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Title: Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (Portulaca oleracea L.) grown in different soilless media in a closed system

Article Type: Research Paper

Section/Category: Bio-based Materials from Crops

Keywords: Purslane; soilless cultivation substrates; yield; proximate composition; phenolic compounds; antioxidant activity.

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Abstract:

Purslane (Portulaca oleracea L.) is a valuable plant and crop with potential industrial uses, yet little is known on how its cultivation could benefit from soilless substrates. This study aimed to assess the effects of different soilless growth media on herbal yields (fresh and dry), proximate chemical composition, total phenolic, flavonoid and anthocyanin content, and antioxidant activity of purslane cultivated in a closed system. The greatest yields over five harvest cycles were obtained with tuff-peatmoss (2:1 mixture) compared with other soilless substrates, although the edible leaves were not as rich in proteins, lipids, minerals, and phenolic compounds. The highest content of proteins (31.4% and 30.4%), lipids (0.68% and 0.75%), total phenolics (646.9 and 684.9 mg/100 g), flavonoids (597.8 and 563.8 mg/100 g), and moisture (92.5% and 93.5%) in the leaves were found in purslane grown in tuff-peatmossperlite (2:1:1) and in zeolitic tuff, respectively. Antioxidant activity of leaf extracts was also the highest in purslane grown in both substrates and was similar to the antioxidant activity of leaf extracts from soil-grown purslane obtained commercially and from the wild. The protein and lipid content obtained with tuff-peatmoss-perlite (2:1:1) and zeolitic tuff were superior to those of soil-grown purslane. The results show that the nutritive and antioxidant qualities of purslane can be enhanced through soilless cultivation and selection of suitable culture media.

July 2019

Opender Koul, PhD Editor-in-Chief, Industrial Crops and Products

Dear Dr. Koul,

Revised manuscript INDCRO-D-19-00886R2

Original Article – "Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (*Portulaca oleracea* L.) grown in different soilless media in a closed system"

M.H. Alu'datt^{1*}, T. Rababah¹, M.N. Alhamad², A. Al-Tawaha³, A.R. Al-Tawaha⁴, K.I. Ereifej¹, G. Al-Karaki³, C.C. Tranchant⁵, S. Kubow⁶. ^{1,2,3}Jordan University of Science and Technology, Jordan; ⁴Al-Hussein Bin Talal University; ⁵Université de Moncton, Canada; ⁶McGill University, Canada. *Corresponding author.

Please receive the revised version of the aforementioned manuscript in which we addressed all the comments and suggestions received. A point-by-point response to reviewers' and editors' comments is also provided in a separate file.

Our paper is the first to report on the yield, proximate composition, total phenolic, flavonoid and anthocyanin contents, and antioxidant activity of purslane (*Portulaca oleracea* L.) grown in different soilless media. The findings have applications for the agricultural production of purslane and should thus be of interest to the readership of *Industrial Crops and Products*.

We thank you for your consideration. We appreciate the opportunity to improve our manuscript and hope that it will be acceptable for publication in your journal.

Please let me know if you have any queries. We look forward to hearing from you.

Best regards,

Dr. Muhammad H. Alu'datt Faculty of Agriculture, Department of Nutrition and Food Technology Jordan University of Science and Technology P.O. Box 3030, Irbid 22110, Jordan E-mail: <u>muhammad.aludatt@mail.mcgill.ca</u>; <u>malodat@just.edu.jo</u>

Files submitted:

Cover letter; Revised manuscript INDCRO-D-19-00886R2; Revised tables and figure; Revised highlights; Authors response to the comments from Reviewers 1 and 2 and Editors.

Manuscript INDCRO-D-19-00886R2 (July 2019)

Revised title: "Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (*Portulaca oleracea* L.) grown in different soilless media in a closed system"

Initial title: "Herbal yield, nutritive composition, phenolic content and antioxidant activity of purslane (*Portulaca oleracea* L.) are influenced by soilless growth medium in a closed system"

Response to the comments from Reviewer 1

Dear Reviewer,

Thank you kindly for your thorough assessment of our work. Your constructive comments are appreciated.

Best regards

	Reviewer's Comment/Authors' response
Comment	The paper entitled: "Herbal yield, nutritive composition, phenolic content and antioxidant activity of purslane (Portulaca oleracea L.) are influenced by soilless growth medium in a closed system" does not present major errors or inconsistencies in its current state.
Response	Thank you for your appreciation of the manuscript.

Manuscript INDCRO-D-19-00886R2 (July 2019)

Revised title: "Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (*Portulaca oleracea* L.) grown in different soilless media in a closed system"

Initial title: "Herbal yield, nutritive composition, phenolic content and antioxidant activity of purslane (*Portulaca oleracea* L.) are influenced by soilless growth medium in a closed system"

Response to the comments from Reviewer 2

Dear Reviewer,

Thank you kindly for your thorough assessment of our work. Your constructive comments are appreciated. They have all been taken into account to improve the manuscript as indicated below. Red font is used to highlight the revisions in the manuscript.

Best regards

	Reviewer's Comment/Authors' response
Comment	Herbal yield, nutritive composition, phenolic content and antioxidant activity of purslane (Portulaca oleracea L.) are influenced by soilless growth medium in a closed system: In general the article is interesting, well written, with future application.
Response	Thank you for your appreciation of the manuscript.
Comment	1. Introduction: Several old references. Update as possible.
Response	The references have been updated whenever possible, with the addition of recent ones, including Rouphael and Kyriacou (2018) , Kopsell et al. (2016) , Alam et al. (2014) , and Uddin et al. (2012) . As noted in the introduction, there is a dearth of information on the impact of soilless culture and substrate on the yield and composition of purslane. Recent reports are even scarcer, which limits the number of recent papers that could be included.
Comment	 2. Materials and Methods: 2.2. Line 106 and 107 - "Cultivation started at the end of February; harvesting began in April and continued until July": Please, insert more details of the harvest, how was it made? Remove the leaves? Close to the root?
Response	Details about the harvesting have been added as suggested (section 2.2.1). The above-ground biomass, which consisted of stems and leaves, was harvested manually by cutting the stems at 5 cm above the level of the soilless substrates.
Comment	2.6. Determination of antioxidant activity using the DPPH assay - Line 186: Why only one method was used? Using other methods, the antioxidant is better evaluated.
Response	The DPPH scavenging assay was used as it is a well-established, simple and reliable method for determining the antioxidant activity of extracts or compounds from plant materials. Our objective in this study was not to compare different methods for assessing antioxidant activity. While we agree that the use of multiple methods would be valuable, this was outside the scope of the study. Suggestions for further research on the antioxidant capacity of purslane products were added to the discussion (section 3.5) to address your comment.
Comment	3. Results and Discussion: Paragraph 252: This paragraph is confuse. Please rewrite the sentence.
Response	This sentence was rephrased as suggested: "This creates suitable conditions for root growth that may support effective growth of purslane. Higher content and availability of nutrients in tuff-peatmoss substrate could explain the enhanced yields as well as height of purslane" (section 3.1).

Comment	<i>Line 331: "Ash content was the highest (29.0%) with tuff-peatmoss-perlite (2:1:1)." However, in the table 4 it can be the highest value for "soil" sample (35.2°), please rewrite the sentence.</i>
Response	This sentence was rephrased to clarify: "Among the soilless media, ash content was highest with tuff-peatmoss-perlite" (section 3.4).
Comment	Conclusion: It should be reduced emphasizing the main findings and applications. Sounds like a abstract.
Response	The conclusion has been reduced, emphasizing the main findings and possible applications.

Manuscript INDCRO-D-19-00886R2 (July 2019)

Revised title: "Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (*Portulaca oleracea* L.) grown in different soilless media in a closed system"

Initial title: "Herbal yield, nutritive composition, phenolic content and antioxidant activity of purslane (*Portulaca oleracea* L.) are influenced by soilless growth medium in a closed system"

Response to Editors' Comments

Dear Editors,

Thank you kindly for your thorough assessment of our work. Your constructive comments are appreciated. They have all been taken into account to improve the manuscript as indicated below. All the comments and suggestions from the reviewers have also been addressed. Red font is used to highlight the revisions in the manuscript.

Best regards

	Editors' Comment/Authors' response
Comment	Discussion requires to show novelty of the study with conclusive comparisons.
Response	As noted in the introduction, there is a dearth of information on the impact of soilless culture and soilless substrates on the yield, composition and phenolic content of purslane. The present study was conducted to address this gap. The novelty of this study was also emphasized in the revised discussion (section 3.1): "To our knowledge, it is the first study to report on the yield, height as well as proximate composition, total phenolic, flavonoid and anthocyanin contents, and antioxidant activity of purslane grown in different soilless media". Conclusive comparisons are presented in sections 3.1 to 3.5, based on the conclusive comparisons shown in tables and figure.
Comment	In addition to the comments of the reviewers, follow the check list below and modify your manuscript accordingly. If an item on the checklist doesn't apply to your manuscript, just skip it. Write in red all changes made to your manuscript in next revision, do not use Word Track changes.
Response	The manuscript has been carefully revised considering all the items in the checklist. Red font was used to indicate the changes made in the manuscript.
1)	<i>Title: Avoid low impact words such as 'effects of', 'influence of', 'characterization of', etc., any part of the title. Title must be declarative, descriptive or a question</i>
	OK, the revised title is descriptive and does not contain low impact words.
2)	Do not use abbreviations in highlights
	OK, the highlights do not contain any abbreviations.
3)	All acronyms must be spelled out in the abstract
	OK, the abstract does not contain acronyms.
4)	Write in third person, avoid personal pronouns, such as we, they, you, I, or our, their, yours
	OK, third person is used throughout the manuscript.
5)	Abstract must have rationale, objective, materials and methods and conclusions. First sentence must be a rationale
	OK, the revised abstract starts with a rationale (lines 26-28), followed by the other elements.
6)	Common names of plants, animals, fungi, etc. must be followed by the Latin name the first time the common name is used. Latin name must include Authority example: maize (Zea mays L.)
	OK, in title, abstract and introduction.

7)	Equations must have the form $y=a + bx$, correct text, figures and tables
	OK, Equations follow this format.
8)	All statistical parameters y, x, n, r^2 , P, p etc must be in Italics in text figures and tables. Use small case r^2 for linear equations , R^2 is used only for non-linear regressions
	OK, this format has been used in the text, tables and figure.
9)	Use significant digits only in values and use . period for decimal separation check all tables and Figures
	OK, all the tables and figure have been checked and corrected when needed.
10)	For currency use only US dollars and Euros
	Not applicable
11)	Justify first column of tables to the left
	OK, first column of tables is justified to the left.
12)	Tables, make sure the independent variables are in the first column. You might need to transpose columns and rows, dependent variables in columns #2 to #n with the unit below.
	OK, the independent variables are in the first column of tables.
13)	Do not start sentences with abbreviations or numbers
	OK, there are no sentences starting with an abbreviation or number.
14)	Abbreviation for number is no
	OK
15)	No space between the unit and Celsius symbol, correct all
	OK, there is no space between the unit and Celsius symbol (e.g., 20°C).
16)	replace 'compared to' with 'compared with', correct all
	OK, "compared with" was used throughout the manuscript.
17)	Add one sentence of rationale to the beginning of your abstract
	The revised abstract starts with a rationale (lines 26-28).
18)	No bold text or values in tables
	OK, there is no bold text or bold values in the tables.
19)	Replace ppm for mg/kg or mg/L
	Not applicable
20)	Tables: Units go below header lines. Delete units from captions. Correct all tables
	OK, all the tables have been checked and corrected when needed.
21)	Add in your manuscript your reply to comments where the question was raised. A future reader of your publication might have a similar question
	OK, our responses to the questions raised were added to the revised manuscript (red font) as suggested.
22)	Format your tables to journal style. No vertical lines and only 3 horizontal lines, top, bottom and line below header
	OK, all the tables have been formatted according to the journal style, with no vertical lines and only 3 horizontal lines in each table.
23)	Only one table per page after references. Move Figures to the end of the text after tables, one

	figure per page with the caption below the Figure
	OK, one table per page after the references, followed by the figure, with the caption below the figure. The illustrations have been submitted in a separate Word document.
24)	<i>Check references format (Johnson, 1993), (Johnson and Smith, 1993), (Johnson et al., 2003). Use Elsevier reference formatting</i>
	Reference format has been checked, ensuring that Elsevier format for this journal is used in the text and reference section.
25)	Tables must stand alone, indicate the meaning of all abbreviations used on the table in a footnote. Footnotes indicators must have small case letter in italics and superscript $(a, b, c \text{ or } x, y z)$ do not use * for footnotes. One line per footnote below the table
	OK, all the tables have been formatted as suggested. * is no longer used for footnotes.
26)	Use 12 August 2016 to indicate dates, not August 12th, 2016
	OK, this format was used.
27)	All units and values are separated by space except % and Celsius degree symbol °C examples: 15 mL, 20 mi, 600 nm, 1000 kg/ha, 46%, 20°C, 8.60 g
	OK, this format was used throughout the manuscript.
28)	Use a comma before the final item in a list of three or more items. For example: "Cores were inside plastic liners, capped, and stored on ice"
	OK, this format was used in lists containing three or more items.

- Highest herbal yields in soilless purslane grown on a combination of tuff-peatmoss.
- Highest protein and lipid contents in purslane grown on tuff-peatmoss-perlite.
- Highest total phenolics and flavonoids in purslane grown on tuff-peatmoss-perlite.
- Highest antioxidant activity in tuff-peatmoss-perlite and zeolitic tuff.
- Soilless purslane is a good source of proteins, lipids, and antioxidant phenolics.

1 2 3	Herbal yield, nutritive composition, phenolic contents and antioxidant activity of purslane (<i>Portulaca oleracea</i> L.) grown in different soilless media in a closed system
4 5	
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7	Rababah ^a , Mohammad N. Alhamad ^b , Abdelrazzaq Al-Tawaha ^c , Abdel Rahman Al-
8	Tawaha ^d , Sana Gammoh ^a , Khalil I. Ereifej ^a , Ghazi Al-Karaki ^c , Hassan R. Hamasha ^e ,
9	Carole C. Tranchant (ORCID https://orcid.org/0000-0002-2026-819X) ^f , and Stan
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27 ABSTRACT

Purslane (Portulaca oleracea L.) is a valuable plant and crop with potential industrial 28 uses, yet little is known on how its cultivation could benefit from soilless substrates. This 29 30 study aimed to assess the effects of different soilless growth media on herbal yields (fresh and dry), proximate chemical composition, total phenolic, flavonoid and anthocyanin 31 content, and antioxidant activity of purslane cultivated in a closed system. The greatest 32 33 yields over five harvest cycles were obtained with tuff-peatmoss (2:1 mixture) compared with other soilless substrates, although the edible leaves were not as rich in proteins, 34 35 lipids, minerals, and phenolic compounds. The highest content of proteins (31.4% and 30.4%), lipids (0.68% and 0.75%), total phenolics (646.9 and 684.9 mg/100 g), flavonoids 36 (597.8 and 563.8 mg/100 g), and moisture (92.5% and 93.5%) in the leaves were found in 37 38 purslane grown in tuff-peatmoss-perlite (2:1:1) and in zeolitic tuff, respectively. 39 Antioxidant activity of leaf extracts was also the highest in purslane grown in both substrates and was similar to the antioxidant activity of leaf extracts from soil-grown 40 41 purslane obtained commercially and from the wild. The protein and lipid content obtained with tuff-peatmoss-perlite (2:1:1) and zeolitic tuff were superior to those of soil-grown 42 43 purslane. The results show that the nutritive and antioxidant qualities of purslane can be enhanced through soilless cultivation and selection of suitable culture media. 44

45

Keywords: Purslane, Soilless cultivation substrates, Yield, Proximate composition,
Phenolics and flavonoids, Antioxidant activity.

48 **1. Introduction**

Common purslane (Portulaca oleraceae L.) is an edible herbaceous plant commonly 49 distributed in much of Europe, the Mediterranean region, the Middle East, Asia, Mexico, 50 the Caribbean and North America. It belongs to the Portulacaceae family, which consists 51 52 of more than 120 species of succulent herbs and shrubs, and grows well in poor soils and hot dry conditions (Cudney et al., 2007). In many parts of the world, including Asia and 53 Mediterranean countries, purslane is grown as a specialty crop valued for its nutritional 54 55 and medicinal properties. Its leaves and stems have a slightly sour and salty taste similar to 56 spinach and are consumed as a leafy vegetable (Chan et al., 2000). Its yellow flower buds are also consumed. Purslane is a rich source of essential nutrients, mainly minerals 57 (Bianco et al., 1998; Uddin et al., 2012), vitamin C, vitamin E and ω -3 fatty acids, 58 particularly α-linolenic acid (Liu et al., 2000; Petropoulos et al., 2015; Simopoulos, 2004), 59 as well as bioactive phytochemicals such as carotenoid and phenolic antioxidants with 60 proposed health benefits (Alam et al., 2014; Erkan, 2012; Kopsell et al., 2016; Uddin et 61 al., 2012). The aerial parts of the plant are used for their antiseptic, anthelmintic, anti-62 63 inflammatory, and antispasmodic properties, and to help manage arthritis, osteoporosis as 64 well as psoriasis (Uddin et al., 2014; Xiang et al., 2005).

Agronomic practices have attracted much attention recently to optimize the factors 65 66 involved in crop management through a better control of plant growth and nutrient requirements to improve plant health, yields, and product quality under greenhouse 67 conditions (Aaby et al., 2010; Atanassova et al., 2007). Soilless culture is considered as 68 one of the main components of sustainable protected horticulture and is gaining particular 69 70 interest in countries with scarce water resources, limited agricultural land, and soil salinity problems (Putra and Yuliando, 2015). Closed soilless techniques offer new opportunities 71 to minimize water losses and maximize the efficiency of fertilizer use, in addition to 72

reducing environmental pollution caused by fertilizer runoff (Rouphael and Kyriacou, 73 74 2018; Rouphael et al., 2004; Van Os, 1999). Other advantages of closed soilless culture, which contribute to its importance on commercial scale, include high yields, cleaner and 75 76 year-long cultivation, and products with minimum herbicide and pesticide residues, which is expected for crops intended for human consumption as foods or health products (e.g., 77 nutraceuticals and nutritional supplements) (Hassanpouraghdam et al., 2010; Martínez et 78 79 al., 2013). Soilless closed systems are also favored to control the growth of pests and insects, reduce contamination and improve the recirculation of plant nutrients. For pest 80 81 control, these systems offer a safer alternative technique to the use of methyl bromide, a 82 now-banned or phased out pesticide used to disinfect soils before planting (Alcon et al., 2010). 83

84 The chemical composition and nutritional quality of some crops have been shown to differ 85 in soil and soilless systems. Strawberries, for instance, were found to have lower values of sugars, total solids, and sugars-to-acid ratio when cultivated in soilless closed systems 86 87 compared with soil in an open system, except when coconut fiber was used as a soilless substrate in an open system (Recamales et al., 2007). Very few studies are available on the 88 89 impact of soilless cultivation on plant phenolic content and antioxidant capacity. Some differences in total phenolic and anthocyanin content have been noted in strawberries 90 91 grown in closed vs. open soilless systems (Hernanz et al., 2007). Except for one report 92 indicating that purslane adapts well to a peat-based floating cultivation system, producing a high yield and lipid content (Cros et al., 2007), there is a dearth of information on the 93 impact of soilless culture and substrates on the yield, nutritional quality, and phenolic 94 95 content of purslane.

96 The aim of the present study was to assess the effects of soilless substrates (tuff, peatmoss,
97 perlite, and their combination) on the yield, height, proximate composition, total phenolic,

flavonoid and anthocyanin contents, and antioxidant activity of common purslane grown
in a closed system. A comparison with soil-grown purslane was also conducted in terms of
chemical composition and antioxidant activity.

101 **2. Materials and methods**

102 2.1. Plant materials and chemicals

103 Purslane seeds were obtained from a local store in Irbid, Jordan. For purslane cultivated in 104 soil (open systems), fresh samples were obtained from three local sources in Irbid, namely the Jordan University of Science and Technology (JUST) campus where purslane was 105 106 cultivated in an open garden (same seeds as for soilless-grown purslane), a local market, and one location in the city where wild (non-cultivated) purslane was found. These 107 samples were designated as "soil", "market", and "wild", respectively, and characterized 108 for chemical composition, phenolic contents and antioxidant activity. All chemical 109 reagents were of analytical grade. 110

111 2.2. Soilless cultivation under closed conditions

112 2.2.1. Soilless cultivation substrates and experimental design

113 Soilless cultivation was conducted in a greenhouse at the JUST campus (Irbid, Jordan) 114 during one growing season. Cultivation started at the end of February; harvesting began in 115 April and continued until July. Germination and subsequent growth after transplantation 116 were under natural light conditions. Ventilation was provided automatically by a cooling 117 system when the air temperature exceeded 28°C. Purslane seedlings were transplanted at the four-leaf stage into multicellular iron trays (20 cm x 300 cm x 25 cm, W x L x D) 118 filled with seven soilless horticultural substrates, namely (1) tuff, (2) peatmoss, (3) 119 120 peatmoss and perlite (2:1), (4) tuff and peatmoss (2:1), (5) tuff, peatmoss and perlite 121 (2:1:1), (6) zeolitic tuff, and (7) tuff and peatmoss (1:1). Tuff, perlite and zeolitic tuff are inorganic components, while peatmoss is organic. Different combinations of these 122

123 materials were tested in this study. The bases of the beds were elevated at a slope of 1.5% with a hole in the tray wall to which a channel was attached to drain excess water which 124 was collected into tanks for reuse. Harvesting of the above-ground biomass, which 125 126 consisted of stems and leaves, was performed manually by cutting the stems at 5 cm above the level of the soilless substrate. This biomass was used to determine the yields, while the 127 other determinations were carried out using the leaves. Treatments were randomly 128 129 assigned to experimental units (i.e., trays) using a randomized complete block design with 130 substrate as factor and three replications per treatment.

131 *2.2.2. Nutrient solution and irrigation system*

Water and nutrients were provided with complete nutrient solutions prepared from commercial fertilizers with some modifications. The procedures for nutrient replenishment and water discharge were applied at the same time to all replicates using a drip irrigation system. The nutrient solutions were prepared freshly once every three weeks. Clark's nutrient solution (Clark, 2008) was pumped from independent tanks. Electrical conductivity and pH values of the nutrient solution were maintained at 2.0-2.5 and 5.5-6.5 dS/m, respectively.

139 2.3. Plant height and yield measurements

Biomass measurements of soilless-grown purslane were taken at five harvesting points during the growing season. Plant height (cm) was measured, then the plants were harvested at 5 cm above the ground. Fresh and dry weights (g) were measured and converted to fresh and dry yields (g/m^2) , respectively. For dry yield, the samples were dried at room temperature for 10 days. Plant material obtained from the first cycle of harvest was used for chemical analyses.

146 2.4. Proximate analysis

147 Proximate chemical composition of the leaves of soilless- and soil-grown purslane was determined according to the AOAC method (1990) with triplicate determinations. 148 Moisture content was determined by drying the samples at 100°C until constant weight. 149 150 Total nitrogen content of the dried samples was determined using the micro-Kjeldahl method and a conversion factor of 6.25 to calculate crude protein content. Total lipids 151 were determined by Soxhlet extraction. For crude fiber, the dried samples were digested 152 153 with 1.25% sulfuric acid and 1.25% potassium hydroxide. Ash content was determined by burning 1 g of dried sample in a muffle furnace at 550°C for 24 h. 154

- 155 2.5. Determination of phenolic compounds
- 156 *2.5.1. Total phenolic content*

157 Half a gram of dried leaves from soilless- and soil-grown purslane was mixed with 50 ml of methanol at 30°C for 12 h, with stirring, to extract the total phenolic constituents. The 158 samples were then filtered into a 50 ml volumetric flask through a filter paper (Whatman 159 no. 42) and the volume was completed to mark. The extracts were kept in the refrigerator 160 161 at 4°C until further analyses. The content of total phenolics in the extracts was determined 162 according to the Folin-Ciocalteu method described by Singleton et al. (1999). Two 163 milliliters of extracts were transferred into a test tube and mixed with 2.5 ml of 10% Folin-Ciocalteu reagent. After 3 min, 2 ml of 10% sodium carbonate solution (Na₂CO₃) was 164 added. The tubes were allowed to stand for 1 h at room temperature, then absorbance was 165 measured in triplicate at 760 nm using a UV-VIS spectrophotometer (SpectroScan 50, 166 167 Biotech Engineering Management Co., UK) against a blank which consisted of methanol instead of test sample. Gallic acid was used as calibration standard, with different 168 concentrations to prepare a standard curve, and the concentration of total phenolics was 169 expressed as gallic acid equivalent (mg of GAE/100 g of sample on a dry weight basis). 170

171 2.5.2. Total flavonoid content

172 Total flavonoid content of the leaves was determined according to the aluminum chloride colorimetric method described by Zhishen et al. (1999). Half a milliliter of methanolic 173 extract was mixed with 150 µl of a 15% sodium nitrite solution (NaNO₂). After 6 min, 174 175 150 µl of a 10% AlCl₃ was added with stirring, then after another 6 min, 2 ml of NaOH solution (4%) and 2 ml of distilled water were added to bring the final volume to 5 ml. The 176 mixture was mixed and allowed to stand for 1 h at room temperature, then absorbance was 177 178 measured in triplicate at 510 nm (UV-VIS spectrophotometer, SpectroScan 50, Biotech 179 Engineering Management Co., UK) against a blank which consisted of methanol. Catechin 180 was used as calibration standard and the concentration of total flavonoids was expressed as catechin equivalent (CE) (mg of CE/100 g on a dry weight basis). 181

182 2.5.3. Total anthocyanin content

Anthocyanins were extracted using the method described by Rabino and Mancinelli (1986). Two grams of dried leaves were mixed with acidified methanol (50 ml, 1% HCl) by stirring at 60°C for 60 min. The resulting extract was filtered by using filter paper (Whatman no. 3) and then kept in the dark in the refrigerator until further analyses. Absorbance was measured in triplicate at 530 nm and 657 nm. Anthocyanin content was expressed as cyanidin 3-glucoside equivalent (CGE) (mg of CGE/100 g on a dry weight basis) and calculated using the following equation:

190 Anthocyanins
$$(mg/g) = \left(\frac{AS530 - (0.25 * AS657)}{29.60}\right) \times Mw \times Df \times \left(\frac{V}{Sw}\right)$$
 (1)

where AS_{530} and AS_{657} are the absorbance at 530 nm and 657 nm, respectively, Mw is the molecular weight of cyanidin 3-glucoside (449.1 g/mol), 29.60 is the extinction coefficient, Df is the dilution factor, V is the total volume (ml), and Sw is the sample weight (g).

The antioxidant activity of purslane leaves was measured using the 2,2-diphenyl-1-196 picrylhydrazyl (DPPH) assay described by Brand-Williams et al. (1995). Sample solutions 197 with different concentrations were prepared from the methanolic extracts of total 198 199 phenolics. For each concentration, an aliquot of freshly prepared DPPH solution in methanol (0.5 mg/ml). The mixture was mixed thoroughly and incubated for 60 min in a 200 201 dark environment at room temperature. Its absorbance was then measured in triplicate at 202 517 nm with a spectrophotometer (UV-VIS SpectroScan 50, Biotech Engineering 203 Management Co., UK). The percentage of DPPH free radical scavenging was calculated 204 using Eq. (2):

205 % Scavenging =
$$\left(\frac{Ab-As}{Ab}\right)$$
 x 100%.....(2)

where Ab is the absorbance of the blank (DPPH solution alone) and As is the absorbance of the test sample. The values of IC_{50} , which represent the extract concentration required to inhibit (scavenge) 50% of the DPPH radicals, were calculated from the plot of percentage scavenging against extract concentration. The values of IC_{50} are inversely proportional to the sample antioxidant activity.

211 2.7. Statistical analyses

The data were analyzed by analysis of variance (ANOVA) performed with MSTAT-C (version 4.0, 1985, Michigan State University, East Lansing, MI, USA) using a least significant difference (LSD) of $p \le 0.05$ for mean separation.

215

216 **3. Results and discussion**

217 3.1. Effect of soilless substrates on purslane fresh yield

Fresh and dry plant yields are important indicators required by purslane growers to assess the economic value of this crop. Moreover, appropriate growth substrates are critical for achieving high crop production, especially when water is a limiting factor. In this study,

the effect of soilless substrate on purslane yield and height was evaluated over five harvest cycles. To our knowledge, it is the first study to report on the yield, height as well as proximate composition, total phenolic, flavonoid and anthocyanin contents, and antioxidant activity of purslane grown in different soilless media.

As shown in Table 1, fresh yield at all the harvest cycles varied significantly depending on soilless substrate. Tuff-peatmoss (2:1) resulted in the highest fresh yields across all the harvest cycles, ranging from 3889 to 6238 g/m² at the 1st and 4th cycles, respectively. The effect of other substrates tended to vary depending on harvest cycle. The second or third highest values of fresh yield after those obtained with tuff-peatmoss (2:1) were achieved with tuff, peatmoss, tuff-peatmoss (1:1), peatmoss-perlite (2:1) or tuff-peatmoss-perlite (2:1:1), depending on harvest cycle.

232 In contrast, zeolitic tuff resulted in the lowest fresh yields, ranging from 121.1 to 3035 g/m^2 at the 1st and 3rd harvest cycles, respectively. Peatmoss-perlite (2:1) and peatmoss 233 also resulted in relatively low fresh yields (265.6 and 3246 g/m²) but only at the 1st and 3rd 234 235 cycles, respectively. Regardless of the soilless substrate, fresh yield initially increased with harvest cycle, followed by a slight reduction after a certain number of harvest, usually 236 after the 4th or 3rd cycle depending on the substrate. This influence of harvest cycle could 237 238 be due to the adaptation of the plants to the substrates and their rapid vegetative growth (including increasing dimensions of leafs and stems) in the initial stages, followed by dry 239 240 matter accumulation and moisture reduction in later stages as the plants mature. Higher yields may reflect relatively large stem diameters, which would be expected to enhance the 241 mechanical strength of the stems and thus their ability to resist breaking and bending under 242 243 growing conditions. Purslane with greater stem diameter may thus be obtained more successfully with tuff-peatmoss (2:1) and after a few harvest cycles. 244

245 These novel findings indicate the superiority of soilless substrates that contain peatmoss for supporting the rapid growth of purslane in a closed system. This is consistent with the 246 high purslane yield obtained in a peat-based floating system as compared with coir and 247 248 perlite (Cros et al., 2007). In our work, tuff-peatmoss (2:1) was particularly effective at all the harvest cycles. This suggests that the nutrient content, availability and specific 249 physicochemical characteristics of peatmoss-containing substrates best meet the 250 physiological needs of purslane for rapid growth and development under the closed 251 252 growing conditions used. This is probably related to the high water holding capacity 253 (WHC), high cation exchange capacity (CEC) and high organic matter content of tuffpeatmoss substrates (Manoloc et al., 2005). Their high CEC helps retain the minerals 254 255 (which reduces nutrient leaching) and enables a gradual release of nutrients over time, 256 while high WHC improves water retention and management. Tuff and peatmoss in 257 combination also improve the structure of the growth substrate, which contributes to proper aeration and drainage. This creates suitable conditions for root growth that may 258 259 support effective growth of purslane. Higher content and availability of nutrients in tuff-

260 peatmoss substrate could explain the enhanced yields as well as height of purslane.

261 High quality peatmoss imparts beneficial physical properties to horticultural growth media in addition to a high CEC (Treadwell et al., 2007). Peat is also considered an important 262 263 sink for atmospheric carbon dioxide, although the time needed to regenerate a peat bog 264 after harvest tends to be quite long (several decades), which can limit the availability of peat from some locations (Raviv et al., 1998; Treadwell et al., 2007). Caution is required 265 when using peat for some plants as it may contribute to the propagation of Pythium 266 267 damping-off, a plant disease caused by Pythium ultimum (Hoitink and Boehm, 1999). Thus, the sterilization of peat-based media has been recommended to eliminate pathogens 268 before basil seeding (Reuveni et al., 2002; Treadwell et al., 2007). 269

270 Combining peatmoss with perlite (2:1) or tuff, peatmoss and perlite (2:1:1) resulted in moderate vields of purslane in our work. In crisp-head lettuce, Gül et al. (2005) found that 271 plant growth was significantly lower with perlite compared with zeolite. The higher 272 273 growth obtained with zeolite was attributed to an increase in the uptake of nutrients since zeolites have a high CEC, which enables them to act as a reservoirs, holding elements in 274 275 their structure for slow release to the rhizosphere (Gül et al., 2005). In contrast, Maloupa and Gerasopoulos (1999) reported that the use of perlite for gerbera cultivation led to a 276 277 higher yield than zeolite. The relatively low yields of purslane obtained with zeolitic tuff 278 in our work suggest that this substrate lacked important characteristics required to support the rapid growth of purslane. Zeolites have a high CEC and a high content of macro and 279 280 micro-minerals, but their organic content is very low (Gül et al., 2005). These 281 characteristics may have limited the provision of essential nutrients by zeolitic tuff or their 282 uptake by purslane, which resulted in lower plant yields compared with the other soilless substrates. Cros et al. (2007) and Biernbaum (2007) noted that the highest yields were 283 284 obtained in plants grown for short time in either peat or vermiculite-based closed cultivation system as compared with coir or perlite. 285

286 *3.2. Effect of soilless substrates on purslane dry yield*

The impact of soilless growth medium on purslane dry yield was significant at all the 287 harvest cycles (Table 2) and was similar to the effect on fresh yield. Consistent with the 288 289 latter, tuff-peatmoss (2:1) resulted in the highest dry yields across all harvest cycles, ranging from 365.1 to 558.6 g/m² at the 1^{st} and 4^{th} cycles, respectively. The effect of other 290 soilless substrates tended to vary depending on the harvest cycle. The second or third 291 highest values of dry yield after those obtained with tuff-peatmoss (2:1) were achieved 292 with tuff, peatmoss, tuff-peatmoss (1:1), peatmoss-perlite (2:1) or tuff-peatmoss-perlite 293 (2:1:1), depending on harvest cycle. These results indicate the superiority of soilless 294

substrates that contain peatmoss for supporting the rapid growth of purslane in a closedsystem.

In contrast, zeolitic tuff resulted in the lowest dry yields, ranging from 8.0 to 266.4 g/m^2 at 297 the 1st and 4th harvest cycles, respectively. Peatmoss-perlite (2:1) resulted in similarly low 298 values at the 1st and 2nd cycles. However, the yields obtained with both substrates 299 increased substantially after a few cycles. At the 3rd, 4th 5th harvest cycles with zeolitic 300 tuff, they were about 2-fold lower than the highest dry yields obtained with tuff-peatmoss 301 (2:1), while they were about 40-fold and 4-fold lower at the 1^{st} and 2^{nd} cycle, respectively. 302 This suggests a relatively rapid adaptation of purslane to zeolitic tuff. Regardless of 303 substrate, dry yield continuously increased across all harvest cycles, but seemed to level 304 off after the 4th cycle, which is consistent with plant adaptation and rapid vegetative 305 growth in the early stages, followed by maturation in later stages. 306

307 *3.3. Effect of soilless substrates on purslane height*

308 The effect of soilless growth medium on purslane height was significant at all the harvest 309 cycles (Table 3) and was consistent with the effect on fresh and dry yields. Tuff-peatmoss (2:1) resulted in the highest plant heights across all the harvest cycles, ranging from 45.87 310 to 67.40 cm at the 1st and 4th cycles, respectively. Tuff-peatmoss (1:1) and peatmoss also 311 produced a high height at the first harvest cycle, which did not differ significantly from the 312 value obtained with tuff-peatmoss (2:1). After the first cycle, the second or third values of 313 314 plant height after those obtained with tuff-peatmoss (2:1) were achieved with tuff, peatmoss, tuff-peatmoss (1:1), peatmoss-perlite (2:1) or tuff-peatmoss-perlite (2:1:1), 315 depending on the harvest cycle. The lowest plant heights across all the cycles were 316 obtained with zeolitic tuff (18.23 to 46.60 cm) and peatmoss-perlite (2:1) (22.80 to 53.80 317 cm). Regardless of the substrate, plant height initially increased with harvest cycle, 318

followed by a slight decline after the 4th cycle, in agreement with the results obtained in terms of fresh yield.

These findings show the superiority of soilless substrates that contain peatmoss for 321 322 supporting rapid purslane growth in a closed system. Similarly, Cros et al. (2007) reported higher plant height in purslane grown in a peat-based floating system as compared with 323 324 coir and perlite. In barley, a significant increase in straw yield and plant height has been reported upon the enrichment of loamy sand soil with a peatmoss-shrimp waste compost 325 326 (Hountin et al., 1995). These researchers evidenced a significant relationship between soil 327 organic carbon and straw yield and plant height, while grain yield was correlated with soil total nitrogen. 328

329 3.4. Proximate composition of purslane leaves from soilless- and soil-grown plants

330 The proximate chemical composition of purslane leaves from soilless-grown and soilgrown plants is shown in Table 4. The effect of soilless substrate was significant on all the 331 332 compositional characteristics considered. Moisture content was the highest (92.5%-93.5%) 333 in purslane leaves grown in peatmoss-perlite (2:1), tuff-peatmoss-perlite (2:1:1), zeolitic tuff, and tuff-peatmoss (1:1). The highest protein contents (29.9%-31.4% of dry weight) 334 335 were obtained with tuff, peatmoss, peatmoss-perlite (2:1), tuff-peatmoss-perlite (2:1:1), zeolitic tuff, and tuff-peatmoss (1:1), while the highest lipid contents (0.68%-0.75%) were 336 obtained with tuff-peatmoss-perlite (2:1:1) and zeolitic tuff. Fiber content (about 12%) 337 338 was relatively constant across soilless substrates. Among the soilless media, ash content was the highest (29.0%) with tuff-peatmoss-perlite (2:1:1), which was comparable to the 339 level found in commercial (market) and wild purslane, but lower than the level (35.2%) 340 341 found in purslane grown in soil at the research facilities.

342 Overall, the leaves of purslane grown in tuff-peatmoss-perlite (2:1:1) and zeolitic tuff 343 stood out as displaying among the highest levels of proteins and lipids. These

344 compositional characteristics are highly valuable from the nutritional standpoint, but they have not been reported before in soilless grown purslane. Purslane lipids are mainly 345 346 unsaturated and rich in ω-3 fatty acids (Petropoulos et al., 2015). The fact that the highest 347 yields of purslane were recorded with another soilless substrate (tuff-peatmoss 2:1) is not entirely surprising. In other food crops, it has been reported that the nutritional quality of 348 349 the crop tends to decline as crop yields increase (Benbrook, 2009; Halweil, 2007). Too much readily available nitrogen (N) in the soil or other growing media generally reduce 350 351 nutrient density as well as flavor of the food (Halweil, 2007). In our study, it is plausible that tuff-peatmoss-perlite (2:1:1) and zeolitic tuff supplied less N to the plant compared 352 353 with tuff-peatmoss (2:1), which may explain the greater content of some nutrients in 354 purslane leaves grown in the former substrates.

355 Significant differences in proximate composition were also found between soilless-grown 356 and soil-grown purslane. Protein and lipid contents were significantly greater in purslane leaves grown in tuff-peatmoss-perlite (2:1:1) and zeolitic tuff compared with soil-grown 357 358 purslane (market, wild and soil). Moisture content was also higher in general in soilless-359 grown leaves compared with soil-grown leaves. For fiber and ash, in contrast, one type of 360 soil-grown purslane (soil) yielded the greatest levels (16.0% and 35.2%, respectively). For soil-grown purslane, our findings are in agreement with those presented by Ezekwe et al. 361 362 (1999) for proteins (22.8 to 25.4%), total carbohydrates (49.0 to 56.1%), ash (15.9 to 21.5%) and moisture (79.4 to 90.6%) in the leaves of different purslane accessions. 363 364 However, the total lipid contents in their study (3.8 to 6.5%) were higher than in our work. For total lipids, our findings are consistent with those reported by Uddin et al. (2012) 365 366 (0.51%) in the leaves of soil-grown purslane.

The differences in proximate composition evidenced in the present study may be explainedby differences in the balance and bioavailability of nutrients in the substrates that were

369 tested. The influence of growth substrate on the chemical composition of vegetables and fruits has been reported in some crops, although not in purslane. In tomatoes, several 370 studies reported higher contents of dry matter, sugar, vitamins, and carotenoids in soilless 371 372 systems compared with soil (Gruda, 2009). A few studies, however, found that these contents were higher in soil-grown tomatoes than in soilless-grown fruits (Gruda, 2009). 373 374 In a review of the effects of organic and inorganic culture media on vegetable quality and productivity under greenhouse conditions, Olle et al. (2012) concluded that it is difficult to 375 376 draw general conclusions on the impact of growth media on vegetable composition as 377 results vary with crop, physicochemical composition of the substrate, and nutrient bioavailability to the plant. 378

379 3.5. Phenolic contents and antioxidant activity of purslane leaves from soilless- and soil380 grown plants

381 The total phenolic, flavonoid and anthocyanin contents of methanolic extracts from purslane leaves varied significantly depending on growth substrate, as illustrated in Table 382 383 5 for plants cultivated under soilless and soil conditions. Variations amongst soilless substrates are discussed first. In soilless-grown purslane, the highest concentrations of 384 385 total phenolics were obtained with zeolitic tuff, tuff-peatmoss-perlite (2:1:1), and tuffpeatmoss (2:1) (684.9, 646.9, and 633.4 mg/100 g, respectively). These values did not 386 387 differ significantly. In contrast, tuff-peatmoss (1:1), tuff, and peatmoss resulted in the 388 lowest levels of total phenolics (456.8, 481.4, and 501.5 mg/100 g, respectively), which did not differ significantly. Flavonoid content was the highest with tuff-peatmoss-perlite 389 (2:1:1) (597.8 mg/100 g), followed by zeolitic tuff (563.8 mg/100 g). Both values differed 390 391 significantly. Tuff-peamoss (1:1) and tuff in resulted in the lowest flavonoid contents (429.1 and 448.6 mg/100 g, respectively), which did not differ significantly. Anthocyanin 392 393 content was the highest with peatmoss (311.7 mg/100 g), which was significantly greater than the second highest values obtained with tuff and zeolitic tuff (294.7 and 289.5 mg/100 g, respectively). Tuff-peatmoss (2:1) yielded the lowest concentration of anthocyanins (196.7 mg/100 g).

397 These findings show that the leaves of purslane grown in zeolitic tuff, tuff-peatmossperlite (2:1:1), or tuff-peatmoss (2:1) are particularly rich in total phenolics. Tuff-398 peatmoss-perlite (2:1:1) and zeolitic tuff also resulted in high levels of flavonoids, while 399 400 peatmoss resulted in high anthocyanin content. Amongst the soilless substrates tested in this study, tuff-peatmoss-perlite and zeolitic tuff appear especially promising as they also 401 402 resulted in the highest levels of proteins, lipids and total solids in the leaves, in addition to high total phenolic and flavonoid contents and intermediate contents of anthocyanins. 403 404 Tuff-peatmoss (2:1), which resulted in the highest plant yields, resulted in high contents of 405 total phenolics and intermediate contents of flavonoids.

406 When all the soilless-grown and soil-grown treatments were compared, the leaves from 407 wild purslane showed significantly higher contents of total phenolics, flavonoids, and 408 anthocyanins (1019, 644.9, and 412.9 mg/100 g, respectively) (Table 5). For total 409 phenolics, the second highest levels were found in plants grown in zeolitic tuff, tuff-410 peatmoss-perlite (2:1:1), tuff-peatmoss (2:1) as well as in purslane from the market and from soil cultivation at our facilities, with no significant difference amongst these values. 411 For flavonoids, the highest contents were found in wild purslane and in market samples. 412 413 Both were significantly higher than the contents obtained with tuff-peatmoss-perlite (2:1:1) and zeolitic tuff. For anthocyanins, the highest levels after wild purslane were 414 found in purslane cultivated in soil at the research facilities, followed by purslane grown in 415 416 peatmoss, with significant differences between these values. Commercial (market) purslane displayed the lowest anthocyanin content compared with the other treatments. 417

418 These findings show that purslane grown in tuff-peatmoss-perlite and zeolitic tuff

419 compared favorably with soil-grown purslane in terms of their richness in total phenolics and flavonoids. For anthocyanin content, peatmoss-grown plants compared favorably with 420 wild purslane and with purslane cultivated in soil at the research facilities. Anthocyanin 421 422 content obtained with peatmoss, zeolitic tuff and tuff-peatmoss-perlite (2:1:1) was superior to those of market purslane. Overall, tuff-peatmoss-perlite and zeolitic tuff stand out as 423 424 soilless substrates of choice as they resulted in high phenolic and nutrient concentrations. 425 Based on these novel findings, the leaves of soilless purslane grown in tuff-peatmoss-426 perlite (2:1:1) or zeolitic tuff could be recommended as rich sources of phenolics and 427 flavonoids for use in various industries including the food industry.

Consistent with the high total phenolic contents of the leaves, the phenolic extracts from 428 429 purslane leaves grown in tuff-peatmoss-perlite (2:1:1) and zeolitic tuff showed the highest 430 antioxidant activity (lowest IC₅₀ values of 0.596 and 0.584 mg/ml, respectively, which did 431 not differ significantly) compared with other soilless substrates, followed by tuff-peatmoss 432 (2:1), as illustrated in Figure 1. The high antioxidant activity found with tuff-peatmoss-433 perlite and zeolitic tuff did not differ significantly from the high antioxidant activity 434 detected in extracts from wild purslane and market purslane (Figure 1). Their antioxidant 435 activity, however, was significantly lower for the extracts from purslane grown in soil at the research facilities. The lowest antioxidant activities (highest IC_{50}) were found with 436 peatmoss (1.247 mg/ml), followed by tuff and tuff-peatmoss (1:1) (1.097 and 1.029 437 438 mg/ml, respectively).

Thus, purslane leaves from cultivation in tuff-peatmoss-perlite (2:1:1) and zeolitic tuff possessed a high antioxidant activity, similar to that of wild and commercial purslane, in addition to high contents of total phenolics and flavonoids. The antioxidant activity obtained with tuff-peatmoss (2:1) was relatively high despite being lower than with tuffpeatmoss-perlite and zeolitic tuff. This is noteworthy because tuff-peatmoss (2:1) is the soilless substrate that resulted in the highest plant yields and height. These findings
suggest that the leaves of soilless purslane grown in the above substrates could be used as
a source of natural preservatives with antioxidant properties. Further research could be
conducted to confirm the antioxidant activity of extracts from soilless purslane using
complementary methods in addition to the DPPH assay which was used in the present
study.

450 There are no published reports of purslane phenolic composition and antioxidant activity 451 when this plant is cultivated in soilless substrates. For wild purslane leaves, the relatively 452 high antioxidant activity in our work (IC₅₀ of 0.56 mg/ml) is consistent with the IC₅₀ value of 0.511 mg/ml reported by Erkan (2012) in methanolic extracts. Uddin et al. (2012) 453 454 reported the phenolic content of soil-grown purslane at different stages of growth. In 455 ethanolic extracts, these researchers found that the total phenolic contents of the leaves 456 ranged from 174.5 to 348.5 mg/100 of fresh weight at 15 and 60 days, respectively. They 457 reported slightly lower total phenolic and flavonoid contents in ethanol extracts (276.8 and 458 41.3 mg/100 g, respectively) than in methanol extracts (360.3 and 49.2 mg/100 g) at day 30. These values seem lower than the levels found in the present work, especially for 459 460 flavonoids. However, it should be kept in mind that their values were expressed on a fresh weight basis, while ours are based on dry weight, which could explain some of the 461 discrepancy. Uddin et al. (2012) reported lower antioxidant activity of the leaf extracts, 462 463 i.e., greater IC₅₀ values (1.71 to 1.30 mg/ml at day 15 and 60, respectively) than in our 464 work (0.2 to 1.2 mg/ml).

Values of IC_{50} of 0.456 and 0.391 mg/ml were reported by Montoya-García et al. (2018) at two different harvest times of soil-grown purslane. These values increased to 0.508 mg/ml upon the application of 300 kg of N/ha to the fertilizer. The values of IC_{50} in their work are comparable to the values found in the present study with soil-grown purslane and with

469 purslane grown in zeolitic tuff and tuff-peatmoss-perlite. Montoya-García et al. (2018) 470 further showed that the decrease in antioxidant activity resulting from nitrogen application was accompanied by a decrease in total flavonoid content and a slight increase in total 471 472 phenolics. Cultivation methods such as organic, conventional, soil and soilless methods in open or closed systems have been showed to influence the phenolic contents of other crops 473 474 (Benbrook, 2009; Hernanz et al., 2007), although no clear trends have been established because of conflicting results. In strawberry fruits, for instance, Asami et al. (2003) 475 reported higher levels of total phenolics from organic and sustainable cultivation 476 477 compared with conventional practices, while Hakkinen and Torronen (2000) found no consistent effect of organic cultivation on the total phenolic content compared with 478 479 conventional cultivation. Wild strawberries have been found to exhibit greater levels of 480 phenolic compounds compared with cultivated fruits (Muthukumaran et al., 2017; Yildiz 481 et al., 2014), as found in the present work with purslane leaves.

482

483 **4.** Conclusions

The findings from this study indicate that purslane soilless cultivation in select culture 484 485 media has promising potential for producing high quality purslane with value-added characteristics, specifically high protein, oil, total phenolics, flavonoids, and antioxidant 486 activity, which are highly valuable and sought after for applications in the food, 487 488 nutraceutical, and pharmaceutical industries, among others. Tuff-peatmoss-perlite (2:1:1) and zeolitic tuff showed particularly high potential with respect to the high contents of 489 proteins, lipids, total phenolics and flavonoids in the leaves. For some applications, tuff-490 491 peatmoss-perlite may be preferred over zeolitic tuff as the latter produced relatively low purslane yields. The highest herbal yields were obtained with tuff-peatmoss (2:1). Other 492 493 soilless substrates could also prove useful depending on the responses or characteristics 494 desired in purslane. This warrants further investigation with consideration of the various

nutrients and biologically active phytochemicals present in this plant. 495

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Table 1

Effect of different soilless substrates on the fresh yield of purslane over five harvest cycles during the growing

Fresh yield (g/m^2)						
Soilless substrates	1 st harvest	2 nd harvest	3 rd harvest	4 th harvest	5 th harvest	Total
Tuff	841.1 ^d	2773 ^{cd}	4026 ^c	4306 ^c	4253 ^b	16199 ^c
Peatmoss	2291 ^b	2718 ^d	3246 ^e	3473 ^d	3308 ^d	15036 ^d
Peatmoss:Perlite (2:1)	265.6 ^{ef}	2217 ^e	4078°	4241 ^c	3708 ^c	14509 ^d
Tuff:Peatmoss (2:1)	3889 ^a	4890^{a}	5868 ^a	6238 ^a	6038 ^a	26923 ^a
Tuff:Peatmoss:Perlite (2:1:1)	$410.0^{\rm e}$	3390 ^b	3651 ^d	3704 ^d	3446 ^d	14601 ^d
Zeolitic tuff	121.1^{f}	1278 ^f	3035 ^e	2758 ^e	2547 ^e	9739 ^e
Tuff:Peatmoss (1:1)	1265 ^c	2948 ^c	4487 ^b	4712 ^b	4332 ^b	17744 ^b

season under closed conditions.

Mean (n=3) separation within columns was by least square difference (LSD) at the 5% level. Means in the same column followed by different letters are significantly different ($p \le 0.05$).

Table 2

Effect of different soilless substrates on the dry yield of purslane over five harvest cycles during the growing season under closed conditions.

		Γ	Dry yield (g/m ²)			
Soilless substrates	1 st harvest	2 nd harvest	3 rd harvest	4 th harvest	5 th harvest	Total
Tuff	62.03 ^d	199.9 ^d	316.1 ^b	412.3 ^b	408.3 ^b	1398 ^b
Peatmoss	178.9 ^b	194.1 ^d	272.4 ^c	341.4 ^c	325.9 ^e	1312 ^c
Peatmoss:Perlite (2:1)	18.77 ^{ef}	157.2 ^e	333.0 ^b	398.7 ^b	385.4 ^c	1293 ^c
Tuff:Peatmoss (2:1)	365.1 ^a	404.9^{a}	439.8 ^a	558.6 ^a	553.2 ^a	2315 ^a
Tuff:Peatmoss:Perlite (2:1:1)	30.8 ^e	281.9 ^b	308.7 ^b	344.4 ^c	339.3°	1305 ^c
Zeolitic tuff	8.00^{f}	97.47^{f}	232.2 ^d	266.4 ^d	245.2^{f}	849.2^{d}
Tuff:Peatmoss (1:1)	94.83 ^c	221.5 ^c	338.9 ^b	394.0 ^b	366.4 ^d	1415 ^b

Mean (n=3) separation within columns was by least square difference (LSD) at the 5% level. Means in the same column followed by different letters are significantly different ($p \le 0.05$).

Table 3

Effect of different soilless substrates on the height of purslane over five harvest cycles during the growing season under closed conditions.

			Plant height (cm)		
Soilless substrates	1 st harvest	2 nd harvest	3 rd harvest	4 th harvest	5 th harvest
Tuff	28.27 ^{bc}	44.07 ^c	48.67 ^d	54.93 [°]	51.00 ^b
Peatmoss	40.80^{a}	44.27°	47.93^{df}	53.67 ^c	49.33 ^b
Peatmoss:Perlite (2:1)	22.80 ^{cd}	35.40 ^d	45.13 ^f	53.80 ^c	47.80^{bc}
Tuff:Peatmoss (2:1)	45.87^{a}	53.87 ^a	59.47 ^a	67.40^{a}	59.53 ^a
Tuff:Peatmoss:Perlite (2:1:1)	32.00 ^b	49.87^{b}	50.67 ^c	58.60^{b}	48.00^{bc}
Zeolitic tuff	18.23 ^d	35.33 ^d	46.60 ^{ef}	44.60^{d}	44.27 ^c
Tuff:Peatmoss (1:1)	42.67^{a}	43.00 ^c	53.00 ^b	57.47 ^b	51.67 ^b
LSD	6.026	2.078	1.715	1.405	4.063

Mean (n=3) separation within columns was by least square difference (LSD) at the 5% level. Means in the same column followed by different letters are significantly different ($p \le 0.05$).

Table 4.

Proximate chemical composition of the leaves of purslane grown in different soilless substrates and in soil.

Treatments	Proteins	Lipids	Crude	Moisture	Ash
	(%)	(%)	fiber (%)	(%)	(%)
Soilless-grown, closed condition					
Tuff	30.3 ^{abc}	0.165^{b}	11.9 ^{bc}	92.0 ^b	24.0^{cd}
Peatmoss	29.9^{abc}	0.046^{b}	11.9 ^{bc}	92.0^{b}	23.0 ^{cd}
Peatmoss:Perlite (2:1)	30.4 ^{ab}	0.046^{b}	11.6^{bc}	93.5 ^a	26.0^{bc}
Tuff:Peatmoss (2:1)	28.0^{bc}	0.083^{b}	11.9 ^{bc}	90.5 [°]	22.0^{d}
Tuff:Peatmoss:Perlite (2:1:1)	31.4 ^a	0.681 ^a	11.9 ^{bc}	92.5 ^{ab}	29.0 ^b
Zeolitic tuff	30.3 ^{abc}	0.755^{a}	11.0 ^{cd}	93.5 ^a	24.0 ^{cd}
Tuff:Peatmoss (1:1)	30.5 ^{ab}	0.088^{b}	13.3 ^b	92.5^{ab}	22.0^{d}
Soil-grown, open conditions					
Market	21.8 ^d	0.156 ^b	9.31 ^d	85.0^{d}	29.5 ^b
Wild	27.8°	0.141^{b}	10.0^{cd}	81.5 ^e	28.9^{b}
Soil, at the research facilities	19.3 ^d	0.193 ^b	16.0 ^a	91.5 ^{bc}	35.2 ^a
LSD	2.516	0.372	2.014	1.317	3.6

Results are expressed on a dry weight basis. Mean (n=3) separation within columns was by least square difference (LSD) at the 5% level. Means in the same column followed by different letters are significantly different $(p \le 0.05)$.

Table 5.

Contents of total phenolics, flavonoids and anthocyanins of the leaves of purslane grown in different soilless substrates and in soil.

Treatments	Total phenolics	Flavonoids	Anthocyanins
	(mg GAE/100g)	(mg CE/100g)	(mg CGE/100g)
Soilless-grown, closed conditions			
Tuff	481.4 ^d	448.6 ^e	294.7 ^d
Peatmoss	501.5 ^d	481.0^{d}	311.7 ^c
Peatmoss:Perlite (2:1)	611.1 ^c	508.6 ^d	288.1 ^e
Tuff:Peatmoss (2:1)	633.4 ^{bc}	500.5 ^d	196.7 ^g
Tuff:Peatmoss:Perlite (2:1:1)	646.9 ^{bc}	597.8 ^b	238.3 ^f
Zeolitic tuff	684.9^{b}	563.8 ^c	289.5^{de}
Tuff:Peatmoss (1:1)	456.8 ^d	429.1 ^e	238.2^{f}
Soil-grown, open conditions			
Market	646.9 ^{bc}	631.9 ^a	139.0 ^h
Wild	1019 ^a	644.9 ^a	412.9 ^a
Soil, at the research facilities	636.5 ^{bc}	395.1 ^f	345.7 ^b
LSD	73.60	28.46	6.397

Results are expressed on a dry weight basis.

Mean (n=3) separation within columns was by least square difference (LSD) at the 5% level. Means in the same column followed by different letters are significantly different ($p \le 0.05$).

GAE: gallic acid equivalents, CE: catechin equivalents, CGE: cyanidin-3-glucoside equivalents.

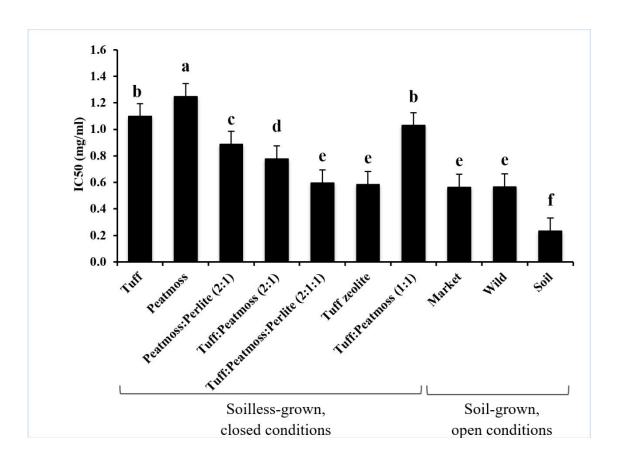


Fig. 1. Antioxidant activity expressed as IC₅₀ of extracted phenolics from the leaves of purslane grown in different soilless substrates and in soil. Different letters indicate significant differences in treatment means from three determinations ($p \le 0.05$).