Preparation and evaluation of a functional effervescent powder based on inclusion complexes of orange oil and β-cyclodextrin derivatives

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Research Article

Keywords: Orange oil, essential oils, 2-Hydroxypropyl-β-cyclodextrin, epichlorohydrin- β-cyclodextrin, effervescent powder, inclusion complex nanoparticles.

Posted Date: November 4th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-2226110/v1

Abstract

Flavoured functional effervescent powders are becoming increasingly popular by consumers due to their health benefits and easy dissolution. In present study, orange flavoured effervescent powders having functional properties were prepared. Orange oil (O) was blended with different essential oils (EOs) having high antioxidant activity. The orange oil and the O-EOs blends were subjected to gas chromatography-mass spectrometry (GC-MS) analysis and evaluation of radical scavenging activity. Combinations of two water soluble β-cyclodextrine polymers, 2-hydroxypropyl-β-cyclodextrine (2-HP-β-CD) and epihydrin-β-cyclodextrin (EPI-β-CD) were prepared at different molar ratios (3: 1 and 1: 3, F1 and F2, respectively). The O-EOs blends that showed the highest antioxidant activities and best odour qualities were encapsulated with F1 and F2, separately. The orange flavoured inclusion complexes were prepared by freeze drying method. The particle sizes of the inclusion complex powders were in the nanoscale. Characterization of the inclusion complexes nanoparticles were performed by scanning electron microscopy (SEM), Fourier transform infrared microscopy (FT-IR). The results confirmed the successful formation of the inclusion complexes. However, inclusion complex of O-EOs blend with F1 (O-F1C-IC) showed the smallest particle size (113.9 ± 15.9 nm), the more negative zeta potential (-27.1 ± 1.27 mV), the highest encapsulation efficiency (95.51%) and best odour quality. Therefore, it was mixed with an effervescent powder having high acceptable characteristics. The orange flavoured effervescent powder showed superior flowability.

1. Introduction

In the upcoming time, the global market for effervescent products will be grown significantly due to their enjoyable sensation, ease in swallowing and health benefits [1]. Global effervescent production market is expected to increase from USD 4.217 million in 2022 to USD 64.08 million in 2029 [2]. Effervescent products contain a mixture of acids such as citric acid (CA) and tartaric acid (TA) with sodium bicarbonate ($NaHCO_3$) that can react quickly after adding water and produce carbon dioxide (CO_2) gas causing a refreshing sparkle effect [1, 3]. Chemical preservatives are used to improve the microbiological and oxidative stability of carbonated beverages, however, rising health consciousness influenced people to shift towards healthy products. The beverages industries are capitalizing on this trend by producing effervescent products having functional properties to improve consumer health and lifestyle [4]. The use of essential oils (EOs) as functional ingredients in food and beverages industries is gaining momentum, both for their relatively safe status, their wide acceptance by consumers and their exploitation for potential multi-purpose functional use [5]. However, the use of EOs as natural preservatives and flavoures is greatly limited by their volatility and low water solubility. In addition to being aromatic compounds, EOs are sensitive to light and oxidation and thus lose their quality. Encapsulation is the main option to protect and preserve flavour in food products [6. In last years, encapsulation technology has been developed significantly for the design and formulation procedures for stabilization, solubilization and delivery of the active compounds in food and pharmaceutical industries [7]. Encapsulation with cyclodextrins (CDs) can improve the stability of sensitive ingredients such as EOs [6], extend their shelf-life, protect them from oxidation and control release of their components [8]. Cyclodextrins have a relatively hydrophilic external surface and a hydrophobic interior cavity [9]. This unique structure allows them to form inclusion complexes. Inclusion complex is a

unique form of chemical structure in which guest molecule is enclosed within the cavity of the host. Among CDs, β-cyclodextrin (β-CD) is the most widely used due to its suitable cavity size for common guests with molecular weight between 200 and 800 Da, availability and reasonable price [10]. However, due to its low aqueous solubility, the use of β-CD is limited in formulations [11]. To overcome this issue, hydrophilic derivatives of β-CD have been investigated. 2-Hydroxypropyl-β-cyclodextrin (2-HP-β-CD) derivative is relatively high aqueous soluble (above 60 g in 100 mL water at 25°C) with low toxicity and satisfactory inclusion ability [12]. Since few years, studies have been carried out toward the use of epichlorohydrin- β-cyclodextrin (EPI-β-CD) polymer [13, 14]. The polymerized form of β-cyclodextrin viz epichloro- β-cyclodextrin remains within the cavity structure of β-CD providing capability of forming inclusion complexes with a variety of guest molecules [15]. Furthermore, it is proved to be more effective compared to its parent β-CD [16]. Orange flavoured effervescent is very popular to consumers. The main objective of the present study was to prepare orange flavoured effervescent powder having promising antioxidant activity and high flavour quality. To achieve this aim the antioxidant activity of orange oil was improved by blending with essential oils (EOs) having high antioxidant activity. A mixture of two soluble derivatives of β-cyclodextrin (2-HP-β-CD and EPI-β-CD) at different molar ratios were used to encapsulate the orange - essential oils (O-EOs) blend that showed the highest antioxidant activity and flavour acceptability. The produced inclusion complexes were freeze dried and those showed the highest entrapping efficiency were subjected to different physicochemical analysis. The effervescent powders of the orange flavoured powder was prepared and subjected to further physical analysis.

2. Material And Methods

2.1. Materials

2.1.1. Orange oil and essential oils

Cooled pressed orange oil was purchased from EL-Marwa Food Industries Company. The essential oils of clean air dried aromatic plants (camphor, basil, marjoram and oregano), purchased from Ferrous Company, Giza, Egypt, were hydrodistilled separately and dried and kept in dark sealed glass vials until analysis.

2.1.2. Chemicals

The authentic volatile compounds, standard n-alkanes (C_8-C_{22}), 2,2-diphenyl-1-picryhydrazyl ($DPPH^{\cdot}$) and 2,2-azinobis (2-ethyl-benzothiazoline-6-sulfonic acid) ($ABTS^{+}$), citric acid(CA), tartaric acid (TA), $NaHCO_3$ and fructose, 2-Hydroxypropyl-β-cyclodextrin (2-HP-β-CD) and epichlorohydrin- β-cyclodextrin (EPI-β-CD) were purchased from Sigma Aldrich Chemical Co. (St. Louis, MO, USA) and Merck (Darmstadt, Germany). All other chemicals were of analytical grade and the solvents were purified and distilled before use.

2.2. Methods

2.2.1. Preparation of orange oil-essential oil blends

Orange oil (100 µL) was blended with different amounts (0.25 µL, 0.50 µL and 0.75 µL) of the essential oils of camphor, basil and marjoram to improve its antioxidant activity, while preserving the characteristic flavour. The blends showed the best results were blended with 0.1 µL of oregano essential oil.

2.2.1.1. Odour acceptability evaluation

The odour acceptability of each orange oil- essential oil (O-EO) blend was conducted by 10 well trained panelists drawn from Food Technology and Nutrition Institute. The panelists sniffed and scored the perceived orange aroma of each blend on a category scale 0.0 (not acceptable) to 10.0 strongly acceptable.

2.2.1.2. Antioxidant activity assays

The antioxidant activity of orange oil, EOs and their blends was assessed by 2,2-diphenyl-1-picrylhydrazyl ($DPPH^{\cdot}$) and 2,2-azinobis (2-ethyl-benzothiazoline-6-sulfonic acid) ($ABTS^{+}$) assays according to Yen and Chen [17], Re et al. [18], Arnao [19], respectively.

Results were expressed as percentage of DPPH or ABTS radical inhibition according to Eq. 1.

Inhibition of radical activity (%) = $[(A_0 - A_s)/A_0] \times 100$ Eq. 1

Where: A_0 is the absorbance of the control sample and A_s is the absorbance of sample. All preparation and assays were performed in triplicate

2.2.1.3. Gas chromatography-mass spectrometry (GC-MS) analysis

Orange oil, EOs and their blends were subjected to GC-MS analysis using a gas chromatography (Agilent 8890 GC System) coupled to a mass spectrometer (Agilent 5977B GC/MSD) and equipped with a HP-5MS fused silica capillary column (30 m, 0.25 mm i.d., 0.25 mm film thickness) according to Fadel et al. [20]. Qualitative analysis of the separated peaks was carried out by matching with data from the library of mass spectra (National Institute of Standard and Technology, NIST) and comparison with those of authentic compounds and published data [21]. While the quantitative analysis was performed by the normalization method.

2.2.2. Preparation of orange flavoured inclusion complexes (O-F-IC)

The inclusion complexes were prepared via freeze-drying method [22]. Two water soluble β-cyclodextrine derivatives, 2-HP-β-CD and EPI-β-CD, were mixed at 3: 1 and 1: 3 (F1 and F2) molar ratios, respectively. Each mixture (2g) was dissolved separately in 15 mL distilled water and stirred at 600 rpm for 30 min. The O-EOs blend showed the best odour acceptability and highest antioxidant activity was selected and added at different contents (200, 250 and 300 mg) to each aqueous solution of the β-CDs mixtures (Table 1S). The solutions were agitated using the vortex till disappearance of any oil droplets then stirred at 150 rpm for 48h to ensure complete entrapment of the O-EOs blend. The obtained inclusion complexes solutions were

lyophilized at − 80°C, under pressure of 0.01 m bar (Alpha-1-4 LSC model, CHRIST, freeze drier, Harz-Germany). The lyophilized inclusion complexes were stored in sealed containers inside a desiccator at 20°C until use.

Table 1
Odour sensory evaluation of orange oil blended with different essential oils.

Blends	Constituents	Odour acceptability
Blend 1	Orange oil + Marjoram oil 100 µL + 0.25 µL	8.8 ± 0.44^{a}
Blend 2	Orange oil + Marjoram oil 100 µL + 0.50 µL	6.2 ± 0.31^{b}
Blend 3	Orange oil + Marjoram oil 100 µL + 0.75 µL	5.6 ± 0.28^{c}
Blend 4	Orange oil + Basil oil 100 µL + 0.25 µL	8.2 ± 0.41^{d}
Blend 5	Orange oil + Basil oil 100 µL + 0.50 µL	6.8 ± 0.34^{e}
Blend 6	Orange oil + Basil oil 100 µL + 0.75 µL	6.0 ± 0.30^{bc}
Blend 7	Orange oil + Camphor 100 µL + 0.25 µL	6.4 ± 0.32^{be}
Blend 8	Orange oil + Camphor 100 µL + 0.50 µL	5.4 ± 0.27^{cf}
Blend 9	Orange oil + Camphor 100 µL + 0.75 µL	5.0 ± 0.25^{f}
Blend 10	Orange oil + Marjoram oil + Oregano oil 100 µL + 0.25 µL + 0.1 µL	6.8 ± 0.29^{e}
Blend 11	Orange oil + Basil oil + Oregano oil 100 µL + 0.25 µL + 0.1 µL	8.4 ± 0.42^{ad}
Blend 12	Orange oil + Camphor oil + Oregano oil 100 µL + 0.25 µL + 0.1 µL	7.4 ± 0.37^{g}
Same letters means insignificant difference at $p < 0.05$		

2.2.2.1. Encapsulation efficiency (EE)

The encapsulation efficiency of each orange flavoured inclusion complex was conducted according to Soliman et al. [23] by using spectrophotometer (Model UV-1601, SHIMADZU, Kyoto, Japan).

Encapsulation efficiency (EE, %) was calculated from the following Eq. 2.

EE (%) = W_0/W_1x 100 Eq. 2

W_0 = Quantity of loaded EO,

W_1 = Initial quantity of EO.

For each loaded sample EE were determined in triplicate.

The antioxidant activity of each entrapped O-EOs blend was determined as mention above.

2.2.3. Characterization of the orange flavoured inclusion complexes.

The following methods were selected to evaluate the physical properties, the particle morphology, group interaction of the O-EOs blend with the mixture of soluble polymers β-CDs (2-HP-β-CD + EPI-β-CD) of the inclusion complexes that showed the highest EE, best odour acceptability and highest antioxidant activity.

2.2.3.1. Scanning electron microscopy (SEM)

The surface morphology of the selected orange flavoured inclusion complexes was examined by scanning electron microscopy (QUANTA FEG 250, Oregon, USA) under 15 Kv accelerating voltage. Each sample was coated with a fine layer of gold/ palladium before viewing under 500 times magnification.

2.2.3.2. Fourier transform infrared spectroscopy (FT-IR) analysis

The possible interaction between the selected O-EOs blend and the mixture of β-cyclodextrine derivatives was detected by FT-IR analysis. The samples were scanned in infrared range of 400–4000 cm^{-1} (JASCO 6100, Tokyo, Japan). FT-IR samples were prepared in the form of potassium bromide pellets.

2.2.4. Preparation of effervescent powder

Different amount of citric acid (CA), tartaric acid (TA) and sodium bicarbonate ($NaHCO_3$) in the ratio of 1: 2: 3.5 were used for the formulation of the effervescent powder [24, 25]. Each formula was dissolved in 50 mL distilled water and investigated regarding their effect on effervescent time, clarity and pH of the obtained solutions. Effervescent time, the moment at which a clear solution was obtained after effervescent completion, was determined using stop watch [1]. Clarity of the obtained solutions was assessed visually, while pH was determined using a digital pH meter (Vstar 92, Orion Versa Star ^{Tm}, Thermo Fisher Scientific

INC, USA) at 25°C. The formula of the effervescent components that showed the acceptable results was selected and mixed with 5 g fructose, as a sweetener, and then mixed well with the selected orange flavoured inclusion complex (O-CDs-IC).

2.2.5. Evaluation of the physical properties of orange flavoured effervescent powder

2.2.5.1. Powder density

The bulk and tapped densities of the powder were determined according to the following equations 3 and 4 [25].

Bulk density (g/mL) = Weight of sample/Bulk volume Eq. 3

Tapped density (g/mL) = Weight of sample/Tapped volume Eq. 4

Bulk volume: is the volume of certain weight of sample measured by a graduated cylinder.

Tapped volume: is the minimum volume of sample reached by tapping the cylinder

2.2.5.2. Determination of compressibility Carr's index

Compressibility is the ability of a powder to decrease in volume under pressure. It is a simple, fast, and accurate method for predicting the powder flow characteristics [25]. Carr's index of the effervescent powder was calculated from Eq. 5.

Carr's Index (%) = Tapped density-Bulk density/Tapped density x 100 Eq. 5

2.2.5.3. Determination of Hausner ratio

The Hausner ratio for the effervescent powder was calculated by the following Eq. 6 [25].

Hausner ratio = Tapped density/ Bulk density Eq. 6

It is related to inter-particle friction and can be used to assess powder flow properties.

2.2.5.4. Measurement of angle of repose

The flowability of the selected orange flavoured effervescent powder was assessed by measuring the angle of repose by a fixed funnel method according to Hussain and Al-khedairy [25]. The angle of repose (θ) of the conical piles produced by allowing the sample flow through a funnel and fall freely onto a surface was calculated according to Eq. 7

$\theta = \tan^{-1}(h/r)$ Eq. 7

h: Height of conical pile ; r: the radius of conical pile

2.2.6. Statistical analysis

The data were statistically processed to estimate the average ± standard deviation (SD) of triplicates. Statistical analysis was performed by one-way analysis of variance (ANOVA) using SPSS program for windows (Version 21) (SPSS, IBM Corporation, Armonk, New York, USA). Least significant differences (LSD) was performed at $P < 0.05$.

3. Results And Discussions

3.1. Chemical composition of the essential oils

The essential oils (EOs) that are expected to possess promising antioxidant activity (camphor, basil, marjoram and oregano) were used to impart the orange oil (O) high antioxidant activity (AOA). The chemical composition and antioxidant activity of orange oil (O) and those of camphor (Ca), basil (Ba), marjoram (Ma) and oregano (Or) were determined (Table 2S, Fig. 1a, b).

Table 2
Volatile compounds identified in orange oil (O) and its blends with camphor (Ca), basil (Ba), marjoram (Ma) and oregano (Or) essential oils.

Peak No.	KI[a]	Volatile compounds[b]	Area (%)[c]						
			Orange	O + Ca	O + Ca + Or	O + Ba	O + Ba + Or	O + Ma	O + Ma + Or
1	939	α-Pinene	0.74 ± 0.04	0.50 ± 0.03	0.53 ± 0.03	0.55 ± 0.03	0.88 ± 0.04	0.64 ± 0.03	0.50 ± 0.03
2	978	Sabinene	0.38 ± 0.02	----	0.28 ± 0.01	0.28 ± 0.01	0.44 ± 0.02	0.34 ± 0.02	0.41 ± 0.02
3	984	β-Pinene	0.56 ± 0.03	----	----	----	----	----	----
4	994	Myrcene	2.35 ± 0.12	1.74 ± 0.19	2.28 ± 0.11	2.09 ± 0.10	3.35 ± 0.17	2.45 ± 0.12	1.77 ± 0.09
5	1001	α-Phellendrene	----	----	0.10 ± 0.01	0.11 ± 0.01	0.27 ± 0.01	0.08 ± 0.00	0.18 ± 0.01
6	1011	α-Terpinene	----	----	0.21 ± 0.01	0.18 ± 0.01	0.08 ± 0.00	0.20 ± 0.01	0.15 ± 0.01
7	1016	Octanal	0.19 ± 0.01	----	0.18 ± 0.01	0.44 ± 0.02	0.28 ± 0.01	0.21 ± 0.01	0.15 ± 0.01
8	1024	ρ-Cymene	----	0.74 ± 0.04	----	----	----	----	0.29 ± 0.01
9	1034	D-Limonene	94.78 ± 4.74	87.16 ± 4.36	92.04 ± 4.36	93.28 ± 4.66	91.67 ± 4.58	93.88 ± 4.69	90.33 ± 4.52
10	1037	1,8-Cineole	----	----	----	0.14 ± 0.01	0.11 ± 0.01	----	----
11	1050	Z-Ocimene	----	----	----	----	0.04 ± 0.00	----	----
12	1064	γ-Terpinene	0.54 ± 0.03	----	----	----	----	----	0.47 ± 0.02

[a]Retention indices.

[b]Compounds listed according to their elution on DB5 column.

[c]Values (average of triplicate determinations) expressed as relative area percentages to total identified compounds.

O + Ca: Orange oil + Camphor; O + Ca + Or: Orange oil + Camphor oil + Oregano oil; O + Ba: Orange oil + Basil oil; O + Ba + Or: Orange oil + Basil oil + Oregano oil

; O + Ma: Orange oil + Marjoram oil; O + Ma + Or: Orange oil + Marjoram oil + Oregano oil

Peak No.	KI[a]	Volatile compounds[b]	Area (%)[c]						
			Orange	O + Ca	O + Ca + Or	O + Ba	O + Ba + Or	O + Ma	O + Ma + Or
13	1095	E-Sabinene hydrate	----	----	----	----	0.07 ± 0.00	----	0.13 ± 0.01
14	1104	Terpinolene	----	----	----	----	----	----	0.31 ± 0.02
15	1106	Linalool	0.26 ± 0.01	1.66 ± 0.08	0.63 ± 0.03	0.51 ± 0.03	0.69 ± 0.03	0.44 ± 0.02	0.39 ± 0.02
16	1150	Camphor	----	1.59 ± 0.08	1.14 ± 0.06	0.09 ± 0.00	0.13 ± 0.01	0.09 ± 0.00	----
17	1180	Terpinen-4-ol	----	----	----	----	----	0.11 ± 0.01	1.15 ± 0.06
18	1193	α-Terpineol	----	----	0.10 ± 0.01	----	----	----	0.25 ± 0.01
19	1199	E-Piperitol	----	----	----	----	0.09 ± 0.00	----	----
20	1201	ρ-Menth-1-en-4-ol	----	0.40 ± 0.02	0.12 ± 0.01	----	----	----	----
21	1203	Octanol acetate	----	----	----	0.16 ± 0.01	----	----	----
22	1207	Thymol methyl ether	----	1.36 ± 0.07	----	----	0.18 ± 0.01	----	----
23	1208	Decanal	0.20 ± 0.01	----	----	----	----	0.28 ± 0.01	----
24	1210	Methyl chavicol	----	----	----	0.29 ± 0.01	0.42 ± 0.02	----	----
25	1211	Z-Piperitol	----	----	0.44 ± 0.02	----	----	----	0.22 ± 0.01

[a]Retention indices.

[b]Compounds listed according to their elution on DB5 column.

[c]Values (average of triplicate determinations) expressed as relative area percentages to total identified compounds.

O + Ca: Orange oil + Camphor; O + Ca + Or: Orange oil + Camphor oil + Oregano oil; O + Ba: Orange oil + Basil oil; O + Ba + Or: Orange oil + Basil oil + Oregano oil

; O + Ma: Orange oil + Marjoram oil; O + Ma + Or: Orange oil + Marjoram oil + Oregano oil

Peak No.	KI[a]	Volatile compounds[b]	Area (%)[c]						
			Orange	O + Ca	O + Ca + Or	O + Ba	O + Ba + Or	O + Ma	O + Ma + Or
26	1253	Carvacrol methyl ether	——	——	0.10 ± 0.01	——	0.09 ± 0.00	——	——
27	1278	β-Terpinyl acetate	——	0.51 ± 0.03	0.18 ± 0.01	——	——	——	——
28	1295	bornyl acetate	——	——	——	0.13 ± 0.01	——	——	——
29	1298	ρ-cymen-2-ol	——	——	——	——	——	——	——
30	1311	Thymol	——	——	0.51 ± 0.03	——	0.16 ± 0.01	——	——
31	1355	Eugenol	——	0.44 ± 0.02	0.20 ± 0.01	——	0.12 ± 0.01	——	——
32	1398	β-Elemene	——	——	——	——	0.04 ± 0.00	——	——
33	1409	β-Caryophyllene	——	1.08 ± 0.05	0.10 ± 0.01	——	0.10 ± 0.01	——	——
34	1438	Z-α-Bergamotene	——	——	——	——	0.04 ± 0.00	——	——
35	1447	α-Humulene	——	——	——	——	0.07 ± 0.00	——	——
36	1462	β-Santalene	——	——	——	——	0.03 ± 0.00	——	——
37	1513	δ-Cadinene	——	——	0.19 ± 0.01	0.13 ± 0.01	0.16 ± 0.01	0.16 ± 0.01	——
38	1598	Cadina-1(10),4-diene	——	1.75 ± 0.09	——	——	——	——	——
Total			100±	98.91±	99.33±	98.38±	99.51±	98.88±	96.70±

[a]Retention indices.

[b]Compounds listed according to their elution on DB5 column.

[c]Values (average of triplicate determinations) expressed as relative area percentages to total identified compounds.

O + Ca: Orange oil + Camphor; O + Ca + Or: Orange oil + Camphor oil + Oregano oil; O + Ba: Orange oil + Basil oil; O + Ba + Or: Orange oil + Basil oil + Oregano oil

; O + Ma: Orange oil + Marjoram oil; O + Ma + Or: Orange oil + Marjoram oil + Oregano oil

D-limonene was the predominant volatile compound in orange oil (94.78%) followed by myrcene (2.35%). The GC-MS analysis of camphor oil revealed that camphor was the major compound (49.99%) followed by β-citronellol (18.59%) and 1-borneol (9.96%) [26]. Thirty three volatile compounds were identified in basil (Ba- EO) among them methyl chavicol (37.65%), linalool (20.98%) and 1,8-cineole (7.54%) were the major compounds [20]. The GC-MS analysis of marjoram (Ma- EO) revealed the presence of 25 volatile compounds, representing 95.44% of the total oil. Terpinen-4-ol (31.23%) was the major identified compound followed by linalool (9.73%) [27]. As shown in Table 2S, the phenolic compound thymol was the major compound (22.27%) in oregano (Or-EO) followed by limonene (20.58%), α-terpineol (11.30%), linalool (7.91%) and thymol methyl ether (7.51%) [28].

3.2. Radical scavenging activity of the essential oils

In the present study DPPH and ABTS radicals assays were used to determine the free radical scavenging activity of orange oil (O) and Ca, Ma, Ba and Or EOs. The radical scavenging activity of both method showed a similar decreasing order such as Ba > Ca > Or > Ma > O (Fig. 1a, b). The highest inhibition of the free radicals DPPH and ABTS by basil EO may be attributed to the presence of linalool and eugenol in considerable concentrations (Table 2S). It was reported that the basil EO of linalool-eugenol chemo type showed high AOA [29]. The synergistic interaction between the antioxidant compounds found in the basil EO such as 1, 8-cineole, ρ-cymene and terpinen-4-ol may have a role to play. The high antioxidant activity of camphor EO may be correlated to the presence of the antioxidant compounds such as camphor [30], 1, 8-cineole, terpinene-4-ol and eugenol. The high scavenging ability of marjoram EO on DPPH and ABTS radicals is mainly correlated to the presence of alcohol terpenes such as terpinen-4-ol (31.23%), linalool (9.73%) and α-terpineol (5.61%). The antioxidant activity of oregano EO is mainly attributed to the presence of the two phenolic compounds thymol (22.27%) and its isomer carvacrol (2.90%) [31], in addition to the synergistic interaction among other antioxidant compounds such as linalool, α-terpeneol, ρ-cymene and 1, 8- cineole (Table 2S)

3.3. Odour sensory evaluation and radical scavenging activity of orange-EOs blends (O-EOs)

Blends of orange oil with Ca, Ba or Ma EOs at different concentrations, 0.25, 0.5 and 0.75 µL /100 µL orange oil, were subjected to odour sensory analysis. As shown in Table 1, the blends containing orange oil and each EO at concentration of 0.25 µL /100 µL orange oil showed the highest scores. Therefore, these blends were selected and subjected to GC-MS analysis and determination of their AOA (Table 2 and Fig. 1c, d). As shown in Fig. 1c, addition of 0.25 µL of each EO (basil, camphor and marjoram) to orange oil increased the inhibition of the free radical DPPH from 19.31% to 57.02, 24.33 and 22.90%, respectively. The scavenging ability of the three blends on ABTS radical showed the same trend as DPPH (Fig. 1d). This finding is correlated to the presence of the antioxidant compounds in the three blends (Table 2). As shown in Fig. 1c, addition of 0.1 µL of oregano essential oil (Or) to the three blends O + Ba, O + Ca and O + Ma significantly ($p < 0.05$) increased the inhibition of free radical DPPH. These results are attributed to the tow phenolic compounds (thymole and carvacrol) which are responsible for the free radical scavenging ability of oregano EO, in additions to other antioxidant compounds (Table 2 and Fig. 1c). As shown from Fig. 1d,

the results of ABTS assay showed the same trend. From Fig. 1c, d and Table 1, O + Ba + Or blend showed the highest scavenging ability (60.79%) and highest odour sensory score, therefore it was encapsulated at different quantities in β-CDs derivatives (F1 and F2) (Table 1S)

3.4. Odour acceptability and encapsulation efficiency of the orange flavoured inclusion complexes

As shown in Table 3, the odour acceptability showed a gradual increase by increasing the amount of O-EOs blend, from 200 to 300 mg, for both inclusion complexes O-F1-IC and O-F2-IC. Increasing the content of O-EOs blend from 200 to 250 showed a significant in increase in the entrapping efficiency EE flowed by a slight decrease by increasing the O-EOs blend to 300 mg. The high EE for all investigated samples may be correlated to the high content of limonene in the O-EOs blend which has high affinity for β-CD encapsulation. The encapsulation of limonene in 2- hydroxypropyl-β-cyclodextrin (2-HP-β-CD) reached 91.8% [32]. As shown in Table 5, O-F1-IC showed higher EE than O-F2-IC. This finding may be correlated to the fact that increasing the concentration of CDs resulted in increasing the spaces occupied by CDs polymers and causing a decrease of the free volume within the polymer matrix (a compact structure with smaller pore sizes) and subsequently the amount of the oil that can be encapsulated within those pores will be decreased [33]. Previous studies revealed that the physicochemical properties of both the CDs polymers (cavity diameter, derivative nature) and the encapsulated guest (geometry, volume and hydrophobicity) play a crucial role in the formation of inclusion complexes [6, 34]. The higher EE in the present study may be attributed to the freeze drying process used in preparation of the inclusion complexes, which prevent the degradation and evaporation of the volatile compounds and thus minimizes their loss [35].

Table 3
Odour acceptability, encapsulation efficiency of the inclusion complexes and radical scavenging activity of encapsulated O-EOs blend.

Inclusion complexes	Odour acceptability	encapsulation efficiency %	radical scavenging activity %
O-F1-IC			
F1A	7.3 ± 0.37^{c}	92.80 ± 0.92^{b}	53.60 ± 0.53^{ce}
F1B	6.2 ± 0.31^{d}	93.65 ± 0.95^{b}	55.00 ± 0.55^{b}
F1C	9.0 ± 0.50^{a}	95.51 ± 0.94^{a}	58.51 ± 0.60^{a}
O-F2-IC			
F2A	4.0 ± 0.22^{e}	91.00 ± 0.90^{c}	51.11 ± 0.50^{d}
F2B	8.0 ± 0.44^{b}	92.10 ± 0.91^{bc}	53.30 ± 0.53^{c}
F2C	8.3 ± 0.45^{b}	$92 .80 \pm 0.92^{b}$	54.53 ± 0.54^{be}
O-F1-IC, O-F2-IC: orange flavoured inclusion complexes			
F1 = 2- HP-β-CD: EPI-β-CD at molar ratio 3: 1; F2 = 2- HP-β-CD: EPI-β-CD at molar ratio 1: 3			
For each column, same letters means insignificant difference at $p < 0.05$			

3.5. Antioxidant activity of the encapsulated O-EOs blend

The antioxidant activity of the encapsulated O-EOs in the inclusion complexes (Table 3) was lower than that of the free O-EOs blend (Fig. 1c). This decrease could be because the cyclodextrine polymers block the functional groups of the active compounds that react with DPPH radical [36], bearing in mind that the cyclodextrine polymers didn't have antioxidant activity. A positive correlation was found between the DPPH scavenging activity and encapsulation efficiency [11]. This result is in agreement with previous studies. The guava life oil encapsulated in 2-HP-β-CD showed slightly lower DPPH scavenging activity than that of the free guava oil [37]. Similar results have been reported by Rakmai et al. [11] who correlated the antioxidant activity of black pepper oil encapsulated in HP-β-CD to the presence of the antioxidant compounds limonene and α-pinene.

3.6. Characterization of the orange flavoured inclusion complexes

As shown in Table 3 inclusion complexes O-F1C-IC, O-F2B-IC and O-F2C-IC showed the best results regarding, odour acceptability, EE and DPPH scavenging activity, therefore were selected to be characterized.

3.6.1. Particle size, polydispersity and zeta potential

As shown in Table 4, the particle sizes of three investigated inclusion complexes were in the nanoparticle scale [38]. Among these samples, O-F1C-IC showed the smallest particle size and highest EE (Table 3 and Table 4). This result is consistent with previous study of Hadian et al. [34] who reported an opposite relationship between EE and size of nanoparticle of β-cyclodextrin containing geraniol. The higher particle sizes of O-F2B-IC and O-F2C-IC than O-F1C-IC may be correlated to their higher content of EPI-β-CD which has a higher molecular weight than 2-HP-β-CD. It has been reported that increases in β-CD concentration induce aggregation and results in increasing the particle size [38–40]. Polydispersity index (PDI) of the inclusion complex nanoparticles was < 0.5. This indicates their stability and homogeneity [34]. The zeta potential of the nanoparticles is considered one of their major characteristics that may be affecting their stability. It is an indicator of the charge presence on the surface of the nanoparticle. The values of negative zeta potential on the investigated nanoparticles ranged from − 16 to -27mV, however, O-F1C-IC showed the more negative zeta potential − 27mV. The high dispersion stability is possible when the zeta potential value is near − 30 mV [41]. The reduced negative charge in O-F2B-IC and O-F2C-IC may be attributed to the lower entrapped O-EOs blend (guest) with higher β-CDs concentration [34].

Table 4
Particle size (PS), zeta potential (ZP) and polydispersity index (PDI) for the selected inclusion complexes.

Inclusion complexes	Particle size (nm)	Zeta potential (mV)	PDI
O-F1C-IC	113.9 ± 15.9[a]	-27.1 ± 1.27[a]	0.334[a]
O-F2B-IC	374.05 ± 20.44[b]	-17.7 ± 1.13[b]	0.399[a]
O-F2C-IC	329.25 ± 7.99[c]	-16.55 ± 0.21[b]	0.398[a]
O-F1-IC, O-F2-IC: orange flavoured inclusion complexes			
F1 = 2-HP-β-CD:EPI-β-CD at molar ratio 3:1; F2 = 2-HP-β-CD:EPI-β-CD at molar ratio 1:3			
For each column, same letters means insignificant difference at $p < 0.05$			

Table 5
Formulation of effervescent powders.

Formula	CA (gm)	TA (gm)	$NaHCO_3$ (gm)	Effervescence time (s)	Clarity	pH
1	0.06	0.12	0.21	19 ± 1.11 [a]	Very clear	6.00 ± 0.01 [a]
2	0.12	0.24	0.42	40 ± 2.75 [b]	Very clear	6.01 ± 0.02 [a]
3	0.24	0.48	0.84	60 ± 3.80 [c]	Very clear	6.03 ± 0.03 [a]
4	0.48	0.96	1.68	105 ± 5.00 [d]	Very clear	6.30 ± 0.30 [a]
5	0.96	1.92	3.36	215 ± 12.32 [e]	Very clear	6.15 ± 0.03 [a]

For each parameter, same letter means insignificant difference at $p < 0.05$. CA = Citric acid; TA = Tartaric acid

3.6.2. Morphology of the orange flavoured inclusion complexes

The particle morphology of the three orange effervescent inclusion complexes was observed under a scanning electron microscope (SEM). As shown in Fig. 2, all samples showed irregular shaped particles. The highly porous nature of the sample confirms their high encapsulation efficiency (Table 3) of the O-EOs blend [42]. Sample O-F1C-IC showed smaller particle size than O-F2B-IC and O-F2C-IC, which consistent with results in Table 4.

3.6.3. Fourier transform infrared spectroscopy analysis (FT-IR) of the inclusion complexes

FT-IR technique was used to investigate the variation of shape, intensity and position of peaks IR absorption peaks of the guest or the host can provide information about the occurrence of the inclusion complex formation. FT-IR spectrum of 2-HP-β-CD showed characteristic bands belonging to saccharides: 3341.47 cm^{-1} (O-H stretching vibration), 2924.67 cm^{-1} (C-H stretching vibration), 1645.39 cm^{-1} (O-H bending vibration), and 1151.38 cm^{-1} (C-O vibration). The bands in the range of 1330.95-1456.76 cm^{-1} were assigned to CH2 and CH3 bending vibrations [42, 43]. The similarity between the spectra of 2-HP-β-CD and EPI-β-CD indicates that the basic structural units of cyclodextrin polymer are preserved in both of them [44, 45]. As shown in Fig. 3, the O-H stretching band showed a remarkable lower frequency in F1 (HP-β-CD: EIP-β-CD, 3: 1 molar ratio) and F2 (HP-β-CD: EIP-β-CD, 1: 3 molar ratio) than their individual components 3341.47 cm^{-1}. This result may be correlated to the formation of aggregates between the two β-CD derivatives when dissolved in water [38]. The main peaks of the selected blend (O + Ba + Or) was observed at 2964, 2917, 1680, 1440, 1375, 886, 797 cm^{-1} (Fig. 3). The peak around 2967 cm^{-1} corresponds to the CH_3 stretching vibration. The peaks around 2917, 1680, 1440, 886, 797 cm^{-1} corresponding to C-H stretching vibration of alkanes, C = O stretching vibration, C = C stretching vibration of alkanes and C = H bending vibration of alkanes, CH-stretching vibration of aromatic and C = C bending vibration of alkanes. These results are in

agreement with [46]. As shown in Fig. 4 the bands of O-EOs spectrum were obscured by the F1 and F2 bands. This result indicates the successful entrapment of the O-EOs blend and formation of the inclusion complexes [11, 37]. In addition, the O-H stretching band in O-F1C-IC was found to be shifted to a lower frequency (3268.90 cm^{-1}) than its blank F1 (3313.27 cm^{-1}). The same trend was found for O-F2B-IC and O-F2C-IC which showed lower frequencies (3290.12 and 3241.19 cm^{-1}, respectively) compared to their blank F2 (3309.47 cm^{-1}). This finding may be correlated to the intermolecular interaction between the O-EO blend and the CDs polymers (F1 and F2) within the inclusion complexes. Similar results were reported by previous studies [6, 11, 37, 47].

3.7. Evaluation of effervescent powder

The results revealed that increasing the amount of effervescent components resulted in increasing the effervescent time (Table 5). Formula 5, showed the long effervescent time (215s), however it was excluded according to the pharmacopeia standard which stated that effervescent time should be < 180s. Moreover, the presence of high amount of citric acid (CA) in the effervescent powder may displace the encapsulated compound from the cyclodextrin cavity and consequently decrease its solubility [25, 48] and hence its amount in the end product. As shown in Table 5, all formula showed a slight acidic, and thus have better palatability [49]. As shown in Table 5, formula 4 showed the best results and thus was selected for the preparation of the orange flavoured effervescent powder.

3.7.1. Physical properties of the orange flavoured effervescent powder

The flowability of the effervescent powder is mainly evaluated by different parameters such as bulk density, tapped density, Carr's compressibility index, Hausner ratio and angle of repose [50, 51].The bulk density and tapped density of the prepared orange flavoured effervescent powder were 0.29 ± 0.01 and 0.35 ± 0.01 g/mL, respectively. It has been reported that a powder with Carr's compressibility index and Hausner ratio lower than 20% and 1.25, respectively is considered to be free flowable [52]. Accordingly, the values of Carr's compressibility index and Hausner ratio in the present study (16.53 ± 0.75 and 1.20 ± 0.01, respectively) indicated good flow properties for the orange flavoured effervescent powder (Table 5). Concerning the angle of repose, the measured value, 34° ± 0.63, also reflected good powder flowability according to the previous study where free flowing powder was imparted when angle of repose ranged between 30–38° [51]. Thus, the evaluated parameters (Table 3S) indicated better packing ability and free flowability of the effervescent powder.

4. Conclusion

Currently, there is an increasing interest to impart the carbonated beverages functional properties. Orange is one of the most popular beverages. In present study, the antioxidant activity of orange oil was improved by blending with different essential oils having high antioxidant activity, while keeping the desired flavour. Combinations of two soluble β-cyclodextrine derivatives, 2-hydroxylpropyl-β-cyclodextrine and epihydrin-β-cyclodextrine, at 3: 1 and 1: 3 molar ratios, respectively, were used to encapsulate the orange essential oils

blend (O-EOs) that showed the highest odour quality and antioxidant activity. The physicochemical analysis, scanning electron microscope (SEM) and Fourier transform infrared spectroscopy (FT-IR) confirmed the successful formation of the orange flavoured inclusion complexes nanoparticles. The slight decrease in the antioxidant activity of the encapsulated O-EOs blend compared to the free blend may be correlated to blocking of the functional groups of the active compounds that react with DPPH radical. Among the investigated inclusion complexes that produced by encapsulation of O-EOs with the mixture of 2-HP-β-CD and EPI-β-CD (at molar ratio 3:1) showed the smallest particle size, the best odour quality, the highest antioxidant activity, the highest encapsulation efficiency and the more negative zeta potential. Therefore, it was used in preparation of the orange flavoured effervescent powder, the physical analysis proved its high quality.

Declarations

Appendix A. Supplementary data Supplementary data to this article can be found at online

References

1. Taymouri, S., Mostafavi, A., Javanmardi, M.: Formulation and optimization of effervescent tablet containing bismuth sub-citrate. J. Rep. Pharma. Sci. **8**, 236-244 (2019). https://doi.org/10.4103/jrptps.JRPTPS_11_19.
2. Persistence market research. Effervescent Products Market Outlook (2022 - 2029). **Effervescent Products Market - Global Industry Analysis, Size, Share, Demand, Sales and Trends by 2020, (2022). (persistencemarketresearch.com)**
3. Ipci, K., Öktemer, T., Birdane, L., Altintoprak, N., Muluk, N.B., Passali, D., Lopatin, A., Belluss, L., Mladina R., Pawankar, R., Cingi, C.: Effervescent tablets: a safe and practical delivery system for drug administration. ENT Updates **6**, 46-50 (2016). https://doi.org/10.2399/jmu.2016001009.
4. Kregiel, D.: Health safety of soft drinks: contents, containers, and microorganisms. Biomed. Res. Int. **2015**,128697 (2015). https://doi.org/10.1155/2015/128697.
5. Yashin, A., Yashin, Y., Xia, X., Nemzer, B.: Antioxidant Activity of Spices and Their Impact on Human Health: A Review. Antioxidants (Basel) **6**, 70 (2017). https://doi.org/10.3390/antiox6030070.
6. Kfoury, M., Balan, R., Landy, D., Nistor, D., Fourmention, S.: Investigation of the complexation of essential oil components with cyclodextrins. Supramol. Chem. **27**, 620-628 (2015). https://doi.org/10.1080/10610278.2015.1051977.
7. Fang, Z., Bhandari, B.: Encapsulation of polyphenols – a review. Trends Food Sci. Technol. **21**, 510-523 (2010). https://doi.org/10.1016/j.tifs.2010.08.003.
8. Pinho, E., Grootveld, M., Soares, G., Henriques, M.: Cyclodextrins as encapsulation agents for plant bioactive compounds. Carbohydr. Polym. **101**, 121–135 (2014). https://doi.org/10.1016/j.carbpol.2013.08.078.

9. Szejtli, J:. Introduction and general overview of cyclodextrin chemistry. Chem. Rev. **98**, 1743–1754 (1998). https://doi.org/10.1021/cr970022c.
10. Waleczek, K.J., Cabral, M.H.M., Hempel, B., Schmidt, P.C.: Phase solubility studies of pure (-) alpha-bisabolol and chamomile essential oil with beta–cyclodextrin. Eur. J. Pharm. Biopharm. **55**, 247–251 (2003). https://doi.org/10.1016/S0939-6411(02)00166-2
11. Rakmai, J., Cheirsilp, B., Mejuto, J.C., Torrado-Agrasar, A., Simal-Gándara, J.: Physico-chemical characterization and evaluation of bio-efficacies of black pepper essential oil encapsulated in hydroxypropyl-beta-cyclodextrin. Food Hydrocol. **65**, 157-164 (2016). https://doi.org/10.1016/j.foodhyd.2016.11.014.
12. Garnero, C., Zoppi, A., Genovese, D., Longhi, M.: Studies on trimethoprim: hydroxypropyl-β-cyclodextrin: aggregate and complex formation. Carbohydr. Res. **345**, 2550-2556 (2010). https://doi.org/10.1016/j.carres.2010.08.018.
13. Gidwani, B., Vyas, A.: Synthesis, characterization and application of epichlorohydrin-β-cyclodextrin polymer. Colloids Surf. B. **114**, 130-137 (2014). https://doi.org/10.1016/j.colsurfb.2013.09.035.
14. Wintgens, V., Amiel C.: Water-soluble γ-cyclodextrin polymers with high molecular weight and their complex forming properties. Eur. Polym. J. **46**, 1915-1922 (2010). https://doi.org/10.1016/j.eurpolymj.2010.06.014.
15. Bai, M-Y., Zhou, Q., Zhang, J., Li, T., Cheng, J., Liu, Q., Xu, W-R., Zhang, Y-C.: Antioxidant and antibacterial properties of essential oils-loaded β-cyclodextrin-epichlorohydrin oligomer and chitosan composite films. Colloids Surf. B. **215**, 112504 (2022). https://doi.org/10.1016/j.colsurfb.2022.112504.
16. Mura, P., Faucci, M.T., Maestrelli, F., Furlanetto, S., Pinzauti, S.: Characterization of physicochemical properties of naproxen systems with amorphous β-cyclodextrin-epichlorohydrin polymers. J. Pharm. Biomed. **29**, 1015-1024 (2002). https://doi.org/10.1016/S0731-7085(02)00142-5.
17. Yen, G-C., Chen, H-Y.: Antioxidant activity of various tea extracts in relation to their antimutagenicity. J. Agric. Food Chem. **43**, 27–32 (1995). https://doi.org/10.1021/jf00049a007.
18. Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice- Evans, C.: Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic. Biol. Med. **26**, 1231–1237 (1999). https://doi.org/10.1016/s0891-5849(98)00315-3.
19. Arnao, M.B.: Some methodological problems in the determination of antioxidant activity using chromogen radicals: a practical case. Trends Food Sci. Technol. **11**, 419–421 (2000). https://doi.org/10.1016/S0924-2244(01)00027-9.
20. Fadel, H.H.M., El-Ghorab, A.H., Hussein, A.M.S., El-Massry, K.F., Lotfy, S.N., Ahmed, M.Y.S., Soliman, T.N.: Correlation between chemical composition and radical scavenging activity of 10 commercial essential oils: Impact of microencapsulation on functional properties of essential oils. Arab J. Chem. **13**, 6815–6827 (2020). https://doi.org/10.1016/j.arabjc.2020.06.034.
21. Adams, R.P.: Identification of essential oil components by gas chromatography/quadruple mass spectroscopy. Allured Publishing Corporation, Carol Stream, IL (2001).
22. Cutrignelli, A., Sanarica, F., Lopalco, A., Lopedota, A., Laquintana, V., Franco, M., Boccanegra, B., Mantuano, P., De Luca, A., Denora, N.: Dasatinib/HP-β-CD inclusion complex based aqueous

formulation as a promising tool for the treatment of paediatric neuromuscular disorders. Int. J. Mol. Sci. **20**, 591 (2019). https://doi.org/10.3390/ijms20030591.

23. Soliman, E.A., El-Moghazy, A.Y., Mohy El-Din, M.S., Massoud, M. A.: Microencapsulation of essential oils within alginate: formulation and in *vitro* evaluation of antifungal activity. J. Encapsulation Adsorpt. Sci. **3**, 48–55 (2013). https://doi.org/10.4236/jeas.2013.31006.
24. Ashutosh, M., Rajesh, K., Mukesh, C.: Formulation, development and evaluation of patient friendly dosage forms of metformin, part-III: soluble effervescent tablets. Asian J. Pharm. **2**, 177-181 (2008). https://doi.org/10.22377/ajp.v2i3.206.
25. Hussain, H.A.M., Al-Khedairy, E.B.H.: Preparation and in vitro evaluation of cyclodextrin based effervescent and dispersible granules of carbamazepine. Int. J. Appl. Pharm. **10**, 290-297 (2018). https://doi.org/10.22159/ijap.2018v10i6.28276.
26. Guo, S., Geng, Z., Zhang, W., Liang, J., Wang, C., Deng, Z., Du, S.: The Chemical Composition of Essential Oils from *Cinnamomum camphora* and Their Insecticidal Activity against the Stored Product Pests. Int. J. Mol. Sci. **17**, 1836 (2016). https://doi.org/10.3390/ijms17111836.
27. Gharib, F.A.E, Teixeira da Silva, J.A.: Composition of total phenolic content and antioxidant activity of the essential oil of four Lamiaceae herbs. Med. Aromat. Plant Sci. Biotechnol. **7**, 19-27 (2012).
28. Sedlaříková, J., Doležalová, M., Egner, P., Pavlačková, J., Krejčí, J., Rudolf, O., Peer, P.: Effect of Oregano and Marjoram Essential Oils on the Physical and Antimicrobial Properties of Chitosan Based Systems. Int. J. Polym. Sci. **2017**, 2593863 (2017). https://doi.org/10.1155/2017/2593863.
29. Pripdeevech, P., Chumpolsri, W., Suttiarporn, P., Mahatheeranont, S.: The chemical composition and antioxidant activities of basil from Thailand using retention indices and comprehensive two-dimensional gas chromatography. J. Serb. Chem. Soc. **75**, 1503-1513 (2010). https://doi.org/10.2298/JSC100203125P.
30. Bouzidi, N., Mederbal, K., Raho, B.: Antioxidant Activity of Essential Oil of *Artemisia herba alba*. J. Appl. Environ. Biol. Sci. **6**, 59-65 (2016).
31. Kulisic, T., Radonic, A., Katalinic, V., Milos, M.: Use of different methods for testing antioxidative activity of oregano essential oil. Food Chem. **85**, 633–640 (2004). https://doi.org/10.1016/j.foodchem.2003.07.024.
32. Hill, L.E., Gomes, C.L., Taylor, T.M.: Characterization of beta-cyclodextrin inclusion complexes containing essential oils (trans-cinnamaldehyde, eugenol, cinnamon bark, and clove bud extracts) for antimicrobial delivery applications. LWT - Food Sci. Technol. **51**, 86-93 (2013). https://doi.org/10.1016/j.lwt.2012.11.011.
33. Sevda, S.B., Rodrigues, L.: The making of pomegranate wine using yeast immobilized on sodium alginate. Afr. J. Food Sci. **5**, 299-304 (2011).
34. Hadian, Z., Maleki, M., Abdi, K., Atyabi, F., Mohammadi, A., Khaksar, R.: Preparation and characterization of nanoparticle β-Cyclodextrin: Geraniol inclusion complexes. Iran J. Pharm. Sci. **17**, 39- 51 (2018).
35. Del Valle, E.M.M.: Cyclodextrins and their uses: a review. *Process Biochem.* **39**, 1033–1046 (2004). https://doi.org/10.1016/S0032-9592(03)00258-9.

36. Kamimura. J.A., Santos, E.H., Hill, L.E., Gomes, C.L.: Antimicrobial and antioxidant activities of carvacrol microencapsulated in hydroxypropyl-beta-cyclodextrin. LWT - Food Sci. Technol. **57**, 701–709 (2014). https://doi.org/10.1016/j.lwt.2014.02.014.
37. Rakmai, J., Cheirsilp, B., Mejuto, J.C., Simal-Gándara, J., Torrado-Agrasar, A.: Antioxidant and antimicrobial properties of encapsulated guava leaf oil in hydroxypropyl-beta-cyclodextrin. Ind. Crops Prod. **111**, 219-225 (2018). https://doi.org/10.1016/j.indcrop.2017.10.027.
38. Loftsson, T., Saokham, P., Couto, A.R.S.: Self-association of cyclodextrins and cyclodextrin complexes in aqueous solutions. Int. J. Pharm. **560**, 228-234 (2019).https://doi.org/10.1016/j.ijpharm.2019.02.004.
39. Hernández-Sánchez, P., López-Miranda, S., Guardiola, L., Serrano-Martínez, A., Gabaldón, J.A., Nuñez-Delicado, E.: Optimization of a method for preparing solid complexes of essential clove oil with β-cyclodextrins. J. Sci. Food Agric. **97**, 420-426 (2016). https://doi.org/10.1002/jsfa.7781.
40. Han, Y., Jia, f., Bai, S., Xiao, Y., Meng, X., Jiang, L.: Effect of operating conditions on size of catechin/β-cyclodextrin nanoparticles prepared by nanoprecipitation and characterization of their physicochemical properties. LWT - Food Sci. Technol. **153**, 112447 (2022). https://doi.org/10.1016/j.lwt.2021.112447.
41. Sahari, M.A., Moghimi, H.R., Hadian, Z., Barzegar, M., Mohammadi, A.: Improved physical stability of docosahexaenoic acid and eicosapentaenoic acid encapsulated using nanoliposome containing α-tocopherol. Int. J. Food Sci. **51**, 1075-1086 (2016). https://doi.org/10.1111/ijfs.13068.
42. Yaşayan, G., Şatıroğlu Sert, B., Tatar, E., Küçükgüzel, İ.: Fabrication and characterisation studies of cyclodextrin-based nanosponges for sulfamethoxazole delivery. J. Incl. Phenom. Macrocycl. Chem. **97**, 175–186 (2020). https://doi.org/10.1007/s10847-020-01003-z.
43. Yuan, C., Liu, B., Liu, H.: Characterization of hydroxypropyl-β-cyclodextrins with different substitution patterns via FTIR, GC-MS, and TG-DTA. Carbohydr. Polym. **118**, 36-40 (2015). https://doi.org/10.1016/j.carbpol.2014.10.070.
44. Poornima, K.N., Deveswaran, R., Bharath, S., BasavaRaj, B.V., Madhavan, V.: Synthesis and evaluation of β-cyclodextrin-epichlorohydrin inclusion complex as a pharmaceutical excipient. J. Fundam. Appl. Sci. **7**, 203-221 (2015). https://doi.org/10.4314/jfas.v7i2.5.
45. Li, R., Yu, J.C., Jiang, Z-T., Zhou, R-H., Liu, H-Y.: A solid-phase fluorescent quenching method for the determination of trace amounts of nitrite in foods with neutral red. J. Food Drug Anal. **11**, 251-257 (2003). https://doi.org/10.38212/2224-6614.2700.
46. Cebi, N., Taylan, O., Abusurrah, M., Sagdic, O.: Detection of Orange Essential Oil, Isopropyl Myristate, and Benzyl Alcohol in Lemon Essential Oil by FTIR Spectroscopy Combined with Chemometrics. Foods **10**, 27 (2021). https://doi.org/10:1-14. 10.3390/foods10010027.
47. Wang, X., Luo, Z., Xiao, Z.: Preparation, characterization, and thermal stability of β-cyclodextrin/soybean lecithin inclusion complex. Carbohydr. Polym. **101**, 1027-1032 (2014). https://doi.org/10.1016/j.carbpol.2013.10.042.
48. Stella, V.J., Rao, V.M., Zannou, E.A., Zia, V.: Mechanisms of drug release from cyclodextrin complexes. Adv. Drug Deliv. Rev. **36**, 3-16 (1999). https://doi.org/10.1016/s0169-409x(98)00052-0.

49. Gould, S., Scott, R.C.: 2-Hydroxypropyl-beta-cyclodextrin (HP-beta-CD): a toxicology review. Food Chem. Toxicol. **43**, 1451-1459 (2005). https://doi.org/10.1016/j.fct.2005.03.007.
50. Al-Okbi, S.Y., Mohamed, D.A., Hamed, T.E., Abd El-Alim, S.H., Kassem, A.A., Mostafa, D.M.: Application of liquisolid technology for promoting the renoprotective efficacy of walnut extracts in chronic renal failure rat model. Drug Dev. Ind. Pharm. **45**, 32-42 (2019). https://doi.org/10.1080/03639045.2018.1515219.
51. Beakawi Al-Hashemi, H.M., Baghabra Al-Amoudi, O.S.: A review on the angle of repose of granular materials. Powder Technol. **330**, 397-417 (2018). https://doi.org/10.1016/j.powtec.2018.02.003.
52. Sarraguça, M.C., Cruz, A.V., Soares, S.O., Amaral, H.R., Costa, P.C., Lopes, J.A.: Determination of flow properties of pharmaceutical powders by near infrared spectroscopy. J. Pharm. Biomed. Anal. **52**, 484-492 (2010). https://doi.org/10.1016/j.jpba.2010.01.038.

Figures

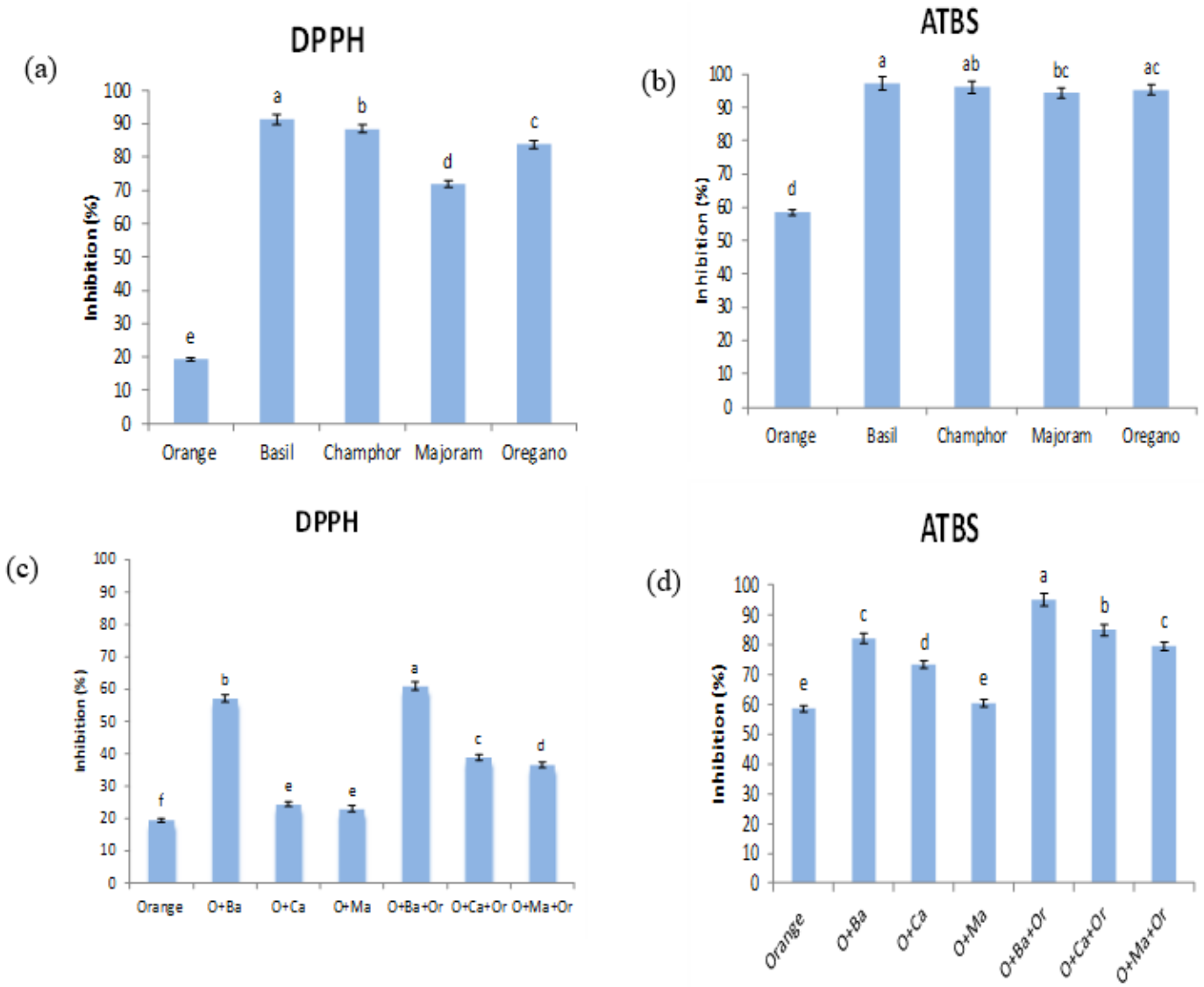

Figure 1

Radical scavenging activity of orange oil, selected EOs and orange blends with essential.

Ba: basil oil, Ca: camphor oil, Ma: marjoram, Or: oregano oil, same letter means insignificant difference at $p < 0.05$.

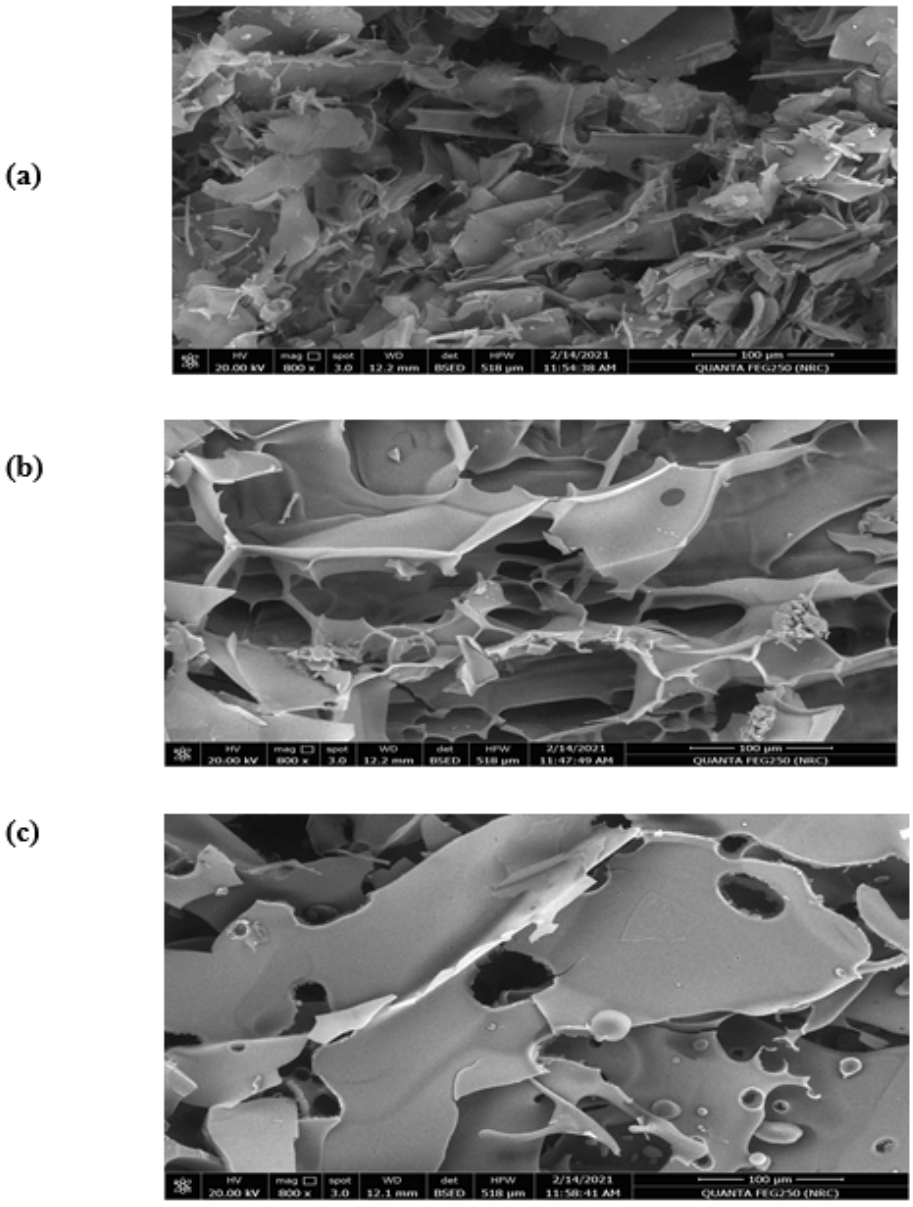

Figure 2

SEM micrographs of the orange flavoured inclusion complexes.

a: O-F1C-IC; b: O-F2B-IC; c: O-F2 C-IC; F1= 2-Hydroxypropyl-β-cyclodextrin: Epichlorohydrin-β-cyclodextrin (2-HP-β-CD: EPI-β-CD at molar ratio 3: 1); F2=2-Hydroxypropyl-β-cyclodextrin: Epichlorohydrin-β-cyclodextrin (2-HP-β-CD: EPI-β-CD at molar ratio 1: 3).

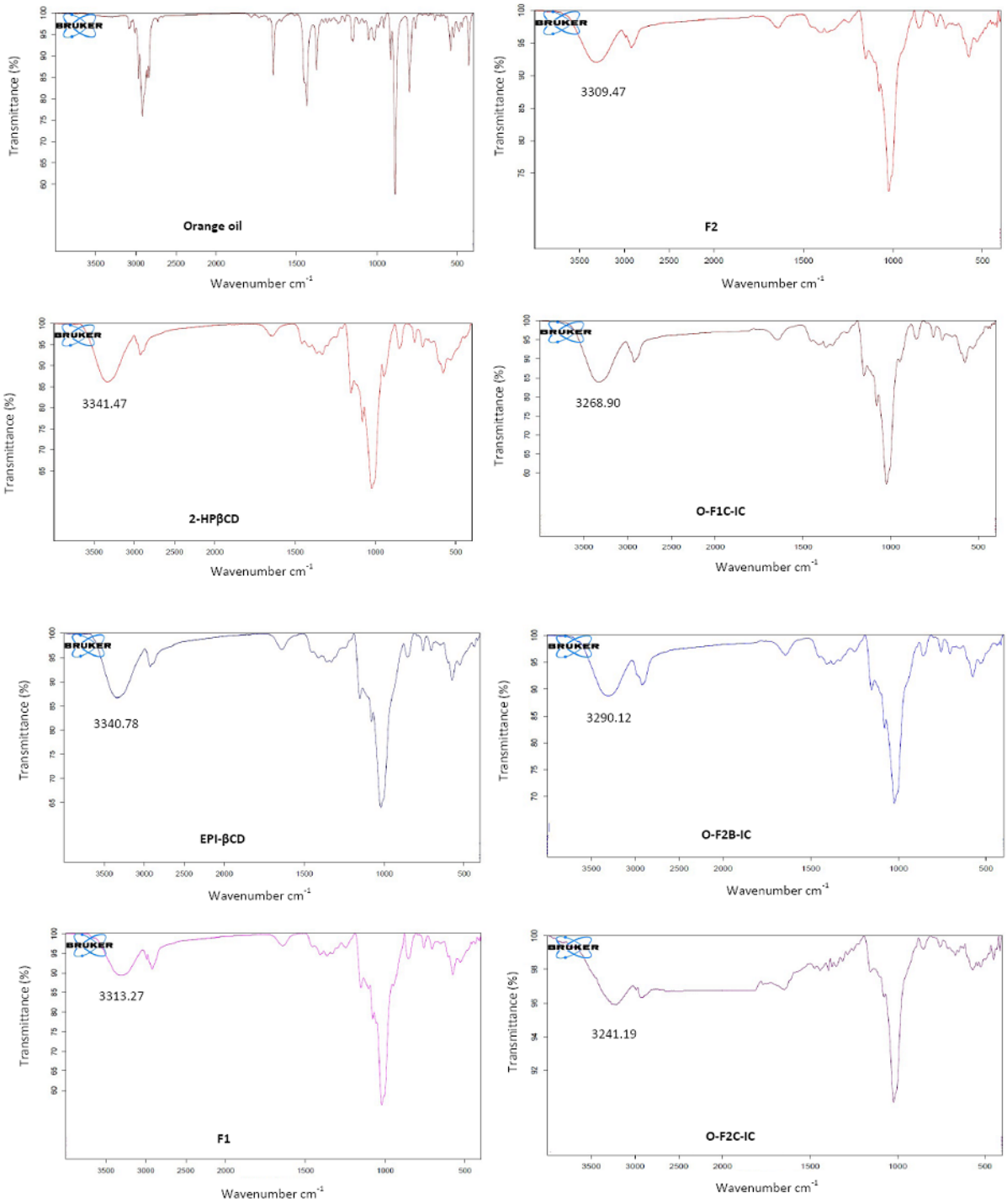

Figure 3

FT-IR spectra of O-EOs blend, F1, F2 and the inclusion complexes (O-F1C-IC, O-F2B-IC and O-F2C-IC).

O-F1C-IC, O-F2B-IC and O-F2C-IC: orange flavoured inclusion complexes; F1= 2-HP-β-CD: EPI-β-CD at molar ratio 3: 1; F2= 2-HP-β-CD: EPI-β-CD at molar ratio 1: 3; 2-HP-β-CD = 2-Hydroxypropyl-β-cyclodextrin: EPI-β-CD = Epichlorohydrin-β-cyclodextrin.

Supplementary Files

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- supplementatrytable.docx
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