

Antioxidant potential, lipid profile and sensory attributes of a functional yogurt formulated with hydroethanolic extract of *Mentha Piperita* L.

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Abstract

This study aims to quantify and identify phenolic compounds in the hydroethanolic extract of *Mentha piperita* L. harvested in the Algerian Sahara by colorimetric and LC-MS/MS analysis, assess its antioxidant capacity by DPPH and ABTS assays and examine the impact of its inclusion at 2 to 6% rates on sensory and oxidative properties (DPPH, ABTS, TEARS) and the lipid profile analyzed by GC-FID in steamed yoghurt.

The extract contained 41.29 mg GAE/ml total phenolics, 22.45 mg QE/ml total flavonoids and only 0.59 mg CE/ml condensed tannins. LCMS/MS analysis detected 26 phenolic compounds, among which rosmarinic acid was the most abundant. DPPH and ABTS assays showed IC₅₀ values of 3.18 and 2.49 mg freeze-dried extract/ml, indicating the high antioxidant potential of the examined extract. Yogurts containing 2% and 4% of this extract were well accepted by the sensory evaluation panel.

The studied extract also significantly increased the antioxidant capacity of yogurt up to 39.51% compared to natural yogurt after 20 days of refrigeration, significantly reduced lipid peroxidation and at a dose of 4%, caused a significant improvement ($p < 0.01$) in the omega content of yogurt, especially alpha-linolenic acid, compared to the control and caused a marked decrease ($p < 0.01$) in the ω -6/ ω -3 ratio in yogurt from 5.21 to 4.11 on day 20 of refrigeration compared with the control. The production of a sensorially acceptable pasteurized yogurt enriched with up to 4% hydroethanol extract of *Mentha piperita* L. as an alternative to synthetic antioxidant additives with health-promoting properties appears feasible.

1. Introduction

Yogurt is one of the most popular fermented dairy products, made by fermentative acidification of milk by specific lactic acid bacteria, including *Streptococcus thermophilus* and *Lactobacillus bulgaricus* (Gahruie et al., 2015). Due to its sensory and nutritional characteristics, it is widely appreciated and consumed worldwide (Jaster et al., 2018). It is an excellent source of protein, calcium, phosphorus, magnesium, zinc, vitamins, conjugated linoleic acid, and other essential fatty acids (Gahruie et al., 2015). However, the nutritional content of this dairy product in bioactive compounds is limited, but it can serve as a good base for enriching its composition with plant-derived additive substances such as phenolic compounds and producing new functional foods for widespread consumption (Alenisan et al., 2017).

In the diversity of plants, *Mentha piperita* L, commonly known as "peppermint", belongs to the family of Lamiaceae. Its leaves are commonly used in the preparation of hot drinks such as traditional Algerian mint tea and herbal teas, as well as in cooked dishes to add flavor and aroma. They are also used to treat several diseases, including colds, musculoskeletal pain, gastrointestinal tract disorders, and inflammation of the mouth and pharynx (Trevisan et al., 2017).

The secret is that peppermint contains numerous phytochemical compounds, including polyphenols, which act very effectively as antioxidants while being less toxic than synthetic molecules. These characteristics make it a major asset for the food industry, as phenolic compounds slow down the oxidative degradation of

ingredients, especially lipids, thus ensuring both good product quality and better nutritional value (Mallick et al., 2016).

This study aims to enhance the functionality of a set style yogurts by supplementing them with hydroethanolic extract of *Mentha piperita* L rich in phenolic compounds at different doses and to monitor the evolution of their organoleptic and antioxidant characteristics during cold storage at 4°C while examining its impact on the lipid profile of the products.

2. Material And Methods

2.1. Material

2.1.1. Plant material

The aerial part, leaves and stems, of *Mentha piperita* L. were collected in September 2018 in Ouargla, southeast Algeria. They were air-dried for 15 days at room temperature, crushed, and then stored in sterile, sealed jars away from light and moisture.

2.1.2. Bacteria

Two strains of lactic acid bacteria, *Streptococcus thermophilus* (ST) (ATCC 19258) and *Lactobacillus delbrueckii* sub sp *bulgaricus* (LB) (ATCC 11842), provided as a donation by SACCO, Italy, and were used for the production of fermented milk and antibacterial tests.

2.2. Methods

2.2.1. Extraction of phenolic compounds

The extraction of phenolic compounds from *Mentha piperita* L was carried out by maceration for 6 hours in a mixture of ethanol: water (80:20) according to the procedure described by Ait Chabane et al. (2021). The hydroethanolic extract was stored at 4°C in amber glass bottles.

Yield (%) = $(m_0/m_1) \times 100$; Where: m_1 is the mass of the residue obtained after evaporation; m_0 is the initial mass of the plant.

2.2.2. Preparation of Yogurts with Mint Extract

Pasteurized cow's milk provided by the industrial group (GIPLAIT), a subsidiary of Mostaganem, was heated to 45°C and distributed into 12 pots of 100 ml. Each set of 3 pots received the same dose (2%, 4%, or 6%) of HEEMP. Three pots to which no extract was added were taken as control. All milk pots were then inoculated with 3% of a starter culture prepared from two strains of lactic acid bacteria in a ratio of 2:1 (2 St/1 Lb), sealed and incubated for 4 hours at 45°C. After fermentation, the cooled yogurts were stored in a refrigerator set at 4°C until the 21st day.

2.2.3. Measurement of bioactive compounds of peppermint extract

2.2.3.1. Total phenolic compounds

The total content of phenolic compounds was determined by the Folin-Ciocalteu method (Saffidine et al., 2015). The absorbance was measured at a wavelength of 765 nm using a Janway 7205 spectrophotometer. The total phenolic compound contents were expressed in mg of gallic acid equivalents per ml of extract (mg GAE/ml), in mg of gallic acid equivalents per g of lyophilized extract (mg GAE/g E), and in mg of gallic acid equivalents per 100mg of peppermint powder (mg GAE/100g PMP) and were deduced by the linear equation ($y = 0.008x + 0.011$; $R^2 = 0.998$) of a standard curve made with gallic acid under the same conditions as the sample.

2.2.3.2. Total flavonoids

Total flavonoids were measured using aluminium trichloride prepared at 2% in methanol (Djenidi et al., 2020). The absorbance was read with a spectrophotometer (Janway 7205) at a wavelength of 430 nm. The total flavonoid content was determined by a standard curve established under identical conditions using quercetin with a linear equation of the form ($y = 0.041 x + 0.007$; $R^2 = 0.999$). The results are expressed in mg equivalent quercetin per ml of extract (mg EQ/ml E), in mg equivalent quercetin per gram of lyophilized extract (mg EQ/g EL), and in mg equivalent quercetin per 100g of peppermint powder (mg EQ/100g PMP).

2.2.3.3. Condensed Tannins

Tannins were quantified using the vanillin method as reported by Mahmoudi et al. (2013). A UV spectrophotometer (Janway 7205) was used to measure the absorbance at 500 nm. The linear equation ($y = 0.001x + 0.013$; $R^2 = 0.994$) of the standard curve established with catechin was used to deduce the condensed tannin content of the *HEEMP*. The results were expressed in mg equivalent catechin per ml of extract (mg EC/ml E), mg equivalent catechin per g of lyophilized extract (mg EC/g EL), and mg equivalent catechin per 100g of peppermint powder (mg EC/100g PMP).

2.2.3.4. Phenolic compound profile

The chemical profile of phenolic compounds contained in *HEEMP* was screened by LC-MS/MS using the method of Yilmaz (2020) with 56 authentic standards. The LC-MS/MS system used consisted of a Shimadzu-Nexera ultra-high performance liquid chromatograph coupled to a tandem mass spectrometer. Chromatographic separation was carried out on an Agilent Poroshell 120 EC-C18 analytical column (150mm × 2.1 mm, 2.7 µm) at a temperature set to 40°C. Two eluents were used, namely eluent A (water + 5 mM ammonium formate + 0.1% formic acid) and eluent B (methanol + 5 mM ammonium formate + 0.1% formic acid). The following elution gradient was used: 20–100% B (0–25 min), 100% B (25–35 min), 20% B (35–45 min). The solvent (5 µL) was injected at a flow rate of 0.5 µL/min. A Shimadzu LCMS-8040 tandem mass spectrometer equipped with an electrospray ionization (ESI) source operating in negative and positive ionization mode was used for detection. LC-ESI-MS/MS data were processed using Lab Solutions software (Shimadzu). Phytochemicals were quantified using an MRM (multiple reaction monitoring) system

optimized for the selective detection and quantification of phytochemical compounds based on screening of specific phytochemical precursor transitions of ion fragments. The mass spectrometry conditions applied were as follows: drying gas flow (N₂) of 15 L/min; nebulization gas flow (N₂) of 3 L/min; DL temperature of 250°C; block temperature of 400°C, and interface temperature of 350°C.

2.2.4. Antioxidant activity of peppermint extract

2.2.4.1. DPPH radical scavenging assay

The protocol described by (Djenidi et al., 2020) was applied. To 100µl of extract or standard, 2.5 ml of freshly prepared methanolic solution of DPPH (0.04 mg/ml) was added. The mixture was vortexed and allowed to react for 30 min in the dark at room temperature. The absorbance was measured at 517nm using a spectrophotometer (Janway 7205). The percentage inhibition of the DPPH radical was calculated using the following equation: % inhibition = [(absorbance of control - absorbance of sample) / absorbance of control] x 100.

2.2.4.2. ABTS⁺ radical scavenging activity

Aqueous solutions of ABTS (7.4 mM) and potassium persulfate (2.6 mM) were mixed in a 1:1 (v/v) ratio and incubated in the dark for 16 hours. Then, 1000µl of the mixture was diluted in methanol until an absorbance of 1.049 ± 0.012 units was obtained at a wavelength of 743nm. Next, 1800 µl of this final mixture was added to 200µl of each extract or standard concentration. After 2 hours of incubation in the dark, the absorbance was read using a spectrophotometer (Janway 7205) at a wavelength of 734 nm (Almusallam et al., 2021). The percentage inhibition of the ABTS radical was calculated using the following equation: % inhibition = [(absorbance of control - absorbance of sample) / absorbance of control] x 100.

2.2.5. Characteristics analysis of yogurt formulated with peppermint extract

2.2.5.1. Sensory analysis

The organoleptic criteria (sour taste, freshness, aftertaste, adhesiveness, cohesiveness, odor, and color) of the yogurts were evaluated on a 10-point scale by a tasting panel of 10 judges aged 22 to 54 years from the Department of Agronomy at the University of Mostaganem (Algeria). The yogurt samples containing 0, 2, 4, and 6% of *HEEMP* were tasted during the post-acidification period on the 1st, 7th, 14th, and 21st day of storage at 4°C.

2.2.5.2. Yogurt oxidative properties

2.2.5.2.1. DPPH and ABTS radical scavenging activity

The antioxidant activity was evaluated on the 1st, 10th, and 20th days of storage at 4°C of experimental yogurts prepared with 0, 2, 4, and 6% of *HEEMP*. 1g of yogurt was centrifuged in a SIGMA centrifuge at a temperature of 4°C for 10 min at 9000 × g. The recovered supernatants were filtered through a syringe filter (ISOLAB Laborgerate GmbH, Eschau/Germany) with a membrane (0.45 µm) (Lee et al., 2021). The filtrates

were then stored at -20°C until their use within a maximum of 15 days. DPPH (Djenidi et al., 2020) and ABTS (Almusallam et al., 2021) tests were performed on the filtrates of the experimental yogurts.

2.2.5.2.2. Determination of thiobarbituric acid reactive substances (TBARS)

Lipid peroxidation was evaluated on the 1st, 10th, and 20th days of storage at 4°C of experimental yogurts prepared with 0, 2, 4, and 6% of *HEEMP* by measuring malondialdehyde (MDA) according to the method of Genot (1996). Aliquots of 2 g of yogurt were placed in glass tubes kept in ice to limit oxidation, then 100 µl of ascorbic acid and 16 ml of a 5% (w/v) TCA solution were added. After homogenization and centrifugation (SIGMA centrifuge) at 8000 × g for 15 min, 2ml of each supernatant were taken and added to 2 ml of TBA (20 mmol/l) in glass tubes and heated for 15 min in a water bath set at 100°C then cooled to room temperature. The absorbance at 532 nm was measured by a spectrophotometer (Janway 7205) against a blank consisting of 2ml TBA and 2ml TCA.

The measured absorbance was converted to MDA equivalent per kg of yogurt (mg EMDA/kg), according to the equation:

$$\text{Mg équivalent MDA/Kg} = (A_{532} \times V_{\text{TCA}} \times V_f) \times (0.72 / 1.56) \times m$$

Where: **A532**: absorbance at 532 nm; **V_{TCA}**: volume of TCA (16 ml); **V_f**: volume of filtrate collected; **m**: mass of the sample (g); **0.72/1.56**: corresponds to the molecular extinction coefficient of the TBA-MDA complex.

2.2.5.3. Yogurt fatty acids

The lipid profile of experimental yogurts prepared with 0% and 4% of mint extract was analyzed by gas chromatography coupled with a flame ionization detector (GC-FID). Cold extraction of the fat from the yogurt samples was carried out on the 1st and 20th day of storage at 4°C according to the method of Folch et al. (1957). The total fat content present in 100 ml of yogurt was calculated as follows:

$$\text{MGT} = \text{P2} - \text{P1}$$

where **P2** is the weight of the flask containing the lipids and **P1** is the weight of the empty flask. using potassium hydroxide according

Fatty acid methyl esters were prepared using potassium hydroxide according to the method described by Günes et al. (2017), and analyzed by gas chromatography GC-FID using a Perkin Elmer Clarus 500 chromatograph equipped with an FID detector and an apolar capillary column (RTX-2330) (USA) (30 m × 0.25 mm and 0.25 µm ID). Helium was used as the carrier gas. The oven temperature was set at 70°C for 2 min, increased from 70°C to 120°C at a rate of 4°C/min, from 120°C to 180°C at a rate of 2°C/min, held at 180°C for 3 min, increased from 180°C to 200°C at a rate of 4°C/min, held at 200°C for 3 min, and increased from 200°C to 230°C at a rate of 7°C/min, and held at 230°C for 5.21 min. The injection volume of the sample was 1 µl. The fatty acids were identified by comparing their retention times with standards, and the peak area percentages of the compounds were calculated based on FID data (Demirtas et al., 2011).

2.2.6. Statistical analysis

In the present study, the parametric results of the experiments performed in triplicate were statistically analyzed using a one-way and two-way analysis of variance (ANOVA) with complete randomization, and a comparison of means using the Newman-Keuls test. The sensory data obtained from a panel of 10 judges was treated using the non-parametric Friedman test. The statistical analysis was carried out using Stat box 6.4 software. The effects of the factors were evaluated at two probability levels of $p < 0.05$ and $p < 0.01$.

3. Results And Discussion

3.1. Phenolic compounds of the extract

In the present study, the 80% aqueous ethanol extraction (80:20; ethanol:water; v/v) of 10 g of the aerial part of *Mentha piperita* L. collected from the Algerian Sahara yielded an interesting extraction yield of 16.16%. According to the results of the colorimetric assay, the plant studied showed a considerable content of phenolic compounds (6.26 g GAE/100g DW) and flavonoids (3.40 g QE/100g DW), while it appears to contain very low concentrations of condensed tannins (0.09 g EC/100g DW) (Table 1). These values are considerably higher compared to those obtained in the hydroethanolic extract of the same plant, *Mentha piperita* L., collected in Iraq, whose total phenolic content was only 1.99 g GAE/100g DW and total flavonoid content was 1.31 g QE/100g DW (**Farnad et al., 2014**). This divergence in results is related to the geographical location and climate, which are external factors that greatly influence the composition of plants, especially those of the genus *Mentha* (**Brahmi et al., 2022**).

However, in a study conducted on *Mentha piperita* L, collected at the beginning of July in the same study region (Ouargla, Algeria) where our plant was cultivated, **Ait Chabane et al. (2021)** found total phenolic contents (154 mg GAE/g DW) and flavonoid contents (49.68 mg QE/g DW) that were significantly lower than those recorded in the present study (336.34 mg GAE and 182.9 mg QE/g DW for total phenolics and flavonoids, respectively). This could be attributed to the harvesting season, which strongly influences the concentration of phytochemical substances and their activities in plants (**Lemos et al., 2017; Ghimire et al., 2021**).

The extraction solvent also plays an important role in the quantitative and qualitative variation of the total polyphenolic composition of plant extracts. In this regard, **Sultana et al. (2009)** demonstrated that ethanol/water mixtures (80:20; ethanol:water; v/v) were able to extract the maximum amount of total phenols from the leaves of *Moringa oleifera* and *Aloe barbadensis* compared to the amounts obtained by using absolute ethanol. The effectiveness of aqueous solvents is linked to the presence of water, which promotes the desorption of the solute from the sample matrix (**Mustafa et al., 2011**) and weakens the hydrogen bonds established between the phenolic compounds and proteins within the plant matrix, thereby offering better solubility to the phenolic compounds (**Wissam et al., 2012**).

Table 1

3.2. Phenolic profile of the extract

The results of the LC-MS/MS analysis presented in Table 2 and Figure 1 revealed the presence of numerous phytochemical compounds in the hydroethanolic extract of *Mentha piperita* L (*HEEMP*), of which 93.35% are phenolic acids, with rosmarinic acid (339.88 mg/g of dry extract) being the major compound. Our results are consistent with the work of several authors who have suggested that phenolic acids, mainly rosmarinic acid and derivatives of caffeic acid, constitute more than 60% of the phenolic compounds in peppermint (**Misan et al., 2011; Mahendran & Rahman, 2020**). Our results also showed the presence of other phenolic acids in *HEEMP* such as quinic, caffeic, chlorogenic, protocatechuic, and p-coumaric acids at levels of 21.026, 1.629, 1.498, 0.918, and 0.406 mg/g of dry extract, respectively. A non-phenolic acid, fumaric acid (1.819 mg/g of dry extract), was also identified in our study. These values are higher than those reported by **Kürekci & Beyazit (2022)**, who demonstrated the presence of quinic, caffeic, and p-coumaric acids in methanolic extract of peppermint at levels of 2.243, 0.625, and 0.317 mg/g, respectively. The importance of caffeic acid has been demonstrated and widely explained, particularly its neuroprotective effects by modulating neuroinflammation and oxidative stress (**Caruso et al., 2022**). Flavonoids are known for their strong ability to trap different types of reactive oxygen species (**Benouchene et al., 2021**). The present study highlighted the presence of flavonoids in the analyzed extract mainly in the form of flavones, accounting for 76.83% of the detected flavonoids and strongly represented by Cyranoside (11.055 mg/g of dry extract) and Cosmosiine (5.424 mg/g of dry extract). These levels are higher than those reported by **Kürekci & Beyazit (2022)** (8.545 and 8.489 mg/g, respectively, for Cyranoside and Cosmosiine) in methanolic extract of *Mentha piperita* L collected in Turkey. In addition, flavanones accounted for 18.17% of the total flavonoids in our extract, which was found to be rich in hesperidin (3.925 mg/g of dry extract), hesperetin (0.248 mg/g), and naringenin (0.212 mg/g). The latter active compound was estimated by **Misan et al. (2011)** at 1.46 mg/g in a hydroethanolic extract of the same plant cultivated in Belgrade. These authors also demonstrated the presence of rutin at 2.18 mg/g of dry extract, whereas in our extract it was evaluated at only 0.638 mg/g of extract. It should be noted that the literature has reported a wide qualitative and quantitative diversity of results regarding the phytochemical composition of *Mentha piperita* L., due to several factors related to the environment and/or the plant itself (physiological state, genetic variation, and stage of harvest) (**Renoz et al., 2022**), extraction, drying, and analysis methods, the harvest season (**Mahendran & Rahman, 2020**), as well as the diversity of the polar characteristics.

Table 2

Figure 1

3.3. Antioxidant potential of the extract

The results shown in Table 3 demonstrated a high inhibition power of the DPPH radical exerted by *HEEMP* with an IC₅₀ (3.18 mg of *HEEMP* lyophilisate/ml) lower than that of ascorbic acid (0.16 mg of ascorbic acid/ml) and quercetin (0.03 mg of quercetin/ml) but considerably more interesting than the results of **Famad et al. (2014)**, who reported an IC₅₀ of 11.2 mg of lyophilisate/ml for the absolute ethanol extract of *Mentha piperita* L cultivated in Iran. It is clear that the use of a hydroethanolic solution at (20:80 v/v) as opposed to absolute ethanol during extraction allows for the obtainment of a peppermint extract richer in phenolic compounds and with a greater antioxidant potential."

A positive correlation between phenolic content and antioxidant activity has been confirmed by many authors (**Souilah et al., 2021**). **Generalić Mekinić et al. (2014)** suggested the existence of a positive relationship between the antioxidant properties of the plant extract and the content of rosmarinic acid, which is considered largely responsible for the antioxidant potential. This hypothesis seems to be confirmed by our results from the LC-MS/MS analysis of the studied *HEEMP*, which revealed an extraordinarily high level of rosmarinic acid. It should be noted that the flavones, which represent 76.83% of the total flavonoids detected by LC-MS/MS, undoubtedly contributed to the increased antioxidant activity of the tested extract.

However, for the same extract, the ABTS test revealed an IC₅₀ (2.49 mg of *HEEMP* lyophilisate/ml) lower than that obtained by the DPPH test (3.18 mg of *HEEMP* lyophilisate/ml). The presence in the extract of substances with absorption bands at the same wavelength as the ABTS[•] radical could explain this increase in absorbance (**Dieng et al., 2017**). Moreover, although it is accepted that the antioxidant effect of phytochemicals is proportionally related to their quantity in the extract, the synergistic effects of some mixtures of compounds cannot be neglected. It has been demonstrated that binary combinations of rosmarinic acid, gallic acid, caffeic acid, chlorogenic acid, rutin, and quercetin allow for better antioxidant activity. However, no antagonistic effect of some of these compounds was found (**Hajimehdipoor et al., 2014**). This is in line with our results, which clearly showed that the *HEEMP* contains a considerably important cocktail of phenolic compounds.

Table 3

3.4. Yogurt sensory analysis

The panelists' perception of sour taste seems to be significantly deteriorated ($p < 0.01$) at high incorporation rates of 6% *HEEMP* in yogurt due to a low production of lactate which gives yogurt its characteristic sour taste, as a result of the antibacterial action of phenolic compounds contained in *HEEMP* against starter bacteria that produce lactic acid by fermenting milk lactose. However, at extract concentrations of 2% and 4% in yogurt, the sour taste seems to be produced at sufficient levels. The 2% yogurt was even better appreciated than the control throughout the post-acidification period, due to its slightly acidic taste resulting from less production of lactic acid by *St* and *Lb*. In addition to lactate, traces of volatile compounds such as menthol and menthone (**Djenane et al., 2012**) that are constituents of the experimental extract seem to form an excellent blend with the usual yogurt flavors (acetaldehyde, diacetyl, acetone and acetylone) when added at a low dose of 2% to enhance acidity and freshness.

The tasting jury qualified the yogurts prepared with 6% *HEEMP* as having a very pronounced unpleasant aftertaste, probably due to the evident bitterness of certain phenolic compounds such as rosmarinic acid and flavonoids (**Weimann & Heinrich, 1997**) brought by the experimental extract at high doses in the product. Adhesiveness criteria, referring to the force required to remove the sticky material in the mouth during food intake (**Kose et al., 2018**), and cohesiveness, corresponding to the deformability of a material before breaking when crushed between the fingers, allowing panelists to appreciate the texturizing character of the lactic curd (**Kose et al., 2018**). The plain yogurt was the most appreciated for these two organoleptic attributes, which seem to be less accepted by panelists with increasing doses of mint extract in yogurt. This could be explained by the decline in the production of exopolysaccharides by the *St* and *Lb* lactic ferments,

whose proliferation and ability to elaborate these complex carbohydrates in the medium seem to be altered by the phenolic compounds of *HEEMP*, given that the rheological quality of yogurt, represented mainly by the sticky and cohesive criteria, is proportionally linked to the amounts of exopolysaccharides produced by *St* and *Lb* bacteria (Rawsan & Marshal, 1997). Our results are in line with those of several authors who have observed a drop in the rheological quality of yogurts during storage at 4°C, following the antimicrobial effect against these two germs exerted by the phenolic compounds of hydroethanolic extracts of *Thymus vulgaris* L. (Khelifi et al., 2018) and *Mentha piperita* L. (Ait Chabane et al., 2021), added notably at a high dose of 6%. Moreover, throughout the storage period, yogurts with 2% and 4% *HEEMP* were better evaluated and appreciated than the control, probably due to the manifestation of a pleasant mixture of odors combining the aromatic components brought by the mint extract, such as menthol, menthone, and limonene (Djenane et al., 2012), and those generated by the fermentative activity of the yogurt bacteria, such as diacetyl, acetaldehyde, and acetone (Beshkova et al., 1998). However, this bacterial activity of *St* and *Lb* producers of flavors and fragrances seems to be attenuated under the antibacterial effect of the phenolic extract introduced at high doses of 6%. Similarly, the tasting jury preferred the color of the yogurt with 2% extract to the natural whitish color of the control yogurt, while the yogurts with 4% and 6% extract seemed to have more intense and less pleasant colors, certainly conferred by the chlorophyll and certain phenolic compounds contained in the extract, including the well-known anthocyanins responsible for characteristic plant dyes (Farnad et al., 2014; Alappat B & Alappat J, 2020) (Table 4, figure 2).

Table 4

Figure 2

3.5. Yogurt antioxidant property

Although yogurt itself has a significant antioxidant function due to bioactive peptides from the metabolism of lactic acid bacteria (Kudoh et al., 2001; Gomez-Ruiz et al., 2008) contributing to the promotion of antioxidant potential and inhibition of lipid peroxidation (Virtanen et al., 2007), the addition of *HEEMP* is likely to further enhance this antioxidant activity. Indeed, on the first day of storage, compared to the control, the addition of only 2% extract to yogurt led to an increase ($p < 0.01$) in the scavenging activity of DPPH and ABTS radicals by 38.66% and 21.14%, respectively, while an extreme supplementation of 6% extract increased ($p < 0.01$) these values by 49.19% and 42.49%, respectively. Our results confirm those of many authors who have observed significant antioxidant activities with increasing levels of phenolic extract from olive leaves (Cho et al., 2020) and argel leaf extract (Mohamed Ahmed et al., 2021) incorporated into yogurt. Moreover, depending on the storage time at 4°C, all experimental yogurts showed a decrease in the scavenging activities of DPPH (9.58; 48.96; 49.09 and 55.33) and ABTS (12.98; 30.30; 35.20 and 36.05) radicals for *HEEMP* doses of 0, 2, 4 and 6%, respectively. However, at the end of the 20th day of storage, *HEEMP*-enriched yogurts still exhibited better antioxidant activities than the control. The reduction in scavenging activities of DPPH and ABTS radicals during storage has been reported by several authors (Cho et al., 2020; Mohamed Ahmed et al., 2021) and could be partly attributed to the formation of a complex between the polyphenols present in the incorporated extract and the proteins of the yogurt, thereby reducing the recovery of phenolic compounds and their antioxidant action (Mohamed Ahmed et al., 2021). The

effects of these interactions on antioxidant power would depend on both the type of phenols and the amino acid composition of the peptide compound, and the resulting protein-polyphenol complexes could therefore reduce or enhance antioxidant activity (Prigent et al., 2003).

On the other hand, the lipid peroxidation of the experimental yogurts was found to decrease (from an average of 1.14 to 0.94, 0.86, and 0.66 mg MDA/kg) proportionally with the addition of different doses (0, 2, 4, and 6%) of *HEEMP*, which seems to provide the products with greater lipid stability compared to the control. Depending on the storage time, a clear increase in TBARS values was observed in all experimental yogurts, with or without *HEEMP* enrichment. This result suggests that the addition of *HEEMP* could improve the lipid fraction and prolong the shelf life of the product during prolonged cold storage. Our results are consistent with those of many authors who have reported that the addition of phenolic extracts from date palm rachis (Almusallam et al., 2021), argel (Mohamed Ahmed et al., 2021), and algae (O'Sullivan et al., 2016) significantly reduces lipid peroxidation and remarkably improves the stability of yogurts.

Table 5

3.6. yogurt fatty acid

The lipid profile of yogurt is an important factor affecting its nutritional and sensory quality (Paszczyk & Tońska, 2022). The present study revealed a slight ($p>0.05$) decrease in the total fat content (MGT) of the products over the storage period. Khalil et al. (2022) also found that the fat content of yogurt decreased during refrigerated storage, which is probably due to the action of lipases from the yogurt starter bacteria. Lipolysis is considered one of the main biochemical phenomena that significantly affect the shelf life of many dairy products (Alirezalu et al., 2019). The addition of *HEEMP* seems to counteract this phenomenon, as the control sample showed a decrease in total fat content of about 0.02%, slightly higher than that observed for the yogurt enriched with *HEEMP*, which showed a decrease of only 0.014% over 20 days of storage. This is attributed to a reduction in lipolysis due to the antibacterial effect exerted by the phenolic compounds of the added *HEEMP* on the growth of the yogurt starter cultures.

Furthermore, the high richness of *Mentha piperita* L. leaves in fatty acids, estimated at about 11.64 mg/g (Savych et al., 2021), suggests that a certain proportion of the plant's lipid fraction was carried into the extract and reinforced the lipid content of the yogurt. In addition, in the present study, a variable increase in certain fatty acids was observed in all experimental samples with or without *HEEMP*, resulting from the variable activity of yogurt starter bacteria that adopt modification of the lipid profile as one of the adaptation strategies to variations in environmental conditions, including acidity of the medium (Tidona et al., 2018; Bahrami et al., 2020).

Moreover, the addition of hydroethanolic extract of *Mentha piperita* L. did not significantly influence ($p>0.05$) the contents of saturated (SFA) and unsaturated (UFA) fatty acids in the experimental yogurts. However, from the beginning to the end of refrigerated storage at 4°C, a dominance of SFA over UFA was observed in the experimental yogurts. A high degree of unsaturation in fatty acids undoubtedly increases their susceptibility to peroxidation (Halliwell et al., 1999), which is responsible for the unacceptable rancid flavors of yogurt. A similar trend in terms of SFA abundance compared to UFA was observed in another study

conducted on yogurts supplemented with fenugreek and chamomile extracts during 14 days of storage at 6°C (Caleja et al., 2016).

The chromatographic analysis (GC-FID) also showed a reduction in the yogurt's contents of short- and medium-chain fatty acids (SMCFA), including butyric acid (C4:0), caproic acid (C6:0), caprylic acid (C8:0), capric acid (C10:0), and lauric acid (C12:0), by adding *HEEMP*. This decrease in SMCFA is not very significant ($p>0.05$) with the addition of *HEEMP*, but it is certainly favorable for health since these fatty acids are most involved in coronary diseases (Bianchi et al., 2017) and in terms of taste since SMCFA have a determining influence on the characteristic flavor of the final product. Butyric, caproic, and caprylic acids impart a sour taste to yogurt (Savinova et al., 2022), while high levels of capric and lauric acids are likely to give it a soapy taste (Su et al., 2018).

Moreover, adding *HEEMP* at 4% to the yogurt made in this study appears to improve its omega-3 content, which is essential for the development and proper functioning of the human body due to its beneficial effects on the brain, heart, eyes, joints, skin, and human behavior (Qin et al., 2017; Almasi et al., 2021). On the 1st and 20th days of storage at 4 °C, the proportion of omega-3 in the yogurt was increased by 0.20% and 0.12% compared to the control, respectively. This increase in omega-3 content can be explained by two hypotheses: the first involves the antioxidant properties of the phenolic compounds in *HEEMP*, which protect this type of fatty acid from potential hydrolysis (Bakry et al., 2019), while the second is the richness of the extract itself in omega-3. A positive correlation between phenolic compounds and omega-3 has been demonstrated in a study on *Thymelaea hirsuta* L (Bouzouina et al., 2016). Additionally, Savych et al. (2021) demonstrated a high proportion of omega-3 in the hydroalcoholic extract of *Mentha piperita* L. leaves using GCMS analysis. Alpha-linolenic acid (ALA, C18:3n3) is among the main omega-3s in yogurt supplemented with 4% *HEEMP*. According to the literature, a diet rich in ALA is associated with a lower risk of cancer, coronary heart disease, and acute myocardial infarction (Lin et al., 2018). ALA is also a precursor for the synthesis of polyunsaturated fatty acids such as EPA (cis-5,8,11,14,17-eicosapentaenoic acid, C20:5n3) and DHA (cis-4,7,10,13,16,19-docosahexaenoic acid, C22:6n3), which are also recognized for their positive effects on the cardiovascular system (Lin et al., 2018). During the experiment, adding the extract to the yogurt significantly ($p<0.01$) improved its ALA content by about 0.19% compared to the control. Furthermore, several studies suggest that a high omega-6/omega-3 ratio is a pathogenic factor for many diseases, including cancer, cardiovascular, inflammatory, autoimmune, and cognitive diseases, as well as obesity (Katan et al., 2021). From a nutritional point of view, omega-6/omega-3 ratios varying from 1:1 to 5:1 are recommended (Lupette & Benning, 2020). In this study, the omega-6/omega-3 ratios of 4.32 and 4.11 recorded on the 1st and 20th day of storage in the 4% *HEEMP* yogurt respectively comply with the accepted norm, unlike the control which had lower values of 6.764 and 5.21.

Table 6

Conclusion

The hydroethanolic extract of *Mentha piperita* L showed a considerable content of phenolic compounds such as rosmarinic acid with strong antioxidant power, which even at a low dose added to the steamed

yogurt, considerably protected its lipid fraction while preserving its organoleptic quality during storage. Thus, compared to the control, the yogurt prepared with 4% peppermint extract stood out for its high antioxidant power, acceptable organoleptic quality, greatly improved values of α -linolenic acid and omega-3, and a ω_6/ω_3 ratio that complies with accepted standards. This product can be suggested as a new functional food that can meet the nutritional and dietary needs of consumers.

Declarations

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- Mahfuz Elmastas. : Aalyse du profil des acides gras par GC-FID.
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Authors declaration

Tous les auteurs ont relu le manuscrit

Data Availability Statement

The authors state data availability in the main file of the manuscript.

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The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper

Conflict of Interest Statement

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Tables

Table 1. Quantification of *HEEMP* phenolic compounds .

Bioactive compounds	Samples	Content
Total phenolic compounds	Plant powder (g GAE/100g PP)	006.26 ±00.16
	Liquid extract (mg GAE /ml <i>HEEMP</i>)	041.29 ± 01.06
	Lyophilized extract (mg GAE /g LE)	336.34 ± 08.66
Total flavonoids	Plant powder (g QE/100g PP)	003.40 ± 00.10
	Liquid extract (mg QE/ml <i>HEEMP</i>)	022.45 ±00.68
	Lyophilized extract (mg EQ/g LE)	182.90 ± 05.54
Condensed tannins	Plant powder (g CE /100g PP)	000.09 ± 00.01
	Liquid extract (mg CE/ml <i>HEEMP</i>)	000.59 ±00.07
	Lyophilized extract (mg CE/g LE)	004.80 ± 00.54
Yield	16.16%	

The results are expressed as mean values followed by the corresponding standard deviations, with a number of repetitions n=3; PP: plant powder; *HEEMP*: hydroethanolic extract of extract of *Mentha piperita* L.; L: lyophilized extract; GAE: gallic acid equivalent; QE: quercetin equivalent; CE: Catechin equivalent.

Table 2. Profile of bioactive compounds in HEMP by LC-MS/MS analysis.

Identified compounds		RT ^a (min)	LOD/LOQ (µg/l) ^f	U ^g	Content (mg/g lyophilized extract)
Organic acids	Fumaric acid	03.90	135.7/167.9	0.0091	01.819 ± 0.02
Phenolic acids	Quinic acid	03.00	25.7/33.3	0.0372	21.026 ± 00.78
	Gallic acid	04.40	13.2/17.0	0.0112	00.15 ± 00.02
	Protocatechuic acid	06.80	21.9/38.6	0.0350	00.918 ± 00.03
	Gentisic acid	08.30	18.5/28.2	0.0167	00.045 ± 00.00
	Chlorogenic acid	08.40	13.1/17.6	0.0213	01.498 ± 00.03
	Caffeic acid	12.10	7.7/9.5	0.0152	01.629 ± 00.03
	<i>p</i> -Coumaric acid	17.80	25.9/34.9	0.0194	00.406 ± 00.08
	Salicylic acid	21.80	6.0/8.3	0.0158	00.103 ± 00.02
	Rosmarinic acid	26.60	16.2/21.2	0.0130	339.88 ± 04.42
Flavones	Cynaroside	23.70	12.1/16.0	0.0366	11.055 ± 00.41
	Cosmosiin	28.20	6.3/9.2	0.0083	05.424 ± 00.05
	Luteolin	36.70	2.6/4.1	0.0313	00.904 ± 00.03
	Chrysin	40.50	1.5/2.8	0.0323	00.004 ± 00.00
	Acacetin	40.70	1.5/2.5	0.036	00.805 ± 00.03
	Apigenin	38.20	1.3/2.0	0.0178	00.364 ± 00.01
Isoflavones	Genistein	36.90	3.7/5.3	0.0337	00.051 ± 0.00
Flavanones	Hesperidin	25.80	19.0/26.0	0.0335	03.925 ± 00.13
	Naringenin	35.90	2.6/3.9	0.0392	00.212 ± 00.01
	Hesperetin	36.70	2.6/3.9	0.0392	00.248 ± 00.01
	Amentoflavone	39.70	2.8/5.1	0.0340	00.003 ± 00.00
Flavonols	Rutin	25.60	15.7/22.7	0.0247	00.638 ± 0.016
	Isoquercitrin	25.60	8.7/13.5	0.0220	00.393 ± 0.0900
	Astragalin	30.40	6.6/8.2	0.0114	00.065 ± 0.000
	Kaempferol	37.90	10.2/15.4	0.0212	00.061 ± 0.001
Phenolic aldehyde	Protocatechuic aldehydes	8.50	15.4/22.2	0.0396	00.164 ± 0.006

The results are presented as mean values and corresponding standard deviations, with a number of repetitions n=03; RTa: Retention Time; LOD/LOQ (µg/L): Limit of Detection/Quantification; Ug (%): relative uncertainty percentage at a confidence level of 95% (k=2).

Table 3. Evaluation of the antioxidant activity of HEMP compared to that of ascorbate and quercetin.

Plant extracts and standards.	IC50		
	DPPH		ABTS
Freeze-dried extract (mg of freeze-dried HEMP)	03.18 ± 00.06		02.49 ±0 0.09
Ascorbic acid (mg of ascorbic acid/ml)	00.16 ± 00.02		01.36 ± 00.12
Quercetin (mg of quercetin/mL)	00.03 ± 00.06		01.17 ± 00.18

The results are expressed as means, followed by the corresponding standard deviations, with a number of repetitions n equal to 3 (n=03); IC50: concentration of extract or standard required to inhibit 50% of the DPPH radical or reduce the trapping of 50% of ABTS. HEMP: hydroethanolic extract of *Mentha piperita* L.

Table 4. Impact of the incorporation of *Mentha piperita*.L extract on the sensorial quality of steamed yogurt.

Sensory attribute	Storages (days)	Extrait concentration				Effect of the extract rate
		0%	2%	4%	6%	
sour taste	1	13 ^d	15,5 ^c	25 ^b	29,5 ^a	p 0,01
	7	15 ^d	14 ^c	27 ^b	36 ^a	p 0,01
	14	18 ^c	15 ^d	28 ^b	34 ^a	p 0,01
	21	20,5 ^c	16,5 ^d	28 ^b	35 ^a	p 0,01
Freshness	1	22,5 ^b	15 ^d	18 ^c	34 ^a	p 0,01
	7	27 ^b	17 ^c	18 ^{bc}	33,5 ^a	p 0,01
	14	27 ^b	16 ^d	18 ^c	33 ^a	p 0,01
	21	29 ^b	13 ^d	16,5 ^c	32,5 ^a	p 0,01
Aftertaste	1	16 ^d	21,5 ^c	29 ^b	33,5 ^a	p 0,01
	7	17,5 ^c	18,5 ^{bc}	29,5 ^b	34,5 ^a	p 0,01
	14	17,5 ^d	23 ^c	30 ^b	35,5 ^a	p 0,01
	21	20,5 ^d	24,5 ^c	28 ^{ab}	30,5 ^a	p 0,01
adhesiveness	1	16,5 ^c	22 ^b	27,5 ^{ab}	32 ^a	p 0,01
	7	12,5 ^d	20 ^c	28 ^b	37,5 ^a	p 0,01
	14	13,5 ^c	22 ^b	30 ^{ab}	34 ^a	p 0,01
	21	13 ^c	21 ^b	31 ^{ab}	35,5 ^a	p 0,01
Cohesiveness	1	16,5 ^c	23 ^b	28 ^{ab}	30,5 ^a	p 0,01
	7	16 ^c	21,5 ^b	29 ^{ab}	33,5 ^a	p 0,01
	14	13,5 ^c	22,5 ^b	29,5 ^{ab}	34 ^a	p 0,01
	21	10,5 ^c	22,5 ^c	30,5 ^b	35 ^a	p 0,01
Odor	1	29,5 ^{ab}	19 ^b	20 ^b	31,5 ^a	p 0,01
	7	28 ^{ab}	18 ^b	19 ^b	30 ^a	p 0,01
	14	25 ^a	18 ^b	20 ^{ab}	27,5 ^a	p 0,01
	21	24 ^b	17 ^c	20 ^{bc}	31,5 ^a	p 0,01

Color	1	16,5 ^c	15 ^c	18 ^b	28 ^a	p 0,01
	7	16 ^c	15 ^c	20 ^b	30,5 ^a	p 0,01
	14	18 ^c	15 ^c	22 ^b	31,5 ^a	p 0,01
	21	17,5 ^c	16 ^c	23 ^b	28,5 ^a	p 0,01

Results are expressed as rank sum, with number of replicates n=10; D:days P 0.05 non-significant effect of the studied factor (rate of hydro-ethanol extract of *Mentha piperita*.L incorporated in yogurt); p 0.01: highly significant effect of the studied factor; a, b: homogeneous groups of statistical comparison of the sums of the ranks two by two.

Table 5. Evolution of antioxidant potential (% inhibition of DPPH and ABTS radicals) and lipid peroxidation (TBARS) level (mg MDA/kg) of experimental steamed yogurt.

Antioxidant properties	Storages (days)	Incorporation rate (%) of pepermint extract (Mean Int. F1XF2, n=03)				Means $\pm$ Standard Error (F2, n=12)
		0%	2%	4%	6%	
DPPH	1	19.36 ^h $\pm$ 00.66	58.02 ^d $\pm$ 0.99	63.25 ^b $\pm$ 0.38	68.55 ^a $\pm$ 0.38	52.30 ^a $\pm$ 0.56
	10	15.65 ⁱ $\pm$ 00.32	51.86 ^f $\pm$ 0.65	59.68 ^c $\pm$ 0.93	62.36 ^b $\pm$ 0.23	47.39 ^b $\pm$ 0.51
	20	09.58 ^j $\pm$ 00.39	48.96 ^g $\pm$ 0.40	49.09 ^g $\pm$ 0.24	55.33 ^e $\pm$ 0.50	40.74 ^c $\pm$ 0.34
Means $\pm$ Standard Error (F1, n=09)		14.86 ^d $\pm$ 0.42	52.95 ^c $\pm$ 0.63	57.34 ^b $\pm$ 0.52	62.08 ^a $\pm$ 0.34	-
ABTS	1	17.38 ^h $\pm$ 00.33	38.52 ^e $\pm$ 0.87	51.19 ^b $\pm$ 0.05	59.87 ^a $\pm$ 0.92	41.74 ^a $\pm$ 0.56
	10	15.67 ⁱ $\pm$ 0.37	31.54 ^g $\pm$ 1.59	43.44 ^d $\pm$ 0.61	45.68 ^c $\pm$ 0.49	34.08 ^b $\pm$ 0.77
	20	12.98 ^j $\pm$ 0.42	30.30 ^g $\pm$ 0.59	35.20 ^f $\pm$ 1.28	36.05 ^f $\pm$ 0.56	28.64 ^c $\pm$ 0.67
Means $\pm$ Standard Error (F1, n=09)		15.34 ^d $\pm$ 0.32	33.45 ^c $\pm$ 0.95	43.28 ^b $\pm$ 0.71	47.20 ^a $\pm$ 0.59	-
TBARS	1	0.78 ^f $\pm$ 0.05	0.67 ^g $\pm$ 0.01	0.58 ^h $\pm$ 0.02	0.32 ^j $\pm$ 0.04	00.59 ^c $\pm$ 0.03
	10	1.11 ^e $\pm$ 0.01	0.72 ^g $\pm$ 0.02	0.65 ^g $\pm$ 0.03	0.48 ⁱ $\pm$ 0.02	00.74 ^b $\pm$ 0.02
	20	1.52 ^a $\pm$ 0.07	1.42 ^b $\pm$ 0.01	1.34 ^c $\pm$ 0.02	1.20 ^d $\pm$ 0.08	1.37 ^a $\pm$ 0.04
Means $\pm$ Standard Error (F1, n=09)		1.14 ^a $\pm$ 0.04	0.94 ^{bc} $\pm$ 0.01	0.86 ^c $\pm$ 0.02	0.66 ^d $\pm$ 0.04	-

The results are expressed as mean values followed by their corresponding standard deviations, with a number of repetitions n=03; h: hours, D: days. All the studied factors (incorporated extract, period, and interaction of the two factors) show a highly significant effect ($p < 0.01$). a, b, c, d, e, f: homogeneous groups of comparison of the means of the factor over time two by two according to the Newman and Keuls test.

Table 6. Fatty acid profile of experimental yogurts (as % of total lipids).

Periods	1st Day		20th Day		Effect of HEMP
Yogurts	0% HEMP	4% HEMP	0% HEMP	4% HEMP	
Total lipids (g/100g)	01.843±0.09	01.844±00.06	01.822±00.03	01.83±00.05	p>0.05
C4:0	01.485 ^b ±00.07	01.425 ^b ±00.04	01.644 ^b ±00.03	01.486 ^b ±00.04	P<0.01
C6:0	01.286±00.06	01.253±00.04	01.329±00.02	01.269±00.03	p>0.05
C8:0	00.98±00.05	00.962±00.03	01.003±00.012	00.965±00.03	p>0.05
C10:0	02.556±0.13	02.479±00.08	02.576±00.042	02.499±00.07	p>0.05
C11:0	00.05 ^a ±00.00	00.049 ^a ±00.00	00.04 ^c ±00.00	00.045 ^b ±00.00	p<0.01
C12:0	03.354±0.17	03.256±00.10	03.318±00.06	03.328±00.09	p>0.05
C13:0	00.091 ^c ±0.01	00.124 ^a ±00.00	00.1 ^b ±00.00	00.092 ^b ±00.00	p<0.05
C14:0	12.042±00.6	11.773±00.36	11.882±00.19	11.97±00.32	p>0.05
C14:1	00.641±00.03	00.616±00.02	00.622±00.01	00.619±00.012	p>0.05
C15:0	01.321±00.07	01.302±00.04	01.304±00.02	01.347±00.04	p>0.05
C16:0	35.249±01.75	35.208±01.07	35.075±00.57	35.926±00.9	p>0.05
C16:1	02.173±00.11	02.253±00.07	02.266±00.037	02.187±00.06	p>0.05
C17:0	00.666 ^a ±00.03	00.615 ^b ±00.02	00.632 ^{ab} ±00.01	00.502 ^c ±00.01	p<0.01
C18:0	08.994±00.45	09.199±00.28	09.14±00.148	09.478±00.25	p>0.05
C18:1n9	24.173±01.2	24.158±00.73	24.317±00.4	23.542±00.62	p>0.05
C18:2n6	02.766 ^a ±00.14	02.556 ^b ±00.08	02.816 ^a ±00.05	02.666 ^{ab} ±00.07	p<0.05
C20:0	00.131 ^b ±00.01	00.153 ^a ±00.01	00.133 ^b ±00.00	00.138 ^b ±00.00	p<0.01
C18:3n3	00.338 ^d ±00.02	00.529 ^b ±00.02	00.448 ^c ±00.01	00.637 ^a ±00.02	p<0.01
C20:1n9	01.148 ^a ±00.06	01.118 ^a ±00.03	00.941 ^b ±00.02	00.986 ^b ±00.03	p<0.01
C20:2	00.051 ^b ±00.00	00.057 ^a ±00.00	00 ^c	00 ^c	p<0.01
C20:3n3	00.093 ^c ±00.01	00.099 ^b ±00.00	00.118 ^a ±00.00	00.052 ^d ±00.00	p<0.01
C22	00.062 ^b ±00.00	00.074 ^a ±00.00	00.076 ^a ±00.00	00.075 ^a ±00.00	p<0.01
C20:3n6	00.149 ^b ±00.01	00.157 ^{ab} ±00.00	00.135 ^c ±0.00	0.166 ^a ±00.00	p<0.01
C24:0	00.198 ^b ±00.01	00.623 ^a ±00.02	00.085 ^d ±0.00	0.107 ^c ±00.003	p<0.01

SFA	68.462±03.39	68.46±02.07	68.338±01.11	69.211±01.82	p>0.05
UFA	31.532±01.56	31.545±00.96	31.663±00.51	30.855±00.81	p>0.05
SFA/UFA	02.171±00.11	02.171±00.07	02.158±00.04	2.243±00.06	p>0.05
MUFA	28.14±01.40	28.146±00.85	28.146±00.46	27.334±00.72	p>0.05
PUFA	03.398±00.17	03.399±00.10	03.517±00.06	3.521±00.09	p>0.05
SMCFA	09.712±00.48	09.426±00.29	09.911±00.16	9.546±00.21	p>0.05
LCFA	90.3±04.47	095.985±02.91	90.09±01.46	90.492±02.38	P>0.05
ω-3	00.431 ^d ±00.02	00.628 ^b ±00.02	00.566 ^c ±00.01	0.689 ^a ±00.02	p<0.01
ω-6	02.915±00.14	02.713±00.08	02.951±00.05	2.832±00.08	p>0.05
ω-6/ ω-3	06.764 ^a ±00.34	04.321 ^c ±00.13	05.214 ^b ±00.09	4.111 ^c ±00.11	p<0.01

The results are expressed as mean values and standard deviations; For each group, the number of repetitions is 03 (n = 03); HEMP: Hydroethanolic extract of *Mentha piperita* L.; p<0.01: highly significant effect of the studied factor (Mint extract content incorporated in yogurt); p<0.05: significant effect of the studied factor; p>0.05: non-significant effect of the studied factor; Means in the same row with different subscripts are significantly different at the 5% probability threshold;; SFA: Saturated Fatty Acids; UFA: Unsaturated Fatty Acids; SFA/UFA: Saturated/Unsaturated Fatty Acid Ratio; MUFA: Mono Unsaturated Fatty Acid; PUFA: Poly Unsaturated Fatty Acid; SMCFA: Short and medium chain fatty acids; ω-3: Omega 3; ω-6: Omega 6.

Figures

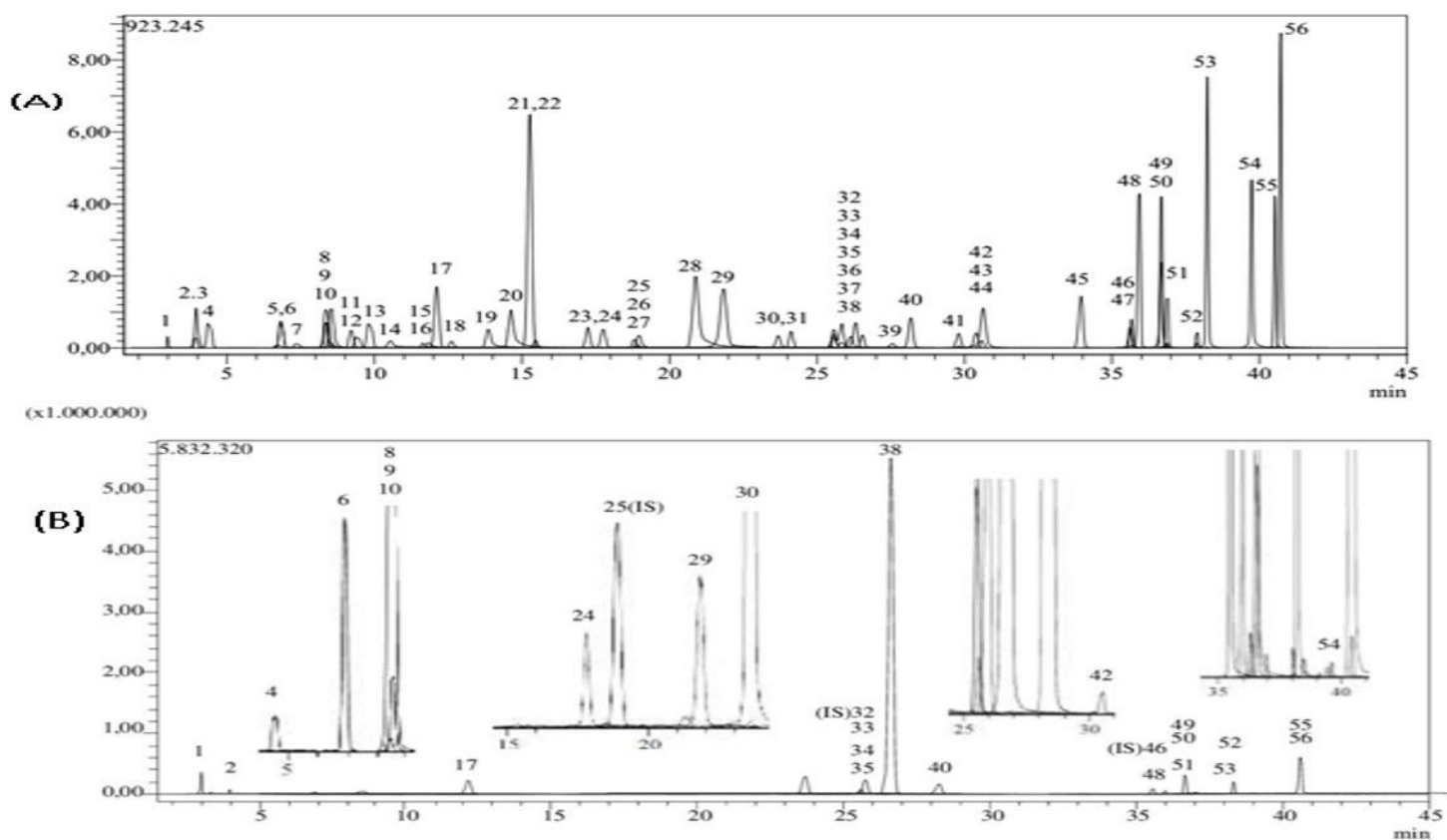


Figure 1

LC-MS/MS chromatograms consisting of (A) TIC (Total Ion Chromatogram) chromatogram of standard phenolic compounds, and (B) chromatogram of ethanol:water extract of *Mentha piperita* L. The TIC chromatogram of standard phenolic compounds includes the following compounds numbered from 1 to 56: (1:Quinic acid, 2 :Fumaric acid, 3 :Aconitic acid, 4 :Gallic acid, 5 :Epigallocatechin, 6 :Protocatechuic acid, 7 :Catechin, 8 :Gentisic acid, 9 :Chlorogenic acid, 10 :Protocatechuic aldehyde, 11:Tannic acid, 12 :Epigallocatechin gallate, 13 :Cynarin, 14 :4-OH Benzoic acid, 15 :Epicatechin, 16 :Vanillic acid, 17 :Caffeic acid, 18 :Syringic acid , 19 :Vanillin, 20 :Syringic aldehyde, 21 :Daidzin, 22 :Epicatechin gallate, 23 :Piceid, 24 :p-Coumaric acid, 25 :Ferulic acid-D3-IS, 26 :Ferulic acid, 27 :Sinapic acid, 28 :Coumarin, 29 :Salicylic acid, 30 :Cyranoside, 31 :Miquelianin, 32 :Rutin-D3-IS, 33 :Rutin, 34 :isoquercitrin, 35 :Hesperidin, 36 :O-Coumaric acid, 37 :Genistin, 38 :Rosmarinic acid, 39 :Ellagic acid, 40 :Cosmosiin, 41 :Quercitrin, 42 :Astragalin, 43Nicotiflorin, 44 :Fisetin, 45 :Daidzein, 46 :Quercetin-D3-IS, 47 :Quercetin, 48 :Naringenin, 49 :Hesperetin, 50 :Luteolin, 51 :Genistein, 52 :Kaempferol 53 :Apigenin, 54 :Amentoflavone, 55 :Chrysin, 56 :Acacetin).

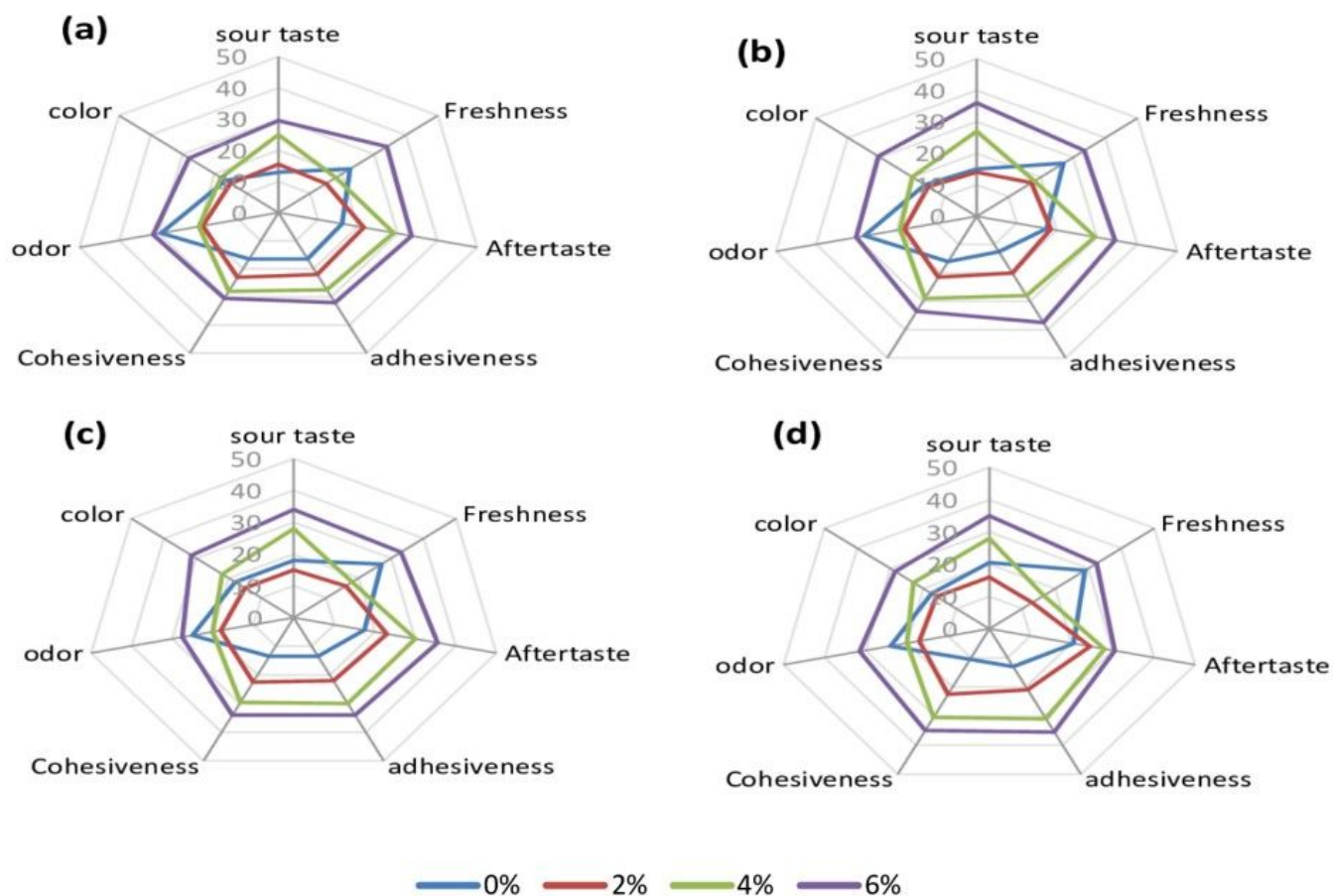


Figure 2

Effect of incorporation of hydroethanolic extract of *Mentha piperita* L. on the sensory quality of a fermented yogurt. (a) Day 1; (b) Day 7; (c) Day 14; (d) Day 21.