# Performance of Two Color Scales for Virgin Olive Oils: Influence of Ripeness, Variety, and Harvest Season

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ABSTRACT: The purpose of this investigation was to compare the performance of the bromthymol blue (BTB) method and the Uniform Oil Color Scale (UOCS) method with different sets of virgin olive oil samples from Andalusia (Spain), namely, 1213 samples from olives at three stages of ripeness, 1008 samples from eight olive varieties, and 1700 samples from olives harvested in four different crop seasons. All oils were extracted in the laboratory by the same procedure. The performance of the two color scales was compared using CIELAB color differences between the oil samples and the nearest standard from each scale. The UOCS performed at least 2.0 times better than the BTB for each of the three stages of olive ripeness, and the difference between the two color scales was statistically significant (P < 0.001). The UOCS performed at least 1.3 times better than the BTB for each of the eight olive varieties studied, and the differences between the two color scales were statistically significant (P < 0.02). The UOCS also performed at least 1.6 times better than the BTB for each of the four harvests analyzed, and the differences between the two color scales were statistically significant (P < 0.001). Using the current oil samples, we could discern no substantial improvements to the UOCS standards.

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**KEY WORDS:** Bromthymol blue method, BTB method, CIELAB, color of virgin olive oils, oil-color measurement, olive ripeness, olive variety, uniform oil color scale, UOCS.

The bromthymol blue (BTB) method (1,2) is currently the official method used to specify the color of virgin olive oils in Spain, the world leader in virgin olive oil production (3). This method is based on a visual comparison between the oil sample and a set of 60 standard solutions (BTB standards). Flaws in the BTB method have been reported (4,5), which have led to the recent proposal of a new color scale (6) called the Uniform Oil Color Scale (UOCS). Like the BTB method, the UOCS method uses visual comparison under specified visual conditions between the oil sample and a set of standards, but this comparison is made with a new set of 60 colors (UOCS standards), in which the color is more uniformly and appropriately distributed in color space than the previous BTB standards. For a set of 1700 virgin olive oil samples, the UOCS method was found to perform roughly twice as effectively as the BTB method (6). Specifically, the average color difference between each of these 1700 virgin olive oils and the nearest standard was reduced from 8.17 (SD: 6.64) CIELAB units (7) with the BTB standards to 3.99 (SD: 3.05) CIELAB units with the UOCS standards. This improved performance of the UOCS method relative to the BTB would be desirable for oil samples with different characteristics, a key question not specifically addressed when the UOCS method was proposed (6).

Olive variety, ripeness, and the oil-extraction process, which have a major impact on the organoleptic properties of the oil, particularly on oil color (8,9), are among the many characteristics influencing the quality of virgin olive oils. These characteristics are closely related to the composition of the extracted oil, especially to the content of chlorophyll and carotene, which are the main pigments responsible for oil color. In the present work, we focused on the main characteristics of olives from the standpoint of the color of their corresponding oils by considering the following three factors: the ripeness of the olive, which is considered a key factor in characterizing the extracted oil (10); the variety of the olive, which is deemed fundamental to the oil produced (11,12); and the harvest season during which the olives were collected, which includes potential differences attributable to many environmental conditions.

Thus, the goal of the current research was to determine how the BTB and UOCS methods were influenced by olive ripeness, variety, and harvest. Our analyses were based on computations from experimental measurements of the color coordinates of virgin olive oils and BTB standards, together with theoretical values of the UOCS standards (6). Visual measurements are not reported in the present work. Specifically, we were interested in identifying potential deficiencies in the performance of the current 60 UOCS standards (6) with respect to any of the characteristics mentioned above. In the case of a serious deficiency, it would be advisable to add new standards (or modify the current ones) before the UOCS method is firmly established for use by researchers or manufacturers.

## **EXPERIMENTAL PROCEDURES**

A set of 1700 samples of virgin olive oils was obtained (13) from olives collected in the most representative production

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 TABLE 1

 Number of Virgin Oil Samples Considered by Stage of Ripeness,

 Variety, and Harvest Season of the Olive

Olive parameters		Number of samples
Ripeness stage	Green	335
	Pintona	628
	Ripe	250
	Total	1213
Variety	Picual	565
	Hojiblanca	83
	Lechín	64
	Manzanilla	154
	Arbequina	75
	Verdial	33
	Picudo	23
	Pico Limón	11
	Total	1008
Harvest season	1994–95	572
	1995–96	502
	1996–97	528
	1997–98	98
	Total	1700

zones in Andalusia (Spain). All the oils were extracted in the laboratory of Almazara Experimental del Instituto de la Grasa (CSIC, Seville, Spain) by the Abencor<sup>®</sup> method (14) following the procedure described in Reference 6. Table 1 shows the number of virgin oil samples used in the current study, distinguishing the stage of ripeness, variety, and harvest season of the olive. Our 1700 oil samples corresponded to olives collected during four different harvests: 1994-95, 1995-96, 1996-97, and 1997-98. The number of oil samples in the first three harvests was similar and was considerably higher than the number in the last harvest. From the olives used to produce the 1700 oil samples, 1008 (59.3%) olive samples were identified as coming from one of the following eight varieties: Picual, Hojiblanca, Lechín, Manzanilla, Arbequina, Verdial, Picudo, and Pico Limón. Except for Arbequina (originally from Catalonia, but also common in Aragon and Andalusia) and Pico Limón, the six remaining olive varieties are considered the main varieties in Andalusia because they occupy a major percentage of the cultivated area or are predominant in a given zone (15). We also knew the harvest date for 1213 (71.4%) of the samples. From this date, we were able to distinguish three different stages of ripeness, based on our previous experience and bearing in mind the characteristics of each variety (13) (Table 2). According to this classification, we were able to assign more than half of our olive samples (51.7%) to the medium-ripe state, corresponding to a blushed appearance called "pintona."

Spectral transmittance measurements of the filtered oil samples were performed immediately after extraction (<1 h) (6). Measured values were referred to a 10-mm pathlength by means of the Lambert–Beer law (16) and were used to compute tristimulus values, assuming the D65 illuminant and the CIE 1964 Supplementary Standard Observer (7). These tristimulus values were transformed to CIELAB, assuming an *n*-hexane measured solution as the reference white.

 TABLE 2

 Olive Ripeness Stages Considered Bearing in Mind Collection Date for Each Variety<sup>a</sup>

Green	D' (		
	Pintona	Ripe	
Before Dec. 9	Dec. 10–Jan. 15	After Jan. 16	
Before Dec. 15	Dec. 16–Jan. 20	After Jan. 21	
Before Nov. 14	Nov. 15-Dec. 30	After Dec. 31	
Before Nov. 14	Nov. 15-Dec. 15	After Dec. 16	
Before Oct. 15	Oct. 16-Nov. 30	After Dec. 1	
Before Dec. 31	Jan. 1–Jan. 31	After Feb. 1	
Before Dec. 31	Jan. 1–Feb. 13	After Feb. 14	
Before Nov. 15	Nov. 16–Dec. 15	After Dec. 16	
	Before Dec. 9 Before Dec. 15 Before Nov. 14 Before Nov. 14 Before Oct. 15 Before Dec. 31 Before Dec. 31 Before Nov. 15	Green         Pintona           Before Dec. 9         Dec. 10–Jan. 15           Before Dec. 15         Dec. 16–Jan. 20           Before Nov. 14         Nov. 15–Dec. 30           Before Nov. 14         Nov. 15–Dec. 15           Before Oct. 15         Oct. 16–Nov. 30           Before Dec. 31         Jan. 1–Jan. 31           Before Dec. 31         Jan. 1–Feb. 13           Before Nov. 15         Nov. 16–Dec. 15	

<sup>a</sup>See Reference 13.

To compare the performance of the BTB and UOCS methods (both having the same number of standards, i.e., 60), we used the color difference between each oil sample and its nearest standard in the corresponding scale. Although the color differences should be computed using recent color spaces, such as DIN99 (17), or recently proposed color-difference formulas, such as CIEDE2000 (18), the results achieved in this way were very similar to those found with the well-known CIELAB system (7). Thus, we used only this latter system, which involves the Euclidean distance between the L\*a\*b\* coordinates of the two color samples, and may be the most familiar to readers. The CIELAB coordinates of the 60 UOCS and 60 BTB standards are provided in Reference 6 and Tables 2 and 3, respectively. For a given group of oil samples, we compared the average and SD of the CIELAB color differences to the nearest standards of each scale; the lower these values, the better the performance of the scale. The color differences were analyzed statistically using nonparametric tests (the Wilcoxon test for related samples, and the Kolmogorov-Smirnov test for unrelated samples) available in SPSS 11.5.1 software (19).

### **RESULTS AND DISCUSSION**

Figure 1 shows the average and SD of CIELAB color differences ( $\Delta E^*_{ab}$ ) for olives at different stages of ripeness (green, pintona, and ripe) using the BTB and UOCS methods. The average CIELAB color differences were in the range of 7.5 to 9.3 for the BTB and 3.7 to 4.5 for the UOCS. The SD of CIELAB color differences (designated by error bars in Fig. 1) for the BTB were also about twice those of the UOCS for the three ripeness stages. The nonparametric Wilcoxon test for related samples indicated that the differences between the BTB and UOCS at each of the three stages of ripeness were statistically significant (P < 0.001). The ratio (BTB/UOCS) of the average CIELAB color difference was in the range of 2.0 (pintona) to 2.1 (green and ripe), and the ratio of the SD was in the range of 2.0 (green) to 2.7 (ripe). Thus, the UOCS outperformed the BTB for each of the three stages of ripeness.

In addition, Figure 1 shows that higher values (i.e., poorer performance) were found for the BTB and UOCS methods for the least ripe (green) olives. The Kolmogorov–Smirnov nonparametric tests for unrelated samples indicated that the differences

	Picual	Hojiblanca	Lechín	Manzanilla	Arbequina	Verdial	Picudo
Green/pintona	1.15	0.73	0.85	1.11	1.22	1.11	
	(1.10)	(0.91)	(1.02)	( <u>1.80</u> )	( <u>0.65</u> )	(0.71)	
Green/ripe	1.16	1.19	_	0.88	<u>1.24</u>	_	_
	( <u>1.20</u> )	(1.39)		( <u>2.65</u> )	( <u>0.41</u> )		
Pintona/ripe	1.00	<u>1.63</u>		0.79	1.01		0.83
	( <u>1.08</u> )	( <u>1.53</u> )		(1.47)	( <u>0.63</u> )		(0.65)
1994–95/1995–96	<u>1.04</u>	1.15	0.76	0.74		0.84	1.02
	( <u>1.76</u> )	(1.42)	(0.54)	( <u>0.60</u> )		(1.43)	(1.10)
1994–95/1996–97	1.07	1.14	0.76	0.60		1.13	1.41
	( <u>1.71</u> )	(1.10)	(0.76)	(0.72)		(1.46)	(1.49)
1994–95/1997–98	0.88	0.80		0.44		_	_
	(1.44)	( <u>0.61</u> )		(0.68)			
1995–96/1996–97	1.03	0.99	0.99	0.82	1.58	1.36	<u>1.38</u>
	(0.97)	(0.77)	(1.41)	(1.20)	(1.14)	(1.02)	(1.35)
1995–96/1997–98	0.85	<u>0.69</u>		0.60	1.20		
	(0.81)	( <u>0.43</u> )		(1.13)	(1.32)		
1996–97/1997–98	0.82	0.70		0.73	0.76	_	_
	(0.84)	( <u>0.56</u> )		(0.94)	(1.15)		

TABLE 3 Ratio of Average CIELAB Color Differences to UOCS Standards (without parentheses) and BTB Standards (with parentheses) for Each Olive Variety at Different Stages of Ripeness and at Different Harvest Years<sup>a</sup>

<sup>a</sup>Statistically significant differences (Kolmogorov–Smirnov test, P < 0.05) for the groups indicated in the first column are underlined.Dashes correspond to cases in which the number of samples in one of the groups was less than 5 (e.g., for all groups of the Pico Limón variety, which were not included in this table).

between the groups pintona and ripe were not statistically significant for either the BTB (P = 0.390) or UOCS (P = 0.971) method. However, the differences were statistically significant (P < 0.05) for both color scales in the groups green–pintona, and green–ripe. This should warn us that the UOCS (as well as the BTB scale) would need to be improved for oils from the least ripe (green) olives. For the UOCS, there were statistically significant differences (P < 0.05) for only one variety (Arbequina, groups green–ripe), as described next.

Figure 2 shows the average and SD of CIELAB color differences  $(\Delta E^*_{ab})$  for olives from the eight varieties studied (Picual, Hojiblanca, Lechín, Manzanilla, Arbequina, Verdial, Picudo, and Pico Limón) using the BTB and UOCS methods. Figure 2 illustrates that very similar values were found for the average color difference and SD by the UOCS. However, this was not true of the BTB scale, where pronounced differences were found, for example, between the Lechín and Verdial varieties. For the UOCS, the average CIELAB color difference changed with olive variety within a narrow range (3.3-4.5 CIELAB units), in contrast to the wider range found for the BTB scale (5.4–14.7 CIELAB units). In general, the same was found for the SD with the UOCS and BTB (error bars in Fig. 2). It is noteworthy that with the nonparametric Wilcoxon tests for related samples, the differences between the BTB and UOCS methods were statistically significant (P < 0.05) for each of the eight olive varieties considered. The poorest results for the UOCS, compared with the BTB, were found for the varieties Arbequina and Pico Limón, although at a 95% confidence level the differences between the two scales were also statistically significant for these two olive varieties (P = 0.008and P = 0.016, respectively). Comparing the BTB and UOCS methods for the different varieties, we found that the ratio (BTB/UOCS) of the average CIELAB color difference was in the range of 1.3 (Arbequina) to 3.4 (Verdial) and that the ratio of the SD was in the range of 0.8 (Arbequina) to 3.3 (Verdial).

We also conducted a pairwise comparison of the eight olive varieties for both the UOCS and the BTB standards using the Kolmogorov–Smirnov tests with unrelated samples. The results showed that for the UOCS, there were no statistically significant differences (P > 0.05) between any two varieties. However, for the BTB there were significant differences (P < 0.05) between 10 pairs of varieties (36%): Picual–Arbequina, Picual–Verdial, Lechín–Verdial, Arbequina–Verdial, Hojiblanca–Lechín, Hojiblanca–Arbequina, Hojiblanca–Picudo, Manzanilla–Arbequina, Manzanilla–Verdial, and Verdial–Picudo.



**FIG. 1.** CIELAB color differences  $(\Delta E^*_{ab})$  for virgin olive oils from olives at three different stages of ripeness [green, pintona (medium-ripe), and ripe; see Table 1], using the bromthymol blue (BTB) and Uniform Oil Color Scale (UOCS) standards. Columns indicate average values, and error bars indicate SD.



**FIG. 2.** CIELAB color differences ( $\Delta F_{ab}^*$ ) for virgin olive oils from eight different varieties of olives (see Table 1) using the BTB and UOCS standards. Columns indicate average values, and error bars indicate SD. For abbreviations see Figure 1.

Figure 3 shows the average and SD of CIELAB color differences ( $\Delta E^*_{ab}$ ) for olives from four different harvests (1994–95 to 1997–98) using the BTB and UOCS methods. The average CIELAB color differences were in the range of 7.0–9.6 for the BTB and 3.8–4.7 for the UOCS. The SD of CIELAB color differences (designated by error bars in Fig. 3) were at least 1.7 times greater for the BTB than for the UOCS among the four harvests studied. From the nonparametric Wilcoxon tests for related samples, the differences between the BTB and UOCS at each of the four harvests were statistically significant (P < 0.001). For the BTB and UOCS at different harvests, the ratio (BTB/UOCS) of the average CIELAB color difference was in the range of 1.6 (harvest 1997–98) to 2.5 (harvest 1994–95), and the ratio of the SD was in the range of 1.7 (harvests 1996–97 and 1997–98) to 3.1 (harvest 1994–95).

The UOCS and BTB methods also were compared pairwise with respect to the four harvests using the Kolmogorov–Smirnov test for unrelated samples. For the UOCS, only one case showed statistically significant differences (P = 0.041), which corresponded to the comparison of harvests 1994–95 and 1996–97. However, for the BTB scale four pairs of harvests (67%) differed significantly (P < 0.05): 1994–95 and 1995–96, 1994–95 and 1996–97, 1994–95 and 1997–98, and 1995–96 and 1996–97.

Thus, as shown in Figures 1 to 3, the results achieved by the UOCS considering olive ripeness, variety, and harvest are worth noting. Unfortunately, the same cannot be said for the BTB scale. The UOCS was superior to the BTB in all cases, and this improvement was always statistically significant according to the Wilcoxon tests (P < 0.05).

Finally, we analyzed the performance of the UOCS and BTB for each of the eight olive varieties individually and compared the three stages of ripeness and the four harvests. Table 3 shows the ratio of average CIELAB color differences between the groups for each of the olive varieties for both the UOCS and BTB, as well as the cases in which the differences between the groups were statistically significant (P > 0.05) based on the Kolmogorov–Smirnov test. Table 3 does not present results for cases



**FIG. 3.** CIELAB color differences  $(\Delta E^*_{ab})$  for virgin olive oils from olives from four different harvests (see Table 1) using the BTB and UOCS standards. Columns indicate average values and error bars indicate SD. For abbreviations see Figure 1.

with fewer than 5 oil samples in one group, because the corresponding statistical results were not considered reliable. This happened, for example, in all the groups of the variety Pico Limón, which contained only 11 olive samples (see Table 1). In Table 3, differences that were not statistically significant (as well as values close to 1.0) are indicative of good performance by the respective color scale (UOCS or BTB). With respect to the degree of maturation, 7 cases registered significant differences with the BTB as opposed to 2 with the UOCS; however, with respect to harvest, 6 cases differed significantly for both the UOCS and BTB. For the UOCS, the most significant differences were found for the variety Hojiblanca.

Although the improvement of the UOCS over the BTB appears to be evident in terms of olive ripeness, variety, and harvest, no clear trends were discerned for efficient modification of the current UOCS method (6). Certainly, adding new standards to the current UOCS might offer a slight improvement of the results found for oils from the least ripe olives and also for olives of the Hojiblanca and Manzanilla varieties. However, it is doubtful whether more standards in the new UOCS would offset the minor numerical improvement achievable in color differences from real oil samples. Currently, we prefer to delay this last step in the development of a final color scale for virgin olive oils until further research with commercial virgin olive oil samples can be reported.

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