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Criteria for HydroSOS Quality Index. Application to Extra Virgin Olive Oil and Processed Table Olives

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Abstract: Water, especially in arid and semiarid regions, is increasingly a disputed commodity among different productive sectors; the pressure for a more sustainable use of water in agriculture will grow. The main strategy to cope with water scarcity is the use of improved, innovative, and precise deficit irrigation management practices which are able to minimize the impact on fruit yield and quality. The aim of this paper was to develop a certification index or hydroSOS quality index for extra virgin olive oil and processed table olives. The hydrosSOS fruits and vegetables are those cultivated under regulated deficit irrigation (RDI). Different indicators in three quality areas ((i) fatty acids, (ii) phenolic compounds, and (iii) sensory attributes) were identified as showing characteristic or typical responses under RDI conditions. Marks or scores were assigned to each one of these indicators to calculate the proposed index. It can be concluded that an extra virgin olive oil (EVOO) or processed table olives are hydroSOStainable foods, if they meet 2 conditions: (i) fulfill the conditions established in the hydroSOS "irrigation" index, and (ii) fulfill the requirements of the hydroSOS "quality" index. HydroSOS quality index will be specific to each crop and variety and will depend on functional and sensory factors.

Keywords: fatty acids; oleuropein; regulated deficit irrigation; saving water; sensory attributes; total phenolic compounds

1. Introduction

In most worldwide agrosystems, it is not possible to get maximum crop yields without an adequate supply of irrigation water to complete rainfall and avoid plant water stress. In this way, a regular fruit yield is obtained and an alternative production pattern is avoided or at least reduced. As the population increases, water, especially in arid and semiarid regions, is increasingly a disputed commodity among different productive sectors. This competition for the water is due to the expansion of urban, touristic, and industrial activities that frequently lead to tensions, conflict between users, and extreme pressure on the environment [1]. It is clear that climate change will inevitably lead to very frequent and severe droughts in a near future [2]. Thus, the pressure for a more sustainable use of water in agriculture will grow as agriculture uses 60% or 1500 trillion liters of the 2500 trillion liters of water it uses each year, which represents 70% of the world's accessible water [3]. Consequently, arid and semiarid agrosystems will be forced to cope with water scarcity and to practice sustainable agriculture. In this sense, the main strategies to cope with water scarcity are: (i) the use of improved, innovative, and precise deficit irrigation management practices which are able to minimize the impact on fruit yield and quality; and (ii) the use of plant materials with low water-demand and/or able to withstand deficit irrigation with minimum impact on yield and fruit quality [4].

Deficit irrigation can improve fruit quality by raising the dry matter percentage and the levels of healthy bioactive compounds [5,6]. Among these compounds, particular mention must be made to a huge variety of secondary metabolites, mainly phenolic compounds. Phenolics represent an important source of biological activities [7] and have frequently been associated with beneficial effects for human health [8]. Regulated deficit irrigation (RDI) is probably the most useful deficit irrigation strategy to improve water saving and, if properly applied, to increase harvest quality without or having minimum impacts in marketable yield. RDI is based in reducing irrigation, or even completely stop irrigation, during the water stress-tolerant phenological periods (non-critical periods) and supplying full irrigation during the water stress-sensitive phenological periods (critical periods) [4,9,10]. On the other hand, full irrigation consists of providing non-limiting water conditions (100% of the evapotranspiration, ET_c) to the crop during the whole cycle of the plant.

Galindo et al. [4] indicated that in the Mediterranean agrosystems to save water and protect the integrity of water resources, there is need for diversification of production and consumption habits. These habits include the use of a broader range of plant species, such as those currently being underutilized and needing a low input of synthetic fertilizers, pesticides, and water. Moreover, these authors indicated that this new approach must be compatible with the consolidation of the cultivation of traditional crops, such as olives, almonds, or grapevines, which are low-water-demanding and certainly very profitable crops.

Recently, Corell, et al. [11] proposed an approach based on scientific knowledge about deficit irrigation and other management techniques for olive trees that quantify the effort to save water in olive orchards in a sustainable way. These authors considered indicators from 4 areas ((i) hydraulic indicators, (ii) horticultural indicators not related to irrigation scheduling, (iii) horticultural indicators related to the moment when deficit irrigation is applied (when?), and (iv) horticultural indicators related to the way deficit irrigation is applied (how?)). The evaluation of these aspects allows the authors to develop a "hydroSOStainable index" (or "hydroSOS index"), that evaluates different aspects at orchard level to improve the sustainability of the water resources.

It has been demonstrated that deficit irrigation can have beneficial effects on the accumulation of bioactive compounds in fruits, and also in consumer satisfaction in "deficit-irrigated" peaches [12], pistachios [13], table olives [14], and olives for extra virgin olive oil (EVOO) [15]. Moreover, in recent studies it has been shown that European consumers think that any effort carried out to obtain "special" products is compensated by their characteristic and special sensory attributes and their health-promoting effects on humans [16]. Also, Noguera-Artiaga, et al. [17] showed that Spanish consumers were willing to pay an extra amount of 1.0 € per kg of hydroSOS pistachios.

For these reasons, it is considered that the global hydroSOStainable index (or hydroSOS index) should be divided into two complementary indexes (Figure 1): (i) a hydroSOStainable "irrigation" index (or hydroSOS irrigation index) for the orchards adequately labelled according to the criteria established by Corell et al. [11]; and (ii) a hydroSOStainable "quality" index (or hydroSOS quality index) for the orchards in which fruits or their derived products have specific and improved physical, chemical, and sensory characteristics, and come from orchards already awarded with the hydroSOS irrigation index label.

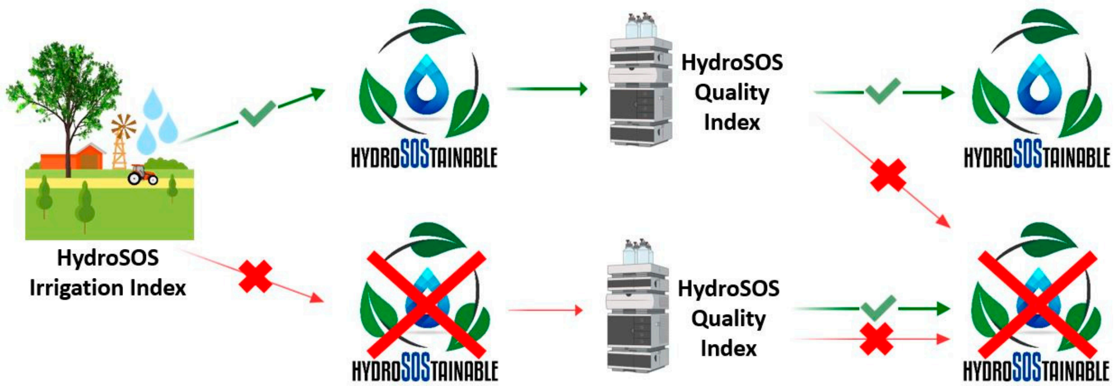

Figure 1. HydroSOStainable index: two complementary indices.

At this last respect, the aim of this paper was to develop a certification index or hydroSOS quality index for extra virgin olive oil and processed table olives from orchards under RDI. To establish this new index, different indicators in three quality areas ((i) fatty acids, (ii) phenolic compounds, and (iii) sensory attributes) were identified as showing characteristic or typical responses under RDI conditions and, thus, different marks or scores have been assigned for each of these indicators.

2. Materials and Methods

The proposed materials and methods [12–15,18–20] are recommended as they have been widely used and are easy to implement. However, other standardized methods can be used and will provide similar and valid results, if properly applied.

2.1. Main Concepts to Consider for the HydroSOS Quality Index

The key concept in the hydroSOS irrigation index is that a proper irrigation management has been used, such as regulated deficit irrigation (RDI) strategy. In order to obtain a successful RDI, it is necessary to monitor the phenology of the crop and control the level of water stress achieved (using different methods such as the pressure bomb technique), and the duration of that water deficit. In olive trees, a moderate water restriction applied during pit hardening, when the resistance to the water deficit in the soil is higher [21–23], allows water saving, without affecting yield and fruit weight and maximizing growers' profit [22,24].

The quality and organoleptic properties of the olives and its oil content is influenced by the implementation of good agronomic practices. It is very complex to define the concept of quality in fruits because it involves many different aspects (physical and chemical characteristics, sensory attributes, healthy effects, technological and economic aspects, etc.). Additionally to this complexity, it is necessary to mention that the quality indicators change over time and may be different in different countries and markets [25]. These are some of the reasons why the protocols used in this area should be revised periodically to incorporate or quantify new or time-dependent elements.

The approach used here, it is the same used by Corell et al. [11] to build the hydroSOS irrigation index in orchards. Both hydroSOS indexes (irrigation and quality) are complementary and have a structure similar to that of the "Seattle Green Factor" [26], which was designed to evaluate green urban zones. The approach followed in this document (hereinafter, "HydroSOS Quality Index") includes different indicators that allow applying the water deficit strategy and getting an enhancement of the quality of fruit and related food-stuffs. These indicators are grouped into chemical and sensory changes influenced only by the water deficit applied and those changes influenced by fruit processing. This approach is due to the fact that the processing of olive fruits to obtain both table olives and olive oil alters both physico-chemical and organoleptic attributes [27,28].

2.2. Preparation of Extra Virgin Olive Oil (EVOO) and Table Olives

Extra virgin olive oil extraction (EVOO). Virgin olive oil is the product obtained exclusively by mechanical or physical means [29]. In addition and to be considered "extra virgin" olive oil (EVOO), it must comply with two requirements: (i) have a maximum free acidity (<0.8 g oleic acid per 100 g) [30], and (ii) have a sensory profile reminding that of freshly harvest olives and with no defects [31]. EVOO was extracted using an Abencor system and the oil obtained was separated by decanting. Oil samples were filtered and stored in amber glass bottles, without headspace, at −18 °C in darkness until analysis.

Table Olives Processing. The Spanish style is the most popular industrial process to produce table olives [32]. This process consists of an initial bleaching treatment followed by fermentation of lactic acid, after addition of the brine [33,34]. During the different stages of this process, changes in the bioactive composition of the olives will happen. For example, during the alkaline treatment, both rutine and luteolin-7-glucoside decrease, while the *p*-coumaric acid increases. The next step of the process is the fermentation, where hydroxytyrosol content increases significantly at the first but remains practically constant throughout the fermentation stage [35]. Additional to polyphenols, α-tocopherol and fatty acids can also decrease during olive processing [28]. However, despite all these changes in the olive and its composition due to processing, environmental factors can also influence these changes. Several studies have shown that water deficit affects the content of fatty acids and to a lesser extent the composition of the volatile compounds. Due to these chemical changes occurring in olives, their organoleptic characteristics are altered [14,18,36].

2.3. Total Phenolic Compounds

The content of total phenolic compounds (TPC) in a methanolic extract can be studied using the Folin–Ciocalteau reagent and the methodology described by Tuberoso et al. [37]. The procedure consists of extracting the TPC of ~3 g of lyophilized olives or ~3 g of olive oil, using as extractant solution methanol/water (80/20 *v*/*v*). The generated biphasic solution is stirred for 2 min and subsequently, the methanolic phase is collected and filtered through a filter with hydrophilic regenerated cellulose with stationary phase (Minisart RC 0.45 µm, 25 mm, Sartorius, Goettingen, Germany). The filtrate is saved and the residual lipid phase is extracted again with 5 mL of extractant solution, collecting the filtered methanolic phase and mixing it with that of the first extraction. The total methanolic phase is evaporated in a rotary-evaporator. The residue obtained after evaporation is reconstituted in 1.5 mL of pure methanol.

2.4. Analysis of Fatty Acids

Fatty acids methyl esters (FAMEs) can be prepared according to ISO-12966-2 [19]. The chromato graphic analysis of the samples after transmethylation is conducted following the method described by ISO-12966-4 [20], with slight modifications. The equipment to be used consisted of a gas chromatograph (model C-17A; Shimadzu Corporation, Kyoto, Japan) connected to a flame ionization detector (FID), equipped with a capillary column CPSil-88 (100 m × 0.25 mm ID. 0.2 µm film thickness; J&W 112-88A7; Agilent Technologies, Santa Clara, CA, USA). The oven program used was as follows: (i) initial temperature 175 °C for 10 min, (ii) rate of 3 °C min^{-1} from 175 to 220 °C, (iii) and hold for 5 min. Samples were injected with a 1:20 split ratio and identification of peaks was made by comparison with FAMEs standards from Sigma-Aldrich. The carrier gas was He and the gases used at the FID detector flame were H_2 (30 mL min^{-1}) and air (350 mL min^{-1}), and He (30 mL min^{-1}) was used as a make-up gas. Detector temperature was 260 °C.

2.5. Sensory Analysis

Organoleptic evaluation of table olives and olive oils must be conducted following the European Council regulation. The panel consisted of eight expert tasters (with over 500 h of experience on vegetable products) from the Department of Agro-Food Technology of Universidad Miguel Hernández

(UMH) and simultaneously, oil samples were sent to the Official Panel of the Murcia Region, to obtain a certified commercial classification of the oils. Panelists were provided with oil and olive samples, ~100 mL and 15 g, respectively, in blue glass cups in the case of the oils, coded using 3-digit numbers. The testing room was approximately at ~21 °C and the samples were kept at ~28 °C. Illumination was a combination of natural and non-natural (fluorescent) light.

For instance, sensory evaluation of the processed olives was conducted according to the procedure described by Cano-Lamadrid et al. [14]. For the descriptive analysis, a numerical scale from 0 to 10, with 0.5 increments was used. Both positive attributes and defects were quantified, including defects (e.g., including fusty/muddy, musty-humid-earthy, winey-vinegary, etc.), and positive attributes (e.g., fruitiness, bitterness and pungency) [38].

3. Results and Discussion

3.1. Quality Indicators for Extra Virgin Olive Oils (EVOOs)

The following concepts are described to highlight those characteristics that have more relevance in the EVOOs obtained using irrigation management strategies (hydroSOS EVOOs):

- Sensory attributes. The quality criteria applied to the EVOO include chemical but also sensory parameters (color, flavor, and volatile compounds), and the sensory analysis must be conducted by an official panel or a panel with wide expertise on application of sensory analysis in research on vegetable products [39,40]. The sensory characteristics of EVOOs are mainly attributed to variations in the composition of phenolic compounds, especially oleuropein, which gives the bitter taste to this product and variations in the profile of volatile compounds, the majority being hydrocarbons, alcohols, aldehydes, ketones, and esters [41,42]. Furthermore, it is possible that alterations in the fatty acids (FAs) of these oils also affect variations in sensory characteristics [43,44]. To obtain the certification hydroSOS, EVOOs must have significant intensity of positive attributes: bitterness, pungency, and especially fruitiness. These attributes can be influenced by the irrigation treatments, and the working hypothesis is that EVOOs coming from fully irrigated orchards will have lower intensity of these positive attributes than those coming from RDI orchards [42].

In this sense, only those EVOOs whose parameters increase their intensity more than 10% with respect to those taken as control [40,42,45] will be considered as hydroSOS-EVOO. The scores to be used for the certification process are those shown in Table 1, and to have the highest score in this section they must have increases of >10% in three sensory attributes.

- Fatty acids. Its profile can vary not only depending on the variety and degree of maturity in which the olives are harvested, but also on environmental factors such as latitude and climate [46,47]. The hydroSOS oil, resulting from trees subjected to water stress, will have the highest score in this section if they show an increase in oleic acid above 5% and a decrease in linoleic acids of more than 10% [48]. The scores to be used for the certification process are those shown in Table 1.
- Total phenolic compounds (TPC). Phenolic compounds have a large impact on the flavor and color of fruit and fruit-based products, because they play an important role in the oxidation of plant tissues [49]. Phenolic compounds and their associated antioxidant activity are important because they promote the resistance of the oil to the development of rancidity, among other deterioration processes. The stability of the oil has been correlated not only with the total amount of phenolic compounds, but also with the presence of certain substances [50]. The level of phenolic compounds in the olive oils can be influenced by several factors such as the cultivar, the degree of maturation and the industrial processes used for the extraction, as well as the environmental conditions being very important the water deficit suffered by trees [51,52].

Table 1. Indicators, levels, and marks of the hydroSOS quality index in extra virgin olive oils.

Indicators	Level	Mark
Sensory attributes: bitter, pungent, and fruity	>10% in three attributes	5
	>10% in two attributes	4
	>10% in only one attribute	2
Fatty acids	Increase > 5% in oleic acid and decrease >10% in linoleic acid	5
	Increase 4.9%–3.0% in oleic acid and decrease 9.9%–7.0% of linoleic acid	4
	Increase 2.9%–1.0% in oleic acid and decrease 6.9%–5.0% in linoleic acid	3
	Increase < 1.0% in oleic acid and decrease < 5.0% in linoleic acid	2
Phenolic compounds	Increase > 30% in TPC (total phenolic compounds) and >20% in oleuropein	10
	Increase > 30% in TPC and 19.9%–15% in oleuropein	9
	Increase 29.9%–20% in TPC and 14.9%–10% in oleuropein	8
	Increase 19.9%–10% in TPC and 9.9%–5% in oleuropein	5
	Increase 9.9%–5% in TPC and 4.9%–2.5% in oleuropein	3

In this sense, some authors have reported that olive oils from trees subjected to water stress during pit hardening contain a higher content of phenolic compounds than those from well-watered trees [39,53]. It is important to mention that although water deficit improves polyphenols contents, when water stress was too high (above 60% ET_c during pit hardening), results were contradictory because the plant metabolism was drastically affected. For this, 60% ET_c is marked as the maximum water stress threshold to be applied [44]. Besides, the content of oleuropein, which is one of the most important compounds within the phenolic fraction, increases under water stress conditions [40]. EVOOs having simultaneous increases above 30% and 20% in TPC and oleuropein, respectively, as compared to the control samples get the highest score in this section [40,54]. The scores used for the certification process are those shown in Table 1.

3.2. *Quality Indicators for Processed Olive Fruits (Table Olives)*

In order to evaluate the certification of hydroSOS table olives, sensory analysis and the content of fatty acids (FAs) of these processed fruits obtained from water stressed trees was studied.

- Sensory evaluation. Water deficit significantly affects the organoleptic characteristics (color, flavor, aroma, and texture) of table olives, because this stress causes the plant defense accumulating bioactive compounds. The green-olive flavor characteristic of green table olives results usually from the synergistic combination of different flavor notes present in fresh olives (fruity, green, vegetable/herbaceous, citrus, vinegar, and even wood notes); these sensory descriptors are due to the simultaneous occurrence of several volatile compounds including alcohols, aldehydes, terpenes, organic acids, and phenolic compounds [55]. Sensory parameters can be altered depending on the intensity of the water stress applied to the trees. In this sense, if the applied stress is moderate, all sensory parameters except sourness, crunchiness, and fibrousness are altered, being bitterness, green-olive flavor, aftertaste and hardness, the most sensitive parameters. In contrast, when the applied water stress is severe, the most sensitive parameter is sweetness [14,18]. Therefore, those processed olives that showed increased green-olive flavor intensity and long aftertaste will be candidates to be certified as hydroSOS products [14]. From the point of view of evaluating the sensory analysis in an objective way, an increase above 10% in the intensity of the green-olive flavor and aftertaste will get the maximum score, while the rest of the scores to be used for the certification process of table olives are shown in Table 2.
- Fatty acids. The most sensitive FA to water stress, according to literature, is linoleic acid; although, this abiotic stress can also affect the content of linolenic acid [36]. In this way, monounsaturated fatty acids (MUFAs) content can also be affected by water stress in olive orchards. According to the literature, the application of RDI in table olives leads to an increase in polyunsaturated fatty

acids (PUFAs) and a decrease in MUFAs [14,18,56]. Table 2 describes how the hydroSOStainability of table olives is evaluated according to their fatty acid profiles.

Table 2. Indicators, levels, and marks of the hydroSOS quality index in table olives. MUFAs, mono-unsaturated fatty acids; PUFAs, poly-unsaturated fatty acids.

Indicators	Level	Mark
Green-olive flavor and aftertaste	Increase > 10% in green-olive flavor and aftertaste	10
	Increases 9.9%–7.5% in green-olive flavor and aftertaste	7.5
	Increases 7.4%–5.0% of green-olive flavor and aftertaste	5
	Increases 4.9%–2.5% of green-olive flavor and aftertaste	2.5
Fatty acids	Increase > 15% of PUFAs and decrease >4% of MUFAs	15
	Increase 14.9%–10.0% of PUFAs and decrease 3.9%–2.0% of MUFAs	10
	Increase 9.9%–5% of PUFAs and decrease 1.9%–0.5% of MUFAs	5
	Increase < 5% of PUFAs and decrease < 0.5% of MUFAs	2

3.3. HydroSOS Quality Labels

From the point of view of quality, the sum of the set of all the considered indicators classifies the products under control into four labels, according to the potential sustainability achieved in water management (Table 3). This index evaluates if the water stress that farmers have applied to the orchard has been appropriate from the point of view of sustainability and has being strong enough to significantly affect also the composition and sensory profile of the final commercial products (EVOO and/or table olives). Besides, the logo associate to this index will help consumers to identify those products that objectively come from a company involved in the preservation of the water resources. The sum of the marks for all evaluated indicators was 20 points for EVOO and 25 points for table olives.

The highest label, Label A, indicates a fully hydrosSOS product (Table 3). Label B was granted to those products getting total scores in the range 16.9–13.0 points for EVOO and 20.9–16.0 points for table olive fruits (Table 3). This label (B) indicates a final product having improved sensory quality and good bioactive composition, but worse than expected for a product grown under soft/moderate RDI. Given this result, a better irrigation schedule must be applied to achieve a moderate water stress; it is important to mention that a too severe stress can lead to similar composition to that of the control products or even worse due to a collapse of the plant metabolism. Label C reaches values between 12.9–10.0 points for EVOO and 15.9–12.5 for table olives (Table 3), which represents final products with some negative results, especially those related to the content of bioactive compounds. In this case, it is possible that the sensory quality begins to give a negative response with respect to the expected results. These results may be related to the fact that these final products are from an orchard which has important deficiencies from the point of view of sustainable irrigation. In view of these results, it is necessary to carry out a new irrigation schedule to achieve an improvement in the applied RDI. Finally, Label D, was below 10 points in EVOO and 12.5 in table olives, indicating that final products have very low quality, in both the analyses of bioactive compounds and sensory profiles. The Label D scores are indicative that RDI is not being applied or it is applied at the wrong stages of the plant cycle or it is reached too intense or soft intensities.

Table 3. Final product indicators, levels, and comments of the hydroSOS quality index in extra virgin olive oils (EVOOs) and table olives.

Product	Label	Points	Comments
EVOOs	A	>17.0	HydroSOS.
	B	13.0–16.9	Interesting results but not hydroSOS yet.
	C	10.0–12.9	Important deficiencies in olive oil characteristics due to an incorrect irrigation management.
	D	<10.0	Very low quality. Regulated deficit irrigation was not properly applied.
Table olives	A	>21.0	HydroSOS.
	B	16.0–20.9	Interesting results but not hydroSOS yet.
	C	12.5–15.9	Important deficiencies in table olives characteristics due to an incorrect irrigation management.
	D	<12.5	Very low quality. Regulated deficit irrigation was not properly applied.

4. Conclusions

From all of the above, it can be concluded that an EVOO or table olives can be labeled as hydroSOS, if they meet two conditions: (i) fulfill the conditions established in the hydroSOS "irrigation" index [11], and (ii) fulfill the requirements of the hydroSOS "quality" index. These requirements can be summarized as follows: (i) for EVOOs, increases in fruitiness, bitterness, pungency, total content of phenolic compounds, oleuropein, and oleic acid and a decrease in linoleic acid; and (ii) for table olives, increases in green-olive flavor, aftertaste, oleic acid and a decrease in linoleic acid.

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