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Labeled Hedonic Scale for the Evaluation of Sensory Perception and Acceptance of an Aromatic Myrtle Bitter Liqueur in Consumers with Chemosensory Deficits

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Abstract: Spices and herbs improve sensory perception and acceptance of foods in subjects with chemosensory deficits. Our study demonstrated that aromatic spices/herbs greatly influenced the sensory perception of an aromatic myrtle bitter liqueur (Mirtamaro) in consumers with olfactory and gustatory deficits. Mirtamaro was obtained by infusion of myrtle leaves/berries and a blend of Mediterranean herbs/plants. We initially evaluated differences in gustatory and olfactory perception of pure stimuli in controls (n = 158), subjects with hyposmia (n = 111 participants), and hypogeusia (n = 34). Subjects with hyposmia and hypogeusia showed a marked reduction in odor threshold, discrimination, and identification, while a noticeable compromise in the perception of basic taste modalities (bitter, salty, sour, and sweet) was detected in participants with hypogeusia. Then, in a subpopulation (n = 111) we evaluated differences in the perception of odor and taste pleasantness, intensity, and familiarity of Mirtoamaro. No significant differences emerged, by a labeled hedonic Likert-type scale, in the perception of Mirtamaro odor and taste in subjects with hyposmia and hypogeusia compared to controls. All groups described similar bitter liqueur sensory attributes, qualifying the use of aromatic herbs/plants as a strategy to enhance sensory perception and acceptance of foods in subjects with chemosensory deficits.

Keywords: chemosensory perception; aromatic herbs and plants; myrtle; hyposmia; hypogeusia

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1. Introduction

The relationship between sensory attributes and consumer acceptance has received great attention [1–3]. The major sensory properties of a food product (appearance, aroma, texture, taste, and irritation) are perceived by the basic human senses [3–5]. The sensory experience of eating greatly influences food intake control [1–4]. Sour, salty, sweet, and bitter are usually considered the basic taste modalities [2,4]. Food flavor is perceived through a combination of gustatory, olfactory, and trigeminal stimuli and the nose provides 80–90% of the food taste quality [2,4]. In addition to the sensory characteristics of the product, consumers' responses to food are also related to other factors such as experience, attitudes and beliefs, and physiological status [3].

One of the physiological factors that affect eating behavior is the decline in olfactory and gustatory function [6–11]. Hyposmia is considered a quantitative olfactory deficit indicating a reduced sense of smell [12]. The olfactory function has a crucial role in human life for avoiding potentially dangerous compounds and for the food taste quality detection and evaluation [13]. A deficit in olfactory function greatly affects food perception and subjects with hyposmia showed a decreased flavor perceptive capacity and food enjoyment, and increased difficulties in detecting spoiled and rotting foods [6–11,13,14]. Moreover, changes in dietary behavior (weight gain or loss), problems in personal hygiene, low quality of life, and an increased risk of depression and anxiety have been reported in subjects with olfactory impairment [6–11,13,14]. Hypogeusia is a quantitative gustatory deficit which may induce sufficient discomfort and possible effects on health [15]. Patients with gustatory deficits may exhibit changes in eating habits with appetite loss and malnutrition [9,15]. Decreased gustatory function has been associated with an enhancement in fat intake, body weight, blood pressure, total cholesterol, and an increased risk of cardiovascular diseases [9].

Smell and taste dysfunctions limit an individual's liking for food and beverages, therefore augmented use of salt, sweeteners, fats, and spices is common in consumers with olfactory or gustatory impairments to compensate for their dysfunctions through activation of the gustatory and trigeminal pathways and induction of a hedonic gratification [6,10,13,16,17]. Intensification of flavor with flavor enhancers and herbs/spices has been indicated as a strategy to ameliorate the perception of aroma/taste and acceptance of food/beverage in consumers with sensory-compromised situations [16].

Aromatic herbs and spices represent an important dietary source of chemical compounds (proteins, fibers, vitamins, minerals, phenolic compounds, essential oils) characterized by nutritional and/or nutraceutical properties, with beneficial effects in the prevention of several pathological conditions (inflammation, oxidative stress conditions, cancer, and cardiovascular/neurodegenerative diseases) [17–19]. The use of fresh herbs and spices plays an important role in modern food preparation, due to their ability to impart unique flavorings, contribute color, and positively affect human health [18–21]. The use of certain spices and herbs, alone or in blends, can reduce or replace sugar in foods [22]. The addition of herbs and spices, instead of or in combination with added salt, has been proposed as a strategy for reducing the diet salt amount [20,21,23,24]. Their addition in low-salt foods increases food palatability and consumer satisfaction, compensating for salt, fat, and energy reduction [20,23,24]. Moreover, volatile compounds from aromatic plants/herbs have been demonstrated to positively influence the perception and acceptance of bitter taste [25,26]. A recent study showed that the use of various spices and herbs improves COVID-19-induced anosmia and ageusia [17]. Moreover, the role of Mediterranean aromatic plants in the enhancement of salty perception in patients with hyposmia has been recently assessed [24].

Aromatic bitter herbal liqueurs are constantly dropping their popularity [25–27]. Italian "Amari" are alcoholic aromatic liqueurs characterized by a marked bitter taste, generally consumed before or after a meal due to their digestive and tonic-restorative properties [26,28,29]. Myrtle (*M. communis* L.) is an aromatic plant, endemic to the Mediterranean basin, with various uses in traditional medicine and cuisine [26,28,30]. The island of Sardinia (Italy) has an ancient tradition in the preparation of "Mirto", a typical after-meal liqueur obtained by hydro-alcoholic maceration of myrtle leaves and/or berries, characterized by a bitter flavor, greatly appreciated for its tonic-digestive, anti-inflammatory, and antioxidant properties [26,28]. Mirtamaro is an aromatic myrtle bitter liqueur characterized by a very intense bitter taste and special aroma, obtained by maceration of myrtle leaves/berries and a complex mixture of Mediterranean aromatic/bitter herbs and plants in a hydroethanolic base according to a secret recipe [26]. We recently demonstrated significant sex differences in the sensory perception of Mirtamaro bitterness [26].

Starting from all these considerations, the objective of the present research was to assess differences in the perception of sensory characteristics (odor and taste) and acceptance of the bitter herbal liqueur Mirtamaro in subjects with hyposmia and hypogeusia compared to healthy controls and explore the potential role of the blends of aromatic plants/herbs in modulating the olfactory and gustatory perception in subjects with chemosensory deficits. Most studies evaluating the effect of olfactory/gustatory disorders on food-related experiences have used a self-rated questionnaire, while few studies reported sensory testing using real food samples in patients with sensory impairment [11,14,31]. To the best of our knowledge, no previous work has been reported on the sensory evaluation of a complex aromatic herbal bitter liqueur in subjects with olfactory and gustatory deficits. To this goal, differences in the olfactory and gustatory perception of pure stimuli in subjects with hyposmia and hypogeusia versus healthy controls were initially determined. The olfactory function was assessed by the evaluation of odor threshold (OThr), odor discrimination

(ODi), and odor identification (OId) with the Sniffin' Sticks test [24,32]. The gustatory function was determined by the "Taste Strips" test, consisting of filter paper strips impregnated with concentrations of each basic taste quality (sweet, bitter, sour, and salty) [26,33]. The correlation between olfactory and gustatory functions was also determined for the three groups of subjects. Then, differences in the sensory perception and acceptance of the herbal bitter liqueur Mirtamaro in healthy controls and subjects with hyposmia and hypogeusia were evaluated considering the rate of the odor and taste pleasantness, intensity, and familiarity dimensions using a labeled hedonic Likert-type scale, previously used for the determination of the sensory properties of various food products [21,24,26,34].

2. Materials and Methods

2.1. Participants

A population of 303 subjects was enrolled, with 158 healthy participants (106 women and 52 men), 111 subjects with hyposmia (70 women and 41 men), and 34 subjects with hypogeusia (13 women and 21 men). An explanatory statement was given to all participants. Then, written informed consent was obtained from all subjects involved in the study. Neurodegenerative disorders, psychiatric conditions, acute and chronic rhinosinusitis, as well as stroke and head and neck injury, were considered as exclusion criteria, as previously reported [21,24,26,34]. All participants were not currently taking medications before the test (for 5 days). Data collections were performed from June 2022 to July 2023. Demographic features (age, weight, height, and body mass index) were collected for all participants. The "Azienda Ospedaliera Universitaria di Cagliari" Ethical Committee (Protocol number: NP/2023/963) approved this study, performed according to the Declaration of Helsinki.

2.2. Evaluation of Olfactory and Gustatory Functions

The Sniffin' Sticks test (Burghart Messtechnik, Wedel, Germany), consisting of pen-like odor-dispensing devices for the determination of odor threshold, discrimination, and identification, was used for olfactory function assessment [24,32] (Figure 1a).

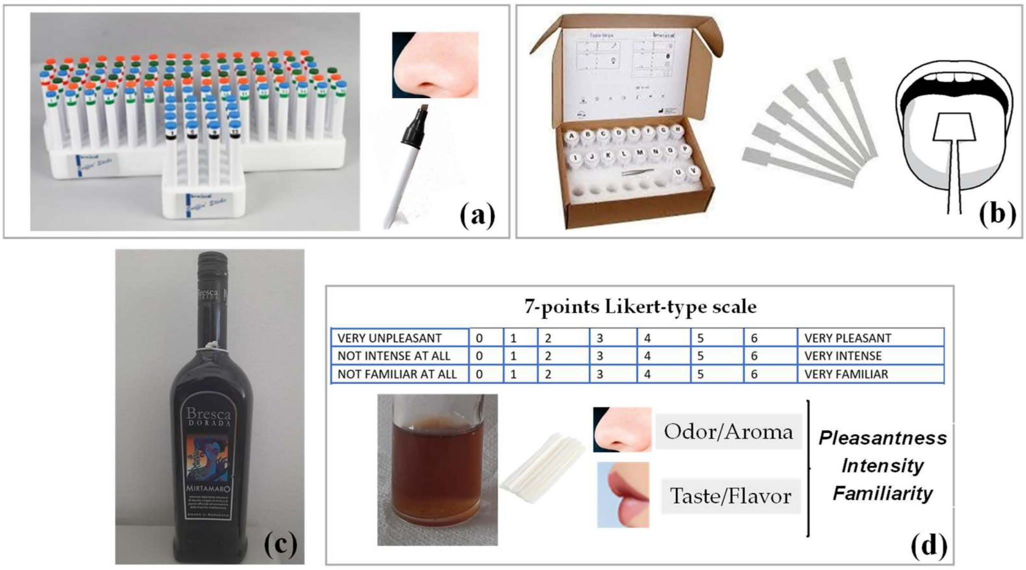


Figure 1. Sniffin' Sticks test for the assessment of olfactory function (**a**), "Taste Strips" test for gustatory function (**b**), the aromatic myrtle herbal-liqueur (Mirtamaro) (**c**), and the hedonic Likert-type scale (7-points, from 0 to 6) to assess pleasantness, intensity, and familiarity dimensions of Mirtamaro odor and taste (**d**).

Sniffin' Sticks test is a validated and commonly used tool for routine clinical assessment of olfactory function and a previous study established its test–retest reliability and validity in subjects with normal sense of smell and in individuals with smell loss [32]. All participants could not smoke or use scented products before the test [24,26]. The odor threshold (OThr) was assessed using 16 stepwise dilutions of n-butanol as previously reported [24,26,35,36]. OThr scores range from 16 (perception of the lowest n-butanol concentration) to 1 (no perception of the highest concentration). Three different pens were used for the odor discrimination (ODi) test, two containing the same odor and the third containing the odor target. The ODi scores ranged from 0 to 16 and were the sum of correct answers [35,36]. The odor identification (OId) test was assessed by 16 common odors [32]. The total olfactory score (TDI) was calculated as the sum of OThr + ODi + OId, and scores as $\leq$16, between 16.25 and 30.5, between 30.75 and 41.25, and >41.5 were indicated functional anosmia, hyposmia, normosmia, and super smellers, respectively [37].

The "Taste Strips" test (Burghart Messtechnik, Wedel, Germany) was used to assess the gustatory function as previously reported [24,26,33]. This test consists of filter paper strips impregnated with 4 concentrations of basic taste qualities: sweet (sucrose: 0.4, 0.2, 0.1, 0.05 g/mL), salty (sodium chloride: 0.25, 0.1, 0.04, 0.016 g/mL), sour (citric acid: 0.3, 0.165, 0.09, 0.05 g/mL), and bitter (quinine hydrochloride: 0.006, 0.0024, 0.0009, 0.0004 g/mL) [24,26,33] (Figure 1b). Drinking water was used to prepare all taste stimuli and for participant mouth-washing before the experiment. The total taste score (TT) was calculated as the sum of correct answers for each taste modality (values ranged from 0 to 16, and scores $\geq$ 9 were considered normogeusia [33]). The "Taste Strips" test is a validated method to evaluate gustatory sensitivity, amply used for clinical assessment of gustatory function not only in healthy subjects [33], but also in patients with chemosensory deficits [24,38].

2.3. Production of the Aromatic Bitter Liqueur Mirtamaro

The commercial myrtle bitter liqueur (Mirtamaro) (Figure 1c) was produced and kindly supplied by the "Bresca Dorada s.r.l.", a company located in Muravera (CA, Sardinia, Italy). Myrtle (*M. communis*) leaves and berries and a complex mixture of aromatic Mediterranean plants and herbs (more than twenty, including fennel, *Citrus* fruits, gentian, licorice, and helichrysum) were subjected to an extensive infusion in a hydroethanolic base according to a secret recipe (the formula was not publicized by the producer) as previously reported [26]. The flavoring and bittering herbs and plants conferred the flavor balance (bitter, balsamic, spicy, and citrus) to Mirtamaro. Both wild and cultivated herbs and plants, fresh and after drying, were used for bitter liqueur preparation, characterized by a 30% (v/v) final alcohol concentration. As reported on the label, sugar was also added as an ingredient to balance the taste and confer palatability [26].

2.4. Determination of Odor and Taste Dimensions (Pleasantness, Intensity, and Familiarity) of the Aromatic Bitter Liqueur Mirtamaro

A subpopulation (111 subjects) was used to assess the sensory properties of Mirtamaro. Participants were classified into three groups: healthy controls (n = 68, 46 women and 22 men), participants with hyposmia (n = 22, 13 women and 9 men), and those with hypogeusia (n = 21, 7 women and 14 men). A hedonic (self-reported) labeled 7-points Likert-type scale was used for the determination of Mirtamaro odor and taste dimensions (pleasantness, intensity, and familiarity) with values ranging from 0—not at all to 6 (0 = very unpleasant and 6 = very pleasant; 0 = not intense at all and 6 = very intense; 0 = not familiar at all and 6 = very familiar), and a value of 3 was considered a neutral point (Figure 1d) [21,24,26,34]. The labeled hedonic Likert Scale is a simple and effective measuring device amply used in food science to measure hedonic differences among foods, beverages, and consumer products and predict their acceptance, suitable for use without extensive training [39].

Before sensory assessment, myrtle bitter liqueur was aliquoted in 2 mL disposable plastic test tubes at room temperature (23 °C) [26]. Initially, after smelling, participants were asked to individuate the most intense subjective aroma descriptors/attributes (presence of flavor notes or aftertastes) of Mirtamaro. Before the taste experiment, participants rinsed their mouths with drinking water. Then, filter paper strips were immersed in a Mirtamaro aliquot and impregnated with the bitter herbal liqueur. After the alcohol removal by strip shaking, participants assessed the Mirtamaro taste (flavor) pleasantness, intensity, and familiarity.

2.5. Statistical Analyses

Initially, the Shapiro–Wilk test was used to calculate the normal data distribution using the software package SPSS software version 25 for Windows (IBM, Armonk, NY, USA). Statistically significant differences (significance level at $p < 0.05$) between data groups (healthy controls and participants with hyposmia and hypogenusia) were performed by the Two-way ANOVA adjusted with the Bonferroni post hoc Multiple Comparisons Test using the SPSS software. The bivariate correlations between different parameters in the three groups were calculated using Pearson's coefficient (r).

3. Results

3.1. Assessment of Olfactory and Gustatory Function

At first, we investigated the differences in olfactory and gustatory function in subjects with hyposmia and hypogeusia compared to healthy controls.

Table 1 shows mean values ± standard deviation (SD) measured for age, sex, weight, height, body mass index (BMI), OThr, ODi, OId, TDI score, sweet, salty, sour, bitter taste perception, and TT score in healthy controls (n = 158), participants with hyposmia (Hypos, n = 111) and with hypogeusia (Hypogeu, n = 34).

Table 1. Demographic and clinical parameters of healthy participants (Controls), participants with hyposmia (Hypos), and with hypogeusia (Hypogeu).

Parameters	**Controls (n = 158)**	**Hypos (n = 111)**	**Hypogeu (n = 34)**
Age (years)	33.0 ± 13.8	36.8 ± 16.5	37.3 ± 16.8
Sex	106 Women/52 Men	70 Women/41 Men	13 Women/21 Men
Weight (kg)	63.8 ± 14.0	66.6 ± 15.7	71.1 ± 11.9 *
Height (m)	1.6 ± 0.1	1.7 ± 0.1	1.7 ± 0.1
BMI	23.5 ± 4.4	24.8 ± 7.9	25.8 ± 3.9
OThr	9.6 ± 3.9	4.1 ± 2.6 ***	6.1 ± 3.7 ***§
ODi	12.8 ± 1.5	10.5 ± 2.4 ***	11.3 ± 3.1 **
OId	13.5 ± 1.2	11.9 ± 2.0 ***	11.9 ± 2.8 ***
TDI score	35.9 ± 3.9	26.5 ± 4.3 ***	29.3 ± 7.9 ***§§
Sweet	3.5 ± 0.7	3.2 ± 1.1 *	2.2 ± 1.2 ***§§§
Salty	3.5 ± 0.7	3.3 ± 0.9 *	2.3 ± 1.2 ***§§§
Sour	2.8 ± 0.8	2.3 ± 1.1 ***	1.3 ± 1.1 ***§§§
Bitter	3.1 ± 1.0	2.9 ± 1.1	1.6 ± 1.2 ***§§§
TT score	12.9 ± 1.6	11.7 ± 2.8 ***	7.4 ± 1.7 ***§§§

Legend: BMI = body mass index; OThr = odor threshold; ODi = odor discrimination; OId = odor identification; TDI score = OThr + ODi + OId; TT score = total taste score. Significant differences between groups (Two-way ANOVA adjusted with the Bonferroni Multiple Comparisons Test): *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$ versus Controls; §§§ $p < 0.001$, §§ $p < 0.01$, § $p < 0.05$ versus participants with hyposmia.

Table 1 showed no significant differences between groups for the mean age, height, and BMI. Instead, a significant increase in weight was observed between subjects with hypogeusia compared to healthy controls [$F_{(2,300)} = 3.914$, $p < 0.05$].

A marked decline in the olfactory function was observed both in subjects with hyposmia and hypogeusia. A noticeable compromise in the gustatory function was detected in subjects with hypogeusia and, to a minor extent, also in subjects with hyposmia.

Regarding olfactory function, a significant reduction in OThr mean scores was observed for subjects with hyposmia and hypogeusia compared to healthy controls [$F_{(2,300)} = 82.671$, $p < 0.001$]. Subjects with hyposmia also showed lower significant mean scores for OThr compared to subjects with hypogeusia ($p < 0.05$) (Table 1). In the ODi mean values, significant differences were found between subjects with hyposmia ($p < 0.001$) and hypogeusia ($p < 0.01$) compared to controls [$F_{(2,300)} = 38.587$, $p < 0.001$]. A significant decrease in OId mean scores, compared to healthy controls, was observed in subjects with hyposmia and hypogeusia [$F_{(2,300)} = 32.436$, $p < 0.001$]. Finally, the total olfactory function (TDI score), significantly decreased in participants with hyposmia and hypogeusia, compared to healthy controls [$F_{(2,300)} = 136.164$, $p < 0.001$].

Sweet taste perception significantly decreased in participants with hyposmia ($p < 0.05$) and hypogeusia ($p < 0.001$) compared to controls [$F_{(2,300)} = 28.627$, $p < 0.001$]. Similarly, significant differences in salty taste perception were found in participants with hyposmia and hypogeusia versus healthy controls [$F_{(2,300)} = 32.067$, $p < 0.001$]. Subjects with hyposmia ($p < 0.05$) and hypogeusia ($p < 0.001$) exhibited a significant decrease in salty taste perception compared to healthy controls. Both participants with hyposmia and hypogeusia showed a significant marked decrease ($p < 0.001$) in sour taste perception versus controls, while subjects with hypogeusia exhibited a significant impairment versus those with hyposmia ($p < 0.001$) [$F_{(2,300)} = 35.011$, $p < 0.001$]. Significant differences were found in the perception of bitter taste between subjects with hypogeusia and healthy controls [$F_{(2,300)} = 25.725$, $p < 0.001$], while no significant differences were observed between participants with hyposmia and controls.

Moreover, subjects with hypogeusia exhibited lower significant ($p < 0.001$) mean scores for sweet, salty, sour, bitter, and total taste perception compared to subjects with hyposmia (Table 1).

Then, the relations between olfactory and gustatory parameters were determined for the three groups. Figure 2a shows the heatmap of Pearson's correlations (r) and significance (p) calculated between ODi, OId, TDI score, sweet, salty, sour, bitter, and TT score in healthy participants (Controls) and participants with hyposmia (Hypos) and hypogeusia (Hypogeu).

In controls, a significant correlation was found between OThr versus OId ($p < 0.05$) and salty taste perception ($p < 0.01$). Moreover, salty taste perception was significantly correlated with the TDI score ($p < 0.01$). Interestingly, no other correlations were observed between olfactory and gustatory function in healthy controls.

Our results showed a greater number of correlations between olfactory and gustatory parameters in participants with hyposmia and hypogeusia than in controls.

Various significant correlations were observed between olfactory and gustatory function in subjects with hyposmia (Hypos), in particular between OThr versus sour taste ($p < 0.05$), ODi versus salty taste perception ($p < 0.01$), and OId versus sweet ($p < 0.01$), salty ($p < 0.05$), sour ($p < 0.01$) taste perception, and TT score ($p < 0.01$). Consequently, the TDI score was significantly correlated with salty ($p < 0.01$) and sour ($p < 0.01$). It was interesting to note that in subjects with hyposmia bitter taste perception was correlated with sweet ($p < 0.01$) and salty ($p < 0.01$) taste. Significant correlations were also observed between TDI score versus salty ($p < 0.01$) and sour ($p < 0.05$) taste perception.

Finally, significant correlations were found between olfactory and gustatory parameters also in participants with hypogeusia (Hypogeu), in particular between OThr versus ODi ($p < 0.05$), OId ($p < 0.01$), and TT score ($p < 0.01$). In addition, important significant correlations were also observed between ODi versus OId ($p < 0.01$), salty ($p < 0.01$), and bitter ($p < 0.05$) taste perception. Moreover, a significant correlation was also observed between TDI score versus salty taste ($p < 0.05$).

Interestingly, significant correlations between TDI and TT scores were observed in participants with hyposmia ($p < 0.01$) (Figure 2c) and hypogeusia ($p < 0.05$) (Figure 2d), while no correlation was observed in healthy controls (Figure 2b), indicating that the olfactory dysfunction may influence gustatory function and vice versa.

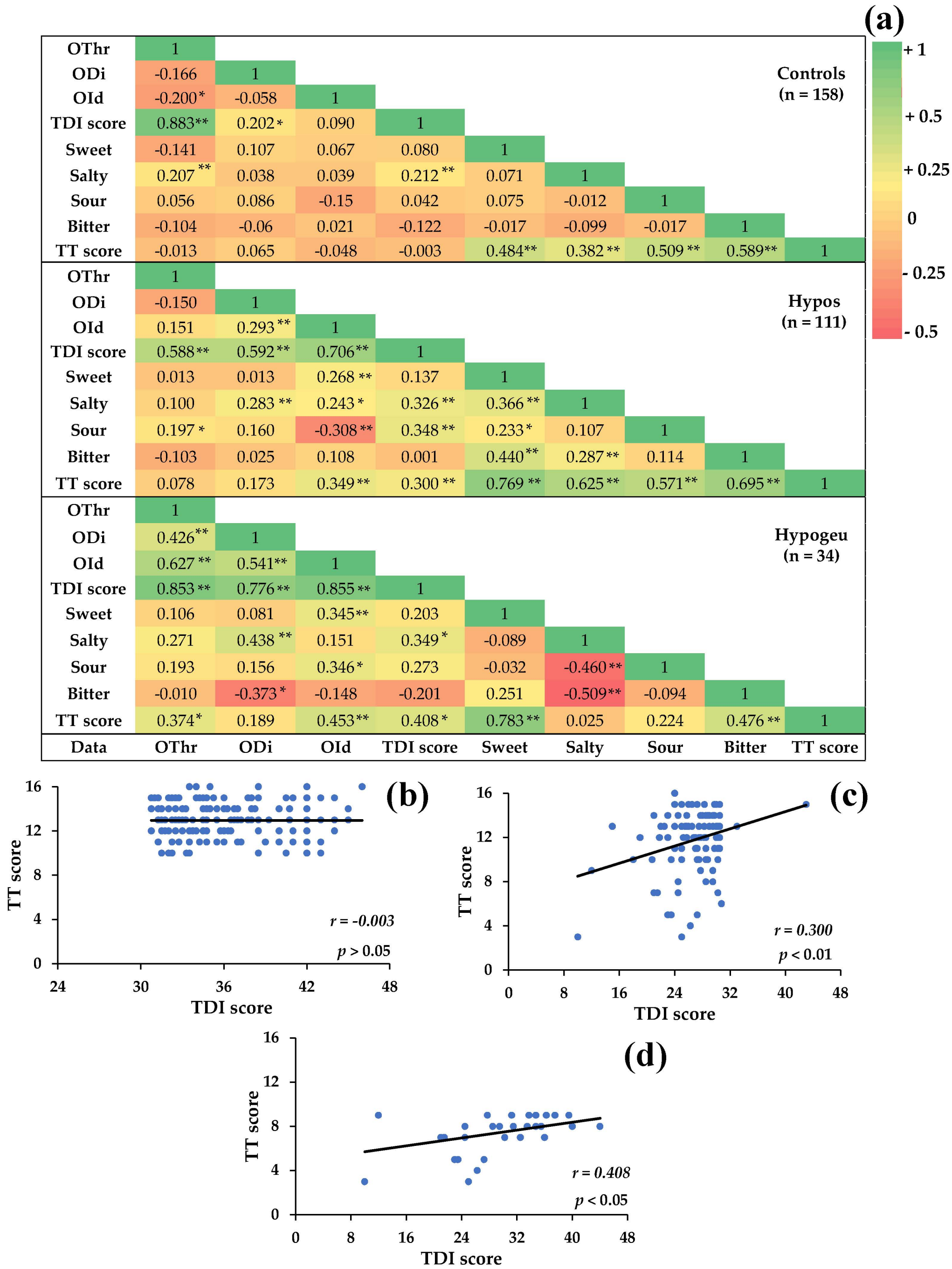


Figure 2. Heatmap of Pearson's correlations (r) and significance (** $p < 0.01$; * $p < 0.05$) calculated between olfactory parameters (odor threshold, OThr; odor discrimination, ODi; odor identification, OId; TDI score = OThr + ODi + OId) and gustatory parameters (sweet, salty, sour, bitter, and TT score = total taste score) in healthy participants (Controls, n = 158) and participants with hyposmia (Hypos, n = 111) and hypogeusia (Hypogeu, n = 34) (**a**). Scatterplots of the relationship between TDI score versus TT score in controls (**b**), participants with hyposmia (**c**), and with hypogeusia (**d**).

3.2. Determination of Odor and Taste Dimensions (Pleasantness, Intensity, and Familiarity) of the Aromatic Bitter Liqueur Mirtamaro

Then, the effect of aromatic herbs/plants on the sensory perception of a food product in consumers with chemosensory deficits was evaluated.

A subpopulation (n = 111) was chosen among all participants to assess the differences in the perception of sensory characteristics/acceptance of the herbal myrtle bitter liqueur Mirtamaro in subjects with hyposmia and hypogeusia versus healthy controls.

Values of demographic and clinical parameters of healthy controls (Controls, n = 68), participants with hyposmia (Hypos, n = 22), and with hypogeusia (Hypogeu, n = 21) enrolled to assess Mirtamaro odor and taste dimensions are reported in Table 2.

Table 2. Demographic and clinical parameters of healthy participants (Controls), participants with hyposmia (Hypos), and with hypogeusia (Hypogeu) enrolled to assess Mirtamaro odor and taste.

Parameters	Controls (n = 68)	Hypos (n = 22)	Hypogeu (n = 21)
Age (years)	32.4 ± 13.4	42.5 ± 22.1 *	37.1 ± 18.8
Sex	46 Women/22 Men	13 Women/9 Men	7 Women/14 Men
Weight (kg)	65.6 ± 13.9	68.7 ± 17.4	71.8 ± 12.5
Height (m)	1.7 ± 0.1	1.7 ± 0.1	1.7 ± 0.1
BMI	24.0 ± 4.7	25.2 ± 6.2	25.9 ± 4.4

Legend: BMI = body mass index. Significant differences between groups (Two-way ANOVA adjusted with the Bonferroni Multiple Comparisons Test): * $p < 0.05$ versus Controls.

In this subpopulation, no significant differences were found in weight, height, and BMI between subjects with hyposmia and hypogeusia compared to controls, while participants with hyposmia exhibited a significant increase ($p < 0.05$) in the age versus healthy controls.

Also in this subpopulation, participants with hyposmia showed a marked decrease in the olfactory function, exhibiting mean scores significantly lower than controls for OThr [$F_{(2,108)} = 21.654$, $p < 0.001$], ODi [$F_{(2,108)} = 12.227$, $p < 0.001$], and OId [$F_{(2,108)} = 20.282$, $p < 0.001$] (Figure 3a).

Regarding the gustatory function (Figure 3b), a significant decrease was observed for salty ($p < 0.05$), and sour ($p < 0.05$) score perception in participants with hyposmia versus healthy controls.

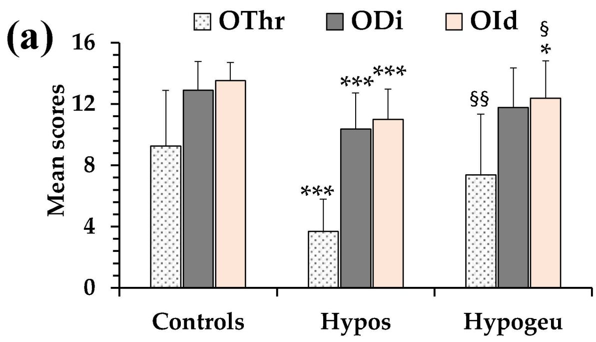
Participants with hypogeusia were characterized by a marked deficit in the gustatory function, exhibiting a significant decrease in sweet [$F_{(2,108)} = 15.557$, $p < 0.001$], salty [$F_{(2,108)} = 17.169$, $p < 0.001$], sour [$F_{(2,108)} = 19.551$, $p < 0.001$], and bitter [$F_{(2,108)} = 12.514$, $p < 0.001$] taste perception compared to healthy controls (Figure 3b), while a significantly lower score than healthy controls was observed for OId ($p < 0.05$) (Figure 3a). Moreover, significant differences were observed in sweet ($p < 0.05$), salty ($p < 0.05$), sour ($p < 0.01$), and bitter taste perception ($p < 0.05$) between participants with hypogeusia versus those with hyposmia (Figure 3b).

Significant differences were observed for the TDI [$F_{(2,108)} = 39.354$, $p < 0.001$] and TT scores [$F_{(2,108)} = 60.429$, $p < 0.001$] between subjects with hyposmia and hypogeusia compared to healthy controls, confirming also in this subpopulation the marked deficits in the olfactory and gustatory function in these two groups of participants and the relation between the two functions.

Then, a 7-points hedonic Likert-type scale was used to evaluate the herbal bitter liqueur odor and taste dimensions (pleasantness, intensity, and familiarity) in the three groups of participants to evidence the potential role of aromatic herbs/spices in modulating sensory perception in consumers with chemosensory deficits [24,26,34].

Initially, participants provided a subjective description of the Mirtamaro odor sensory properties (aroma), and the results obtained for healthy controls and participants with hyposmia and hypogeusia are listed in Table 3.

Regarding Mirtoamaro odor (aroma), all groups of participants individuated the presence of alcohol, myrtle as the main contributor to the sensory properties of the product, and bitter compounds. Due to the complex composition of the herbal bitter liqueur, the individuation of specific components (herbs/spices) of the flavoring mixture was very difficult for all participants. In general, healthy controls furnished more sensory descriptors than subjects with hyposmia and hypogeusia such as the presence of specific aromas (licorice, juniper, chinotto, orange, woody, spicy, mint, etc.). However, all groups individuated the note of aromatic herbs.



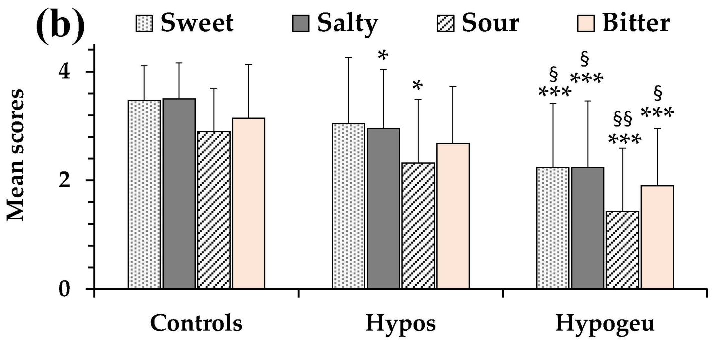


Figure 3. Mean values ± standard deviation (SD) of odor threshold (OThr), odor discrimination (ODi, odor identification (OId) (**a**), sweet, salty, sour, and bitter taste perception (**b**) measured in healthy participants (Controls, n = 68) and participants with hyposmia (Hypos, n = 22) and hypogeusia (Hypogeu, n = 21). Significant differences between groups (Two-way ANOVA adjusted with the Bonferroni Multiple Comparisons Test): *** $p < 0.001$, * $p < 0.05$ versus Controls; §§ $p < 0.01$, § $p < 0.05$ versus participants with hyposmia.

Table 3. Odor (aroma) and taste (flavor) perceived attributes of the myrtle herbal liqueur (Mirtamaro) measured in healthy participants (Controls, n = 68), participants with hyposmia (Hypos, n = 22) and hypogeusia (Hypogeu, n = 21).

Subjects	Sensory Input	Sensory Perceived Attributes
Ctrl	Odor	Liqueur; bitter liqueur; myrtle; herbs; alcohol; bitter; very bitter; chinotto; licorice; juniper; berries; orange; woody; spicy; nicotine; coffee; Sambuca; medication; pungent; natural essences; rum; coffee; balsamic herbs; orange; mint.
	Taste	Bitter liqueur; myrtle berries; myrtle; bitter; very bitter; spicy; alcohol; strong; bitter myrtle; wine; herbs note; *Citrus* note; sour; initially sweet, then very bitter; strong; chinotto aftertaste; slightly bitter; honeyed aftertaste; chinotto aftertaste; medication; mint aftertaste; quinine; Sambuca.
Hypos	Odor	Bitter liqueur; aromatic bitter liqueur; licorice; Sambuca; alcohol; Marsala; aromatic herbs; medication; spicy.
	Taste	Very bitter; bitter; very bitter with a sour note; bitter liqueur; whisky; propolis; rum; sweet; sweet with caramelized aftertaste; initially sweet, then bitter.
Hypogeu	Odor	Liqueur; myrtle; alcohol; bitter; bitter liqueur; spirit; aromatic herbs; natural essences; Marsala.
	Taste	Sweet; sweet with caramelized aftertaste; bitter; myrtle; alcohol; spirit; initially sweet, then bitter; liqueur.

Concerning the taste subjective perceived attributes (flavor) of Mirtoamaro (Table 3), most participants belonging to all groups individuated bitterness as the main taste perception (the attributes were bitter or very bitter), indicating an initial sweet taste perception followed by a bitter taste. Moreover, all groups recognized the occurrence of myrtle extract and alcohol. However, healthy controls furnished more sensory descriptors for Mirtamaro taste than subjects with hyposmia and hypogeusia, indicating the presence of specific aromas (herbs and *Citrus* notes and chinotto, honeyed and mint aftertaste).

Figure 4 shows the ratings of odor pleasantness (P), intensity (I), and familiarity (F) dimensions measured for the odor (Figure 4a) and taste (Figure 4b) of the bitter liqueur measured in healthy participants (Controls), participants with hyposmia (Hypos) and hypogeusia (Hypogeu).

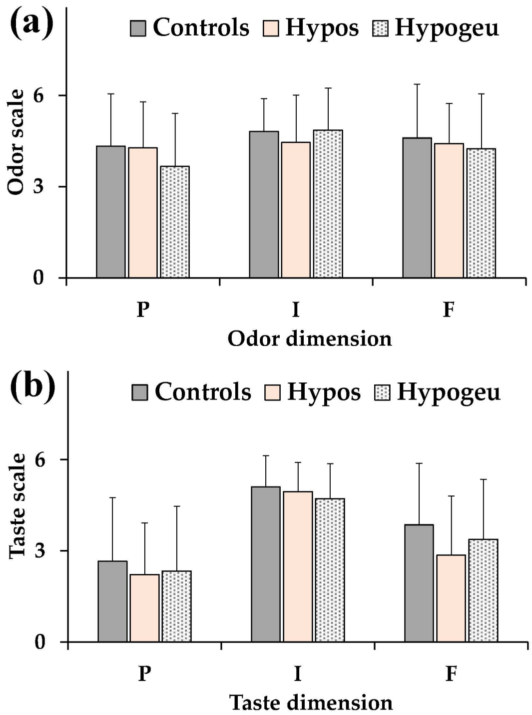


Figure 4. Pleasantness (P), intensity (I), and familiarity (F) dimensions determined for the odor (**a**) and taste (**b**) of the aromatic myrtle herbal liqueur (Mirtamaro) measured in healthy participants (Controls, n = 68), participants with hyposmia (Hypos, n = 22) and hypogeusia (Hypogeu, n = 21). No significant differences were measured by Two-way ANOVA.

All groups of participants showed high mean scores for odor pleasantness, intensity, and familiarity dimensions (Figure 4a). Interestingly, no significant differences were observed in the perception of Mirtamaro odor pleasantness, intensity, and familiarity dimensions between participants with hyposmia and hypogeusia compared to healthy controls.

In general, for all groups of participants lower ratings of Mirtamaro taste pleasantness and familiarity than the same odor dimensions were measured (Figure 4). Moreover, the taste intensity of Mirtamaro (TI) was mainly identified with the intensity of its bitterness.

Furthermore, no significant differences were observed in the perception of bitter liqueur taste pleasantness, intensity, and familiarity between participants with hyposmia and hypogeusia compared to healthy controls.

Obtained results indicated that participants with hyposmia and hypogeusia perceived Mirtoamaro odor and taste dimensions similarly to healthy controls, despite marked differences observed in olfactory and gustatory functions among the three groups.

Finally, correlations between OThr, bitter taste intensity, pleasantness, intensity, and familiarity of Mirtamaro odor and taste were calculated in healthy controls and participants with hyposmia and hypogeusia (Figure 5a).

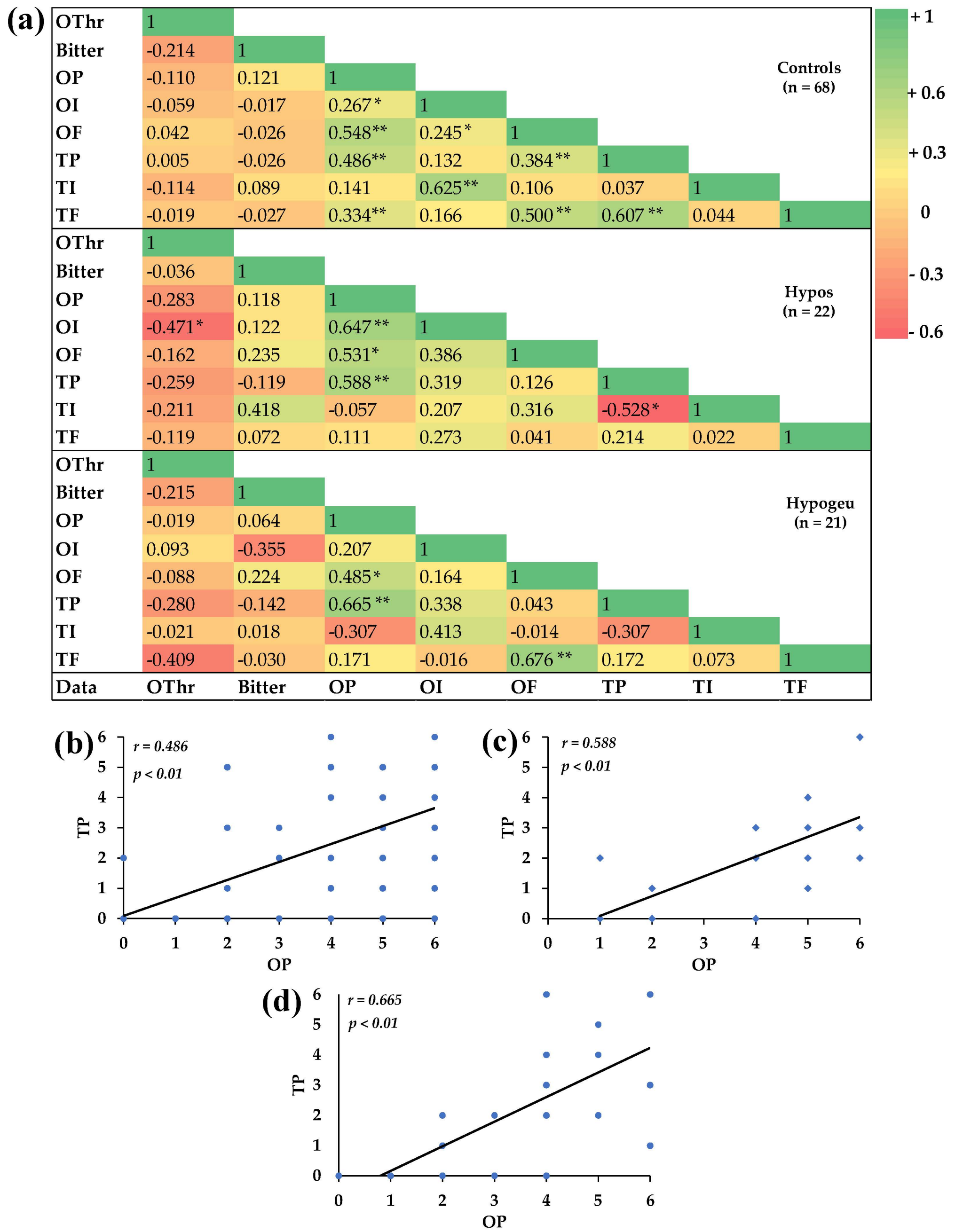


Figure 5. Heatmap of Pearson's correlations (r) and significance (** $p < 0.01$; * $p < 0.05$) calculated between odor threshold (OThr), bitter taste intensity, Mirtamaro odor pleasantness (OP), odor intensity (OI), odor familiarity (OF), taste pleasantness (TP), taste intensity (TI), and taste familiarity (TF) in healthy participants (Controls, n = 68) and participants with hyposmia (Hypos, n = 22) and with hypogeusia (Hypogeu, n = 21) (**a**). Scatterplots of the relationship between OP score versus TP score in Controls (**b**), participants with hyposmia (**c**), and with hypogeusia (**d**).

Strong correlations were determined between different Mirtoamaro odor (OP, OI, OF) and taste dimensions (TP, TI, TF) in all groups of participants.

Healthy controls exhibited more correlations between several Mirtoamaro odor and taste dimensions than participants with hyposmia and hypogeusia. Significant correlations were evidenced for OP/OI, OP/OF, OI/OF, OP/TP, OP/TF, OI/TI, OF/TP, OF/TF, and TP/TF in healthy controls, while no significant correlations were observed between OThr and bitter taste perception versus different Mirtoamaro odor and taste dimensions.

Participants with hyposmia showed a significant correlation between OThr and Mirtamaro odor intensity (OI) (r = −0.4717, $p < 0.05$). In addition, in this group, significant correlations were determined for OP/OI, OP/OF, OP/TP, and TP/TI (Figure 5a), whereas significant correlations between OP/OF, OP/TP, and OF/TF were observed for participants with hypogeusia.

Interestingly, no significant correlations were observed between bitter taste intensity (bitter) and Mirtamaro taste intensity (TI) in all groups, demonstrating that the perception of Mirtamaro flavor was not only due to the bitter perception but to the combination of the different aromatic notes/aftertastes of herbs and plants present in the mixture.

Moreover, a strong correlation was found between odor pleasantness/odor familiarity in all groups, indicating that familiarity with the liqueur aroma influenced its likability.

Scatterplots of the relationship between odor pleasantness score versus taste pleasantness score in controls, participants with hyposmia, and with hypogeusia are reported in Figure 5b–d, respectively. A strong correlation OP/TP was found between healthy controls (r = 0.486, $p < 0.01$), participants with hyposmia (r = 0.588, $p < 0.01$), and with hypogeusia (r = 0.665, $p < 0.01$) (Figure 5), highlighting the important role of aromatic plants/herbs in modulating the Mirtamaro acceptability independently of the individual chemosensory impairment.

4. Discussion

Sensory properties of foods are principally the outcome of the interaction of food characteristics/quality (such as physical structure and chemical/nutritional properties) and the consumer's characteristics (gender, age, attitude, and physiological/psychological state) and its environment (family, cultural traditions, and education) [1–5,40]. Among sensory and personal factors, health conditions significantly influence consumer choice [3,40]. It has been reported that the decline in olfactory and gustatory function greatly affects food sensory perception and eating behavior [6–11,13–16]. Olfactory and gustatory dysfunctions limit the individual's liking for food and beverages, leading to poor appetite, possible malnutrition, and worsening of other diseases [16]. Several studies evaluated the effect of olfactory/gustatory disorders on food-related experiences using a self-rated questionnaire [14,31], whereas sensory testing using real food items under controlled test conditions in patients with sensory impairment has rarely been used [11,24,31].

In this study, we assessed for the first time the perception of odor/taste sensory characteristics and acceptance of the bitter herbal myrtle liqueur Mirtamaro in subjects with hyposmia and with hypogeusia compared to healthy controls. Myrtle leaves, berries, seeds, and essential oils are amply used for the food aromatization and cosmetic/pharmaceutical applications due to their anti-inflammatory and antioxidant activities [26,28,30,41]. Taking into consideration the growing popularity of the bitter taste among consumers [25–29], Mirtoamaro was prepared by the maceration of myrtle leaves and berries together to a complex secret blend of aromatic and bitter Mediterranean plants/herbs [26,28]. Aromatic plants and spices are traditionally used in the Mediterranean area to prepare beverages and the most used plants for conferring bitterness to aromatic bitter liqueurs belong to *Gentiana*, *Artemisia*, and *Achillea* species [28]. The herbal liqueur Mirtamaro is characterized by a strong bitter taste and balsamic, spicy, and citrus flavor [26].

Initially, the occurrence of significant changes in olfactory and gustatory perception was explored in subjects with hyposmia and hypogeusia compared to healthy controls. Our results showed a marked decline in the olfactory function (OThr, ODi, and OId) in subjects with hyposmia. Moreover, these subjects perceived sweet, salty, sour, and total taste as less intense than healthy participants. Participants with hypogeusia showed a marked reduction in sweet, salty, sour, and bitter perception versus healthy participants, coupled with an impairment of the olfactory function.

Interestingly, the presence of significant positive correlations between total olfactory and total taste scores (TDI/TT) in participants with hyposmia and hypogeusia indicated a marked interconnection between olfactory and gustatory function in consumers with chemosensory deficits. The olfactory function is well-known to be closely associated with the gustatory function [11]. Both gustatory and olfactory systems send their inputs to associative brain areas which play an important role in food ingestion or rejection and food quality determination [42]. Food perception is a multisensory event determined by the integration of gustatory function, smell, physical sensations, such as food temperature and consistency, and trigeminal perceptions [3–5,42]. Previous studies evidenced a high rate of comorbidity between gustatory and olfactory impairment [7,11].

Then, differences in the perception of sensory characteristics (odor and taste) and acceptance of the bitter herbal myrtle liqueur Mirtamaro were assessed in subjects with hyposmia and hypogeusia compared to healthy controls. Differences in the grade of intensity, familiarity, and pleasantness of Mirtamaro odor (aroma) and taste (flavor) were measured in the three groups using a 7-points labeled hedonic Likert-type scale, a method amply used for the evaluation of food and beverage acceptability in the food industry [39]. We previously applied this method for the determination of the sensory properties of salted and semi-dried mullet ovary products in healthy controls [34] and to assess the salty perception of saline solutions prepared with sea salts flavored with Mediterranean aromatic plants in healthy participants and subjects with hyposmia [21,24].

Considering the Mirtamaro aroma and flavor subjective sensory evaluation, all groups of participants individuated the occurrence of bitter compounds, myrtle, aromatic herbs, and alcohol, However, healthy controls furnished more sensory descriptors than subjects with chemosensory deficits. All groups indicated the odor of the aromatic bitter liqueur as very familiar and pleasant, while the Mirtamaro taste was described as less pleasant and familiar than the odor. Moreover, bitterness represented for all groups the main Mirtamaro taste modality. As previously observed for other bitter alcoholic beverages, all groups indicated an initial sweet taste perception followed by a strong bitter taste [25]. Interestingly, despite the existence of marked alterations in the olfactory and gustatory functions, no significant differences were observed in the perception of the bitter liqueur odor and taste pleasantness, intensity, and familiarity between participants with hyposmia and hypogeusia compared to healthy controls.

Previous study reported that subjects with olfactory dysfunction were significantly less able to recognize several categories of food items during consumption using taste, smell, and mouthfeel combined [14]. Another study evidenced that subjects with olfactory dysfunction rated ortho-nasal odors of several foods (dark chocolate, peanut butter, and caramel) as less pleasant, intense, and familiar in comparison to controls [11]. Most individuals with taste dysfunction report distorted taste perception for common foods such as meats, eggs, coffee, fresh fruits, and carbonated beverages [43].

Consumers with olfactory or gustatory deficits try to compensate for the decrease in food enjoyment using an increased quantity of salt, sweeteners, fats, condiments, and spices [6,10,13,16,17,31,43,44]. Herbs and spices are normally used to increase food flavor and liking/preference for foods characterized by low palatability [20,45]. The use of flavor enhancers (monosodium glutamate) and herbs/spices has been indicated as a strategy to ameliorate the perception of aroma and taste and acceptance of beverage/food in consumers with sensory-compromised situations [16,17,31,46]. The use of spices and herbs has been recently proposed as a strategy for the improvement of taste (ageusia) and smell (anosmia) loss in coronavirus-induced disease (COVID-19) patients [17]. Moreover, we previously demonstrated that the addition of Mediterranean plants and herbs (rosemary, fennel, myrtle, orange, and saffron) to sea salt enhances the salty taste perception in patients with olfactory dysfunction [24]. Herbs and spices furnish numerous molecules with the ability to interact with receptors, compensating for chemosensory impairment [46].

Our results confirmed the use of blends of aromatic herbs/plants as a strategy to enhance sensory perception in subjects with chemosensory deficits. Regarding Mirtamaro odor (aroma), the similar rating in aromatic liqueur odor intensity perception measured especially in participants with hyposmia compared to healthy controls demonstrated an evident modulatory effect of liqueur aromatic compounds, derived from the blend of aromatic herbs/plants, in potentiating olfactory perception. More than twenty aromatic herbs/plants (among others fennel, gentian, licorice, *Citrus* fruits, and helichrysum) were blended with myrtle leaves/berries according to a secret recipe (the formula was not publicized) to obtain Mirtamaro [26]. The main Mirtamaro aroma components previously identified were 1,8 cineole, methyl chavicol (estragole), octanoic acid, alpha-terpineol, carvone, fenchone, terpinen-4-ol, *orto*-cymene, linalool, limonene, gamma-terpineol, (E)-anethole, and alpha-thujene [26]. Eucalyptol (1,8-cineole), characterized by eucalyptus and camphor-like odor, and methyl chavicol (estragole), characterized by spicy, green, herbal, fennel, and anise odor, were the most abundant volatile Mirtamaro compounds (49% of total volatile compounds) [26]. Both compounds, being characterized by a pleasant odor, possibly contributed to the Mirtamaro odor intensity and pleasantness [26]. Volatile organic aroma compounds are perceived in the nasal cavity through the smell sensory organs (ortho-nasal smell) [4]. The activation of brain areas by olfactory stimuli evokes emotions and numerous associations [34,47]. Mirtamaro odor pleasantness was strictly correlated to odor familiarity in all groups, suggesting that familiarity with Mirtamaro influenced its likability. The habitual consumption of a liqueur or food has been previously demonstrated to rise its acceptability [3,34].

Regarding Mirtamaro taste (flavor), analogous rating in aromatic liqueur taste intensity perception determined in participants with hypogeusia compared to healthy controls also demonstrated the modulatory effect of Mirtamaro flavor compounds derived from aromatic plants/herbs in ameliorating the gustatory perception. The absence of significant correlations between bitter taste intensity (bitter) and Mirtamaro taste intensity (TI) in all groups confirmed that the Mirtamaro flavor perception was not only ascribable to the sole bitter taste perception but to the combination of the different aromatic notes/aftertastes of herbs and plants present in the complex mixture. The perception of food flavor is due to an integration of gustatory and olfactory information, involving the gustative perception of volatile compounds by retro-nasal olfaction (aroma), soluble/non-volatile compounds (basic tastes), and chemical sensations by the trigeminal nerve activation [2–4,24,26,34]. Aroma and taste activate central cognitive pathways to produce the perception of flavor and a complex relationship has been demonstrated between multisensory flavor perception and specific configurations of volatile organic compounds in drink and food products [4,25,48]. The aromatic substances present in alcoholic liqueur may influence bitterness perception [25]. Essential oils and phenolic compounds (phenolic acids, flavonoids, anthocyanins, etc.) are reported as the most important phytochemicals in myrtle berries, leaves, and obtained liqueurs [26,28,41]. Therefore, Mirtamaro flavor perception in participants with hyposmia and hypogeusia, as well as in healthy controls, was the result of a multisensory perception of many chemical components derived from the mixture of aromatic herbs/plants, including bitter compounds, volatiles, and non-volatile polar components. Mirtamaro non-volatile polar components may modulate the taste perception in the tongue, whereas its aromatic compounds, liberated in the mouth, may be responsible for the flavor perceived by retro-nasal olfaction [26].

Finally, the strong correlation between Mirtamaro odor and taste pleasantness (OP/TP) in participants with hyposmia and with hypogeusia as well as in healthy controls confirmed the association between gustatory and olfactory perception and the important role of aromatic compounds from plants/herbs in the positive modulation of the Mirtamaro acceptability [21] independently of the individual chemosensory impairment.

The results of the present study contribute to the existing literature on chemosensory deficits and provide useful information on the role of aromatic herbs and spices in positively modulating the sensory perception and acceptance of foods/beverages in consumers with chemosensory deficits. The limitation of this study is the low number of consumers with hyposmia and hypogeusia in the subpopulation selected for the sensory evaluation of Mirtoamaro. Further studies are needed to explore the modulatory effect of mixtures of aromatic herbs/spices on the sensory properties of specific foods/beverages in a large number of subjects with olfactory and gustatory deficits to individuate the best blends that confer an aroma and taste appealing to this category of consumers.

5. Conclusions

In this study, we evaluated the perception of sensory characteristics (odor and taste) and acceptance of a commercial bitter herbal liqueur Mirtamaro in subjects with hyposmia and hypogeusia compared to healthy controls. Participants with hyposmia and hypogeusia showed ratings in odor and taste familiarity, pleasantness, and intensity of the myrtle bitter aromatic liqueur Mirtamaro like healthy participants. Moreover, all groups of subjects furnished similar sensory descriptors for Mirtoamaro odor and taste.

There is a great interest in individuating approaches to enhance the perception of food/beverage aroma and taste in subjects with sensory-compromised situations. Our results provide new evidence about the important role of blends of aromatic spices and herbs in ameliorating food sensory perception in consumers characterized by chemosensory deficits. The results of the current studies hold some practical implications for product developers in an industrial setting. The addition of selected blends of aromatic herbs and spices to different foods/beverages represents a healthy approach for potentiating the flavor perceptive capacity and food enjoyment in consumers with olfactory and gustatory deficits and the design of special foods that meet the needs of this category of consumers may represent new business opportunities for food companies.

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Informed Consent Statement: Written informed consent was obtained from all subjects involved in the study.

Data Availability Statement: The datasets generated and analyzed during the current study are available from the corresponding author upon reasonable request. The data are not publicly available due to privacy.

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Abbreviations

Othr: odor threshold; ODi: odor discrimination; OId: odor identification; TDI: total olfactory score; TT: total taste score; OP: odor pleasantness; OI: odor intensity; OF: odor familiarity; TP: taste pleasantness; TI: taste intensity; TF: taste familiarity.

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