2017). With this in mind, other research areas of interest lie in the nutritional values of these species and whether these values and palatability change seasonally, spatially or with cultivation (López-Pérez et al. 2017; Long et al. 2020).

Table 4 Aroma description of volatile organic compounds (VOCs) associated ($P < 0.05$) with *Caulerpa lentillifera*

Volatile organic compound	Aroma descriptor
(*E,E*)−2,4-Heptadienal	Fishy, seafood aroma
2-Decenal	Fishy, seafood aroma
Pentanal	Coffee, nut-like, chocolate*
β-Cyclocitral	Grassy
(*Z*)−2-Heptenal	Fishy, seafood aroma

Aroma descriptions come from (Peinado et al. 2014; The Good Scents Company 2021; Zhu et al. 2021; Urlass et al. 2023, 2021). Asterisks indicate aroma descriptors originating from isolated VOCs from PubChem (2024) and The Good Scents Company (2021) databases

Conclusion

This study investigated the palatability of just four Tasmanian seaweeds; there are over 750 other seaweed species in Tasmania, many of which have been eaten by the Tasmanian aboriginal community for millennia (Scott 2018; Thurstan et al. 2018; Guiry and Guiry 2023; Hurd et al. 2023). The positive results of this research furthers previous research that supports the potential for native Australian seaweeds as novel seafoods and participate in the rapidly expanding global seaweed market, especially given Australia's relatively unpolluted coasts and diversity of seaweeds (Skrzypczyk et al. 2019; Kelly 2020; Visch et al. 2023). Well planned sustainable seaweed aquaculture may help to offset the rising demand for food and other goods. This could occur in addition to reducing pressures related to food production using agricultural land and freshwater and promoting biodiversity and climate change mitigation (Godfray et al. 2010; Déléris et al. 2016; Duarte et al. 2017; Forbes et al. 2022). The challenge for western cultures lies in supporting and promoting this rise of seaweeds as a food, to the point where seaweeds are taken beyond the traditional foods borrowed from other cultures and combined into original dishes that appeal to the wider cultural palate.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s10811-025-03538-5.

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Authors' contributions M.L-L. constructed the research aims, collected samples, organised and conducted the experiments, processed and analysed the data, prepared the figures and wrote the manuscript text. S.S. advised on the construction of quantitative sensory experiments and associated data analyses. M.W. advised on the construction of the human sensory experiment. M.T., R.dN., C.L.H., and J.W. constructed the research aims. All authors critically reviewed the manuscript.

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Data availability Data can be made available subject to the ethics approval conditions of this work.

Declarations This research was conducted with approval from the University of Tasmania Human Research Ethics Committee under project title 'Palatability of Tasmanian native Caulerpa seaweeds' project ID 27272.

Competing interests Masayuki Tatsumi and Rocky de Nys are both employed at Sea Forest Ltd.

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