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Level of Agreement in Subjective Selection of Gingival Colour

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Abstract: *Background and Objectives*: Primary outcome: To assess the level of agreement between the objective and subjective methods for recording gingival colour in each area of the gingiva. Secondary outcome: To compare performance of the subjective visual method of gingival colour selection by a male observer and a female observer. *Materials and Methods*: A chromatic study was conducted on a total of 101 participants, in five gingival zones, from the free gingival margin to the mucogingival line, using a SpectroShade Micro spectrophotometer for the objective method and 21 ad hoc ceramic gingival shade tabs for the subjective method. A man and a woman of the same age, with the same amount of clinical experience in dentistry, selected the tab that most resembled the colour of participants' gingiva. The "chromatic error" was then assessed by calculating the colour difference (using the Euclidean and CIEDE2000 formulae) between the CIELAB coordinates of the shade tab selected and the objective coordinates of the gingiva. The unweighted Kappa coefficient was used to calculate the level of agreement between observers. *Results*: For the male observer, the mean chromatic errors varied between ΔEab* 10.3 and 13.1 units, while for the female observer, the mean errors varied between ΔEab* 11.1 and 12.8: these differences were not statistically significant. Similarly, no statistically significant differences were found between the mean chromatic errors for the five gingival zones in either the male operator ($p = 0.100$) or the female operator ($p = 0.093$). The minimum level of agreement (unweighted Kappa) between the observers ranged from 0.1 to 0.4. *Conclusions*: Subjective selection of gingival colour was very inaccurate, by both the male observer and the female observer, for any area of the gingiva, with no differences identified between them. The level of agreement between the observers was low. These findings suggest that gingival colour should not be determined using solely subjective methods, given that the chromatic errors significantly exceeded the clinical acceptability threshold for gingiva (4.1 units for ΔEab* and 2.9 units for ΔE00). Both observers showed a tendency to select gingival shade tabs that were redder and bluer than the objective colours.

Keywords: gingiva colour; agreement between objective and subjective methods; perception of colour gender

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1. Introduction

All forms of therapeutic rehabilitation performed in dentistry should aim to achieve a good level of dental functionality and aesthetics. To meet the second of these objectives, it is vital for the "white aesthetics" to be in harmony with the so-called "pink aesthetics" [1]. Several studies have noted that colour is the aesthetic factor in dentistry which patients deem most important [2–4], although it is also the factor that is most difficult to determine, convey, and reproduce accurately. Colour is a psychophysical sensation that results when the human visual system captures light reflected from an object [5]. Three factors influence this perception of colour [6]: the observer, the light source used, and the object, defined by its own chromatic information. Selecting dental colour by making a direct comparison

with the colour of the tooth in question—or the adjacent teeth, if missing teeth are being replaced—is the classic, most widely used methodology [7–9]. The subjectivity involved in the visual selection of colour in dentistry is well-documented [10–14], as is the lack of consistency, given the range of variables that have been seen to condition it, such as the following: the observer's experience, theoretical and practical training, gender, age, and emotional state, as well as ocular fatigue, ambient light and colour, etc. With the aim of reducing the errors involved in the traditional colour selection method and increasing the accuracy and reliability [14] of dental shade matches, objective electronic devices have come into use [15–18] which record dental colour using CIELAB colour coordinates and also provide the resulting colour as given in dental shade guides (Vita Classical or Vita 3D Master) [14,19]. Comparisons between visual selection and instrumental methods have suggested that the latter provides better results [20–23], although recent publications have recommended a combination of various methods [8,18,24,25].

There is no consensus in prior studies about whether there is any difference in the subjective perception of the dental colour space by men and women. Some have concluded that men and women perceive dental colour in a similar manner [11,26–32], while other articles [11,33–36] have identified statistically significant differences in favour of women's identification of colour.

To enable the quantitative study of colour, the Commission Internationale de l'Éclairage (CIE, International Commission on Illumination) has defined a colour space as a uniform, three-dimensional space of colour coordinates formed by three axes: (1) the L* coordinate, which corresponds to the vertical axis and refers to the lightness of the colour, from black when L* = 0 to white when L* = 100; (2) the horizontal a* coordinate—a positive value for a* indicates the amount of red, while the more negative the a* coordinate is, the more green is present; and (3) the horizontal b* coordinate—an increasingly positive value for b* indicates a larger amount of yellow, while an increasingly negative b* coordinate indicates a larger amount of blue [37]. In dentistry, the most frequently used formulae for quantitatively calculating the difference between two objects are the Euclidean formula (ΔEab*) [37] and the CIEDE2000 formula, whose purpose is to solve the problem of discrepancies between colour measurements performed with colorimeters and the human eye [15,38,39].

The aforementioned results apply solely to the dental colour space and have yet to be explored in the gingival colour space, due primarily to the lack of a gold standard for the gingival shade guides used in dentistry [40–42]. There is, therefore, a lack of knowledge about how subjective visual perception works within the "pink" colour space. There has been a similar lack of research on the difference in the size of the "chromatic errors" (the colour difference between the colour coordinates of the subjectively selected shade tabs and the objective colour coordinates provided with electronic devices) made by men and women in the gingival colour space and whether such errors have clinical relevance in practice.

This study had the following objectives: (1) to determine the chromatic errors made by a male observer and a female observer—both of whom were dentistry graduates—by calculating the difference between spectrophotometer readings and the colour they selected visually, using the Euclidean and CIEDE2000 formulae, and to assess whether these errors depended on the gingival zone (two papillae, free gingival margin, attached gingiva, and mucogingival line) and (2) to determine the CIELAB coordinates of the subjective gingival colour selections made by each observer in the various gingival regions and examine the level of agreement between them, identifying whether there were any statistically significant differences between the observers.

The null hypotheses of this study are the following: (1) the chromatic error made in subjective colour selection is the same in all five gingival zones; (2) the level of agreement between the two observers for the same gingival zone is acceptable and the chosen CIELAB coordinates are homogeneous.

2. Materials and Methods

2.1. Study Design

This cross-sectional study has been approved by the institutional Bioethics Committee (CBE.USAL-16/11/15). Prior to inclusion, all participants were verbally informed about the objectives and signed to confirm their participation. The criteria for enrolling subjects onto this study included: (1) having natural dentition at (1.1; 1.2; 2.1; 22); (2) falling within the age range of 18 to 90 years; (3) having healthy gingival tissue; (4) lacking melanin pigmentation; and (5) being of Caucasian race. Two dentists participated in the subjective selection of gingival colour as observers: (1) a woman of 24 years and (2) a man of 24 years, both of whom had theoretical and practical training on gingival colour in dentistry, no vision disorders, and two years of clinical experience.

2.2. Subjective Selection of Gingival Colour

For the subjective selection of gingival colour, 21 samples of HeraCeram® ceramic gingiva were used, manufactured by Heraeus Kulzer (Kulzer GmbH, Hanau, Germany). These samples were created using a silicone mould—Smile Line New Architect Wax-up Assistant Anterior: form B, large—the dimensions of which were approximately 10.6 mm × 61.6 mm × 33.2 mm. They were mounted on a Smile Line My Shade Guide in order of decreasing lightness (Figure 1). HeraCeram® colours G2, G4, G5, G6, G7, and G8 were used to make the 21 samples. These products were used as the six basic colours, on the basis of which 15 further samples were created by mixing the colours (the proportions of which were accurate to the nearest 25%), applying changes of 25% to obtain each new mixture. The mixtures used for the 15 additional samples were as follows: 75% colour G2 mixed with 25% colour G4; 50% G2 with 50% G4; 25% G2 with 75% G4; 75%G4 with 25% G5; 50% G4 with 50% G5; 25% G4 with 75% G5; 75% G7 with 25% G8; 50% G7 with 50% G8; and 25% G7 with 75% G8 [33].

Figure 1. Ad hoc gingival shade guide.

Three colour readings were conducted on each of the 21 shade tabs, after calibration, using the Spectroshade Micro (MHT Optic Research AG), which had undergone reliability and precision testing beforehand. All these subjective colour measurements were performed on the same dental cabinet in a neutral grey setting, with ambient light from Phillips daylight-colour fluorescent tubes (TDL 95/65), following the manufacturer's instructions. The resulting colour space values obtained were as follows: L*, a*, and b* (Table 1).

Table 1. Colour coordinates of the colour tabs produced using controlled ceramic mixtures.

Sample	Mixture Percentage	L*	a*	b*
1	100% G2	61.3	21.2	13.0
2	100% G4	36.8	33.5	12.4
3	100% G5	56.7	25.7	8.0
4	100% G6	60.2	20.9	13.6
5	100% G7	58.1	24.0	8.9
6	100% G8	48.9	29.9	13.8
7	75% G2 + 25% G4	56.8	22.4	17.4
8	50% G2 + 50% G4	45.2	32.8	12.5
9	25% G2 + 75% G4	57.7	24.7	10.9
10	75% G4 + 25% G5	57.0	22.6	17.7
11	50% G4 + 50% G5	55.9	28.4	6.2
12	25% G4 + 75% G5	41.5	33.0	16.4
13	75% G5 + 25% G6	44.7	32.6	13.7
14	50% G5 + 50% G6	63.9	16.4	21.6
15	25% G5 + 75% G6	37.7	32.7	12.9
16	75% G6 + 25% G7	45.0	33.0	15.1
17	50% G6 + 50% G7	61.7	18.6	18.0
18	25% G6 + 75% G7	45.0	32.7	16.6
19	75% G7 + 25% G8	43.1	35.2	19.4
20	50% G7 + 50% G8	39.3	34.1	15.8
21	25% G7 + 75% G8	60.1	21.2	11.1

2.3. Participants

Initially, 101 volunteers participated in this study. The colour of the five gingival zones of tooth number 1.1—mesial papilla, distal papilla, free gingival margin, the middle zone, and the upper zone (mucogingival line) of the attached gingiva—was recorded objectively (using spectrophotometry) (Figure 2). Next, the female operator subjectively selected (using our ad hoc gingival colour guide of 21 shade tabs) the tab she considered closest to the objective gingival colour in each of the five gingival zones. Subsequently, in a session on a different date, the male operator conducted subjective selection of gingival colour, using the same gingival colour guide and process described above, in the five gingival zones shown in Figure 2. His selections were made for 47 of the initial 101 participants, given that 54 participants did not attend the second session. Therefore, it was only possible to compare the subjective selections made by the two operators for these 47 participants. All the colour readings were made on the same dental cabinet and in the same ambient conditions.

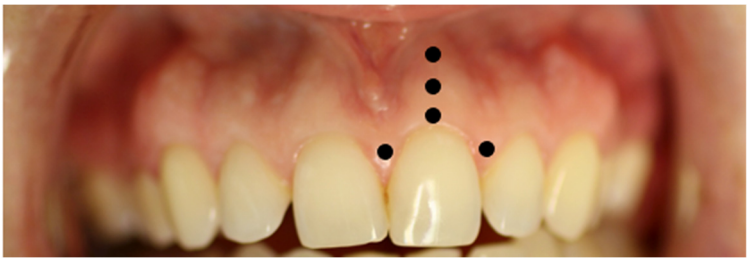

Figure 2. Areas in which gingival colour readings were taken.

2.4. Objective Gingival Colour Readings

For the objective colour readings, a spectrophotometer was used: the "Spectroshade Micro" (MHT Optic Research, Niesderhasli, Switzerland), which has a configuration of 45°/0°, corresponding to lighting and recording, respectively. Three colour readings were taken per participant in each of the five areas of analysis, resulting in a total of 15 spectrophotometric readings per participant. The aim of taking multiple readings was to calculate the arithmetic mean of the colour coordinates (L*, a*, and b*) in each gingival location. All the colour readings were made by the same female operator. The methodology used in this study is illustrated in Figure 3.

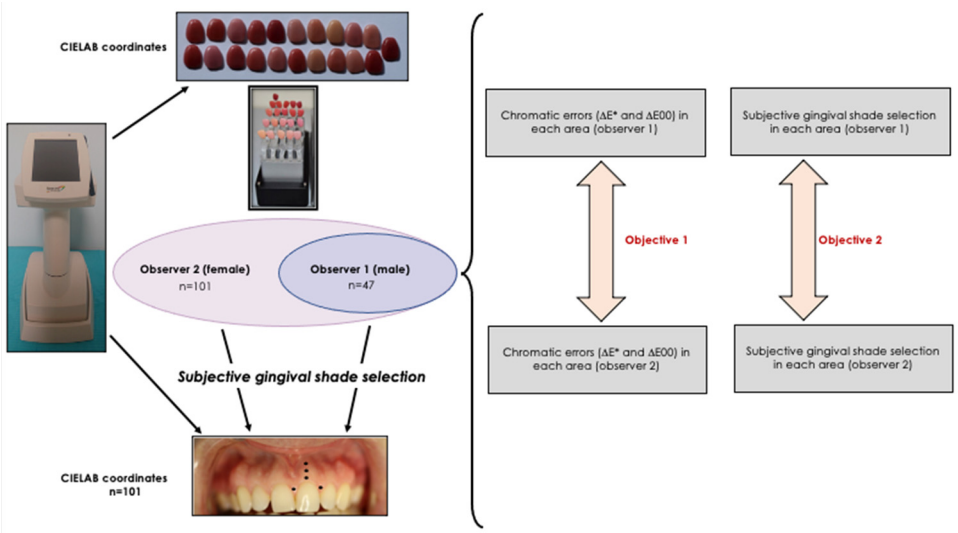

Figure 3. Outline of the methodology used to meet this study's objectives.

"Chromatic error" is defined in this study as the colour difference between the CIELAB coordinates of the shade tab selected subjectively and the CIELAB coordinates provided via the objective spectrophotometric method. The chromatic error was calculated using two formulae: the Euclidean formula (ΔEab*) and the CIEDE2000 formula (ΔE00).

Euclidean formula (ΔEab*):

$$\Delta Eab^* = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

in which ΔL*, Δa*, and Δb* are the differences in the respective colour coordinates [37].

The CIEDE2000 colour difference formula was created on the basis of the Euclidean formula, but it involves a more complex series of mathematical operations [15,38,39]. It is characterised by the use of weighting functions and parametric factors to correct the effects of luminosity, chroma and hue, and a rotation term that corrects the interaction between chroma and hue in the blue region:

$$\Delta E_{00} = \sqrt{\left(\frac{\Delta L^*}{k_L S_L}\right)^2 + \left(\frac{\Delta C^*}{k_C S_C}\right)^2 + \left(\frac{\Delta H^*}{k_H S_H}\right)^2 + R_T\left(\frac{\Delta C^*}{k_C S_C}\right)\left(\frac{\Delta H^*}{k_H S_H}\right)}$$

In CIEDE2000 computation, under baseline conditions, all the parametric factors are set to 1 [43].

2.5. Statistical Analysis

One-way analysis of variance (ANOVA) was performed to determine if statistically significant differences existed in the means of the colour differences between the coordinates of the chosen shade tabs and the objective coordinates of the patients' gingiva for the five gingival regions. Unweighted kappa coefficients of agreement between the choices made

by the male observer and the female observer were calculated and interpreted according to Landis and Koch (1977): 0 to 0.2 signifying slight agreement; 0.2–0.4: fair agreement; 0.4–0.6: moderate agreement; 0.6–0.8: substantial agreement; and >0.8 almost perfect agreement [44]. The proportions of each shade tab selected by the male observer and the female observer were compared with the χ^2 test of homogeneity of proportions. Finally, the *t* test for paired samples was performed to determine if there were any statistically significant differences between the "errors" in the chromatic selections of the two observers.

3. Results

Table 2 shows the means and standard deviations of the colour differences—calculated using the Euclidean formula (ΔEab*) and the CIEDE2000 formula (ΔE00)—between the colour coordinates of the shade tab chosen by the male observer and the patient's objective colour coordinates (recorded using the spectrophotometer).

Table 2. Means (standard deviations) of the colour differences between the colour coordinates of the shade tabs chosen by the male observer and the objective colour coordinates of the participants' gingiva (*n* = 47).

"Chromatic Errors" of the Male Operator		
	CIELAB (ΔEab*)	**CIEDE2000 (ΔE00)**
Mesial papilla	12.08 (5.30)	9.48 (4.99)
Distal papilla	10.29 (4.92)	8.96 (4.95)
Free gingival margin	13.12 (5.53)	10.30 (5.09)
Middle zone	11.96 (4.10)	9.05 (3.69)
Mucogingival line	11.87 (4.80)	8.64 (5.05)

There were no statistically significant differences between the mean values of the colour differences with both formulae, in the five gingival zones (CIELAB: F = 1.972, $p = 0.100$; CIEDE2000: F = 0.848; $p = 0.496$).

Table 3 shows the means and standard deviations of the differences between the colour coordinates of the shade tab chosen by the male observer and each objective colour coordinate of the patients' gingiva. There were statistically significant differences between the three coordinates in the five gingival areas. In all of these areas, the a* coordinate had the lowest level of "fit" between the subjective and objective readings. In contrast, the L* coordinate had the best "fit".

Table 3. Means (standard deviations) of the differences between each colour coordinate of the shade tabs selected by the male observer and the objective colour coordinates of the patients (*n* = 47).

	ΔL*	**Δa***	**Δb***	***p* Value**
Mesial papilla	0.79 (8.83)	3.61 (6.32)	−3.23 (5.88)	<0.001
Distal papilla	0.98 (8.55)	4.75 (5.53)	−2.30 (5.68)	<0.001
Free gingival margin	−1.06 (9.28)	4.59 (6.97)	−3.51 (6.02)	<0.001
Middle zone	−2.37 (7.44)	6.26 (5.43)	−3.10 (4.75)	<0.001
Mucogingival line	2.86 (7.51)	6.60 (4.88)	−3.05 (4.94)	<0.001

Figure 4 shows the differences in each colour coordinate for the 47 patients for whom the male observer selected the shade tab he considered closest to their gingival colour, by gingival zone.

Figure 4a shows the differences in the L* coordinate (lightness) between the selected shade tab and the gingiva, by gingival zone. Positive differences in L* (ΔL*) indicate that the chosen gingival shade tab was lighter than the gingiva, while the opposite is true for negative differences in L*. As can be seen in this figure, the number of data points located above the ΔL* = 0 line is practically the same as the number of data points located below it for the mesial and distal papilla (25 vs. 22 and 24 vs. 23, respectively); for the free gingival

margin and the middle zone, there are fewer data points above the line than below (21 vs. 26 and 18 vs. 29, respectively), indicating that the male observer selected more shade tabs that were darker than the gingiva than tabs that were lighter; finally, at the mucogingival line, the number of data points above the L* = 0 line is greater than the number below it (27 vs. 20), showing that the male observer chose more tabs that were lighter than the gingiva than tabs which were darker in that zone. However, the proportion of data points for which ΔL* > 0 does not differ significantly in the different gingival zones ($\chi^2 = 4.257$, $p = 0.372$).

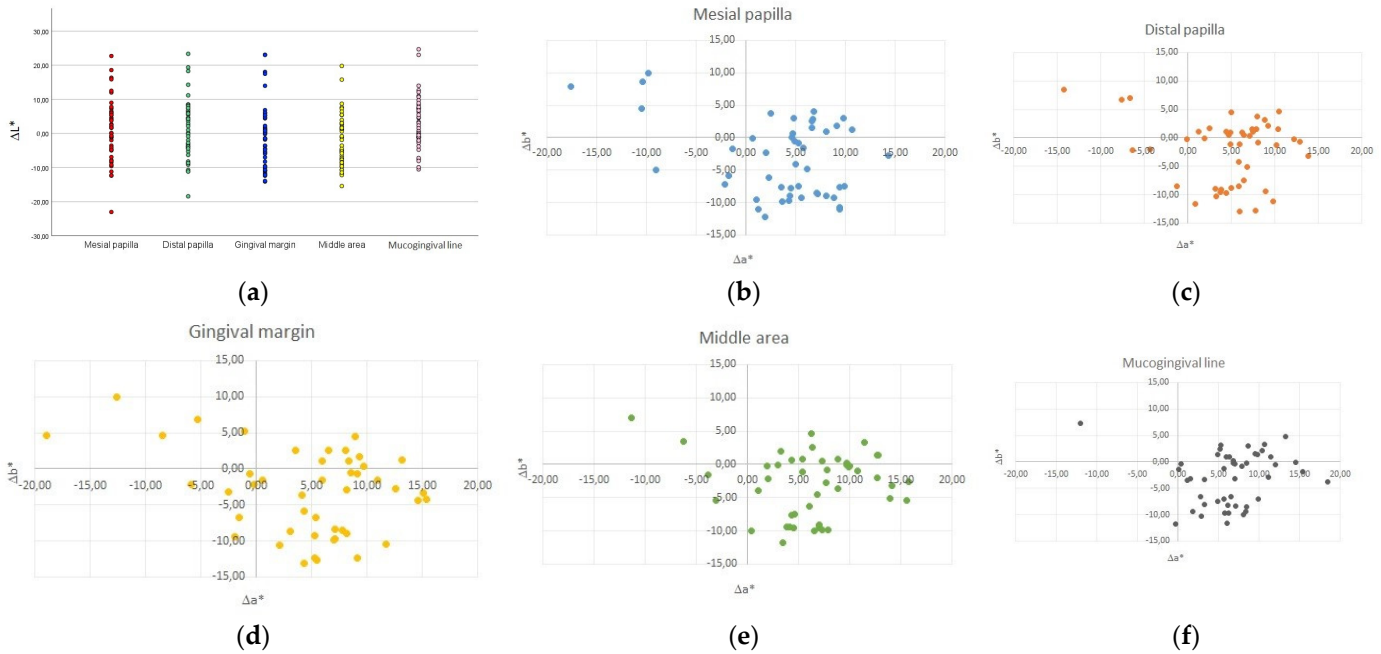

Figure 4. Direction and extent of CIELAB colour coordinates (male observer). (**a**) ΔL* distribution; (**b**–**f**) Δa* − Δb* in the mesial papilla, distal papilla, free gingival margin, middle zone, and mucogingival line, respectively.

Figure 4b–f illustrate the differences between the a* coordinate and the b* coordinate of the shade tab selected and the gingiva, for each gingival zone. A data point located in the first quadrant (Δa* > 0 and Δb* > 0) shows that the tab selected contains more red (less green) and more yellow (less blue) than the gingiva; a data point located in the second quadrant (Δa* > 0 and Δb* < 0) shows that the tab selected contains more red (less green) and less yellow (more blue) than the gingiva; a data point located in the third quadrant (Δa* < 0 and Δb* < 0) shows that the tab selected contains less red (more green) and less yellow (more blue) than the gingiva; finally, a data point located in the fourth quadrant (Δa* < 0 and Δb* > 0) shows that the tab selected contains less red (more green) and more yellow (less blue) than the gingiva. As these figures show, in all zones, there is a larger number of data points in the second quadrant than in the others, indicating that most of the shade tabs selected by the male observer contained more red and less yellow than the participants' objective colour coordinates. There is no significant difference between the five gingival zones when it comes to the proportion of data points in each quadrant ($\chi^2 = 12.728$, $p = 0.389$).

Table 4 shows the means and standard deviations of the colour differences—calculated using the Euclidean formula (ΔEab*) and the CIEDE2000 formula (ΔE00)—between the colour coordinates of the shade tab chosen by the female observer and the patient's objective colour coordinates. With 101 participants and five gingival zones, a total of 505 colour readings were taken.

Table 4. Means (standard deviations) of the colour differences between the colour coordinates of the shade tabs chosen by the female observer and the objective colour coordinates of the participants' gingiva (n = 101).

"Chromatic Errors" of the Female Operator		
	CIELAB (ΔEab*)	**CIEDE2000 (ΔE00)**
Mesial papilla	12.80 (5.31)	10.22 (4.94)
Distal papilla	12.05 (4.82)	9.02 (4.55)
Free gingival margin	11.88 (4.84)	9.27 (4.20)
Middle zone	11.11 (4.46)	8.61 (4.04)
Mucogingival line	12.76 (5.31)	10.21 (5.69)

There were no statistically significant differences between the mean values of the colour differences, in the five gingival zones (CIELAB: F = 2.004, p = 0.093; CIEDE2000: F = 2.374; p = 0.051).

Table 5 shows the means and standard deviations of the differences between the colour coordinates of the shade tab chosen by the female observer and the objective colour coordinates of the patients' gingiva. There were statistically significant differences between the three coordinates in the five gingival areas. In all of these areas, the a* coordinate had the lowest level of "fit" between the subjective and objective readings, in contrast with the L* coordinate, for which the fit was best in all the areas, except at the mucogingival line.

Table 5. Means (standard deviations) of the differences between each colour coordinate of the shade tabs selected by the female observer and the objective colour coordinates of the participants (n = 101).

	ΔL*	**Δa***	**Δb***	***p* Value**
Mesial papilla	−1.21 (9.97)	4.27 (6.75)	−1.52 (5.16)	<0.001
Distal papilla	0.84 (8.20)	5.36 (6.16)	−2.21 (5.47)	<0.001
Free gingival margin	−1.22 (8.08)	4.88 (5.93)	−3.31 (5.38)	<0.001
Middle zone	1.02 (7.81)	4.11 (5.37)	−3.11 (5.19)	<0.001
Mucogingival line	5.44 (9.05)	4.43 (5.83)	−1.42 (4.98)	<0.001

When comparing these results for divergence from the objective colour coordinates for the man and the woman, no statistically significant differences were found in any gingival area, except the middle zone, in which there was a statistically significant difference between ΔL* (−2.37 vs. 1.02, p = 0.014) and Δa* (6.26 vs. 4.11, p = 0.025) for the male and the female observer.

Figure 5 shows the differences in each colour coordinate for the 101 patients for whom the female observer selected the shade tab she considered closest to their gingival colour, by gingival zone.

As can be seen in Figure 5a, the number of data points located above the ΔL*=0 line is practically the same as the number of data points located below it for the mesial and distal papilla (50 vs. 51 and 53 vs. 48, respectively); for the free gingival margin, there are fewer data points above the line than below (46 vs. 55), indicating that the female observer selected more shade tabs that were darker than the gingiva than tabs that were lighter in that zone; finally, for the middle zone and the mucogingival line, the number of data points above the line is greater than the number below it (62 vs. 39 and 74 vs. 27, respectively), showing that the female observer chose more tabs that were lighter than the L* coordinates provided via the spectrophotometer in these zones. For the female observer, there were significant differences in the proportion of data points for which ΔL* > 0 in the five different gingival zones (χ^2 = 20.136, p < 0.001).

Figure 5b–f show that, in all the gingival zones, there is a larger number of data points in the second quadrant than in the others, indicating that most of the shade tabs chosen by the female observer contained more red and less yellow than the gingival colours of the

participants. The proportion of data points in each quadrant did not differ significantly between the five gingival zones (χ^2 = 12.147, p = 0.434).

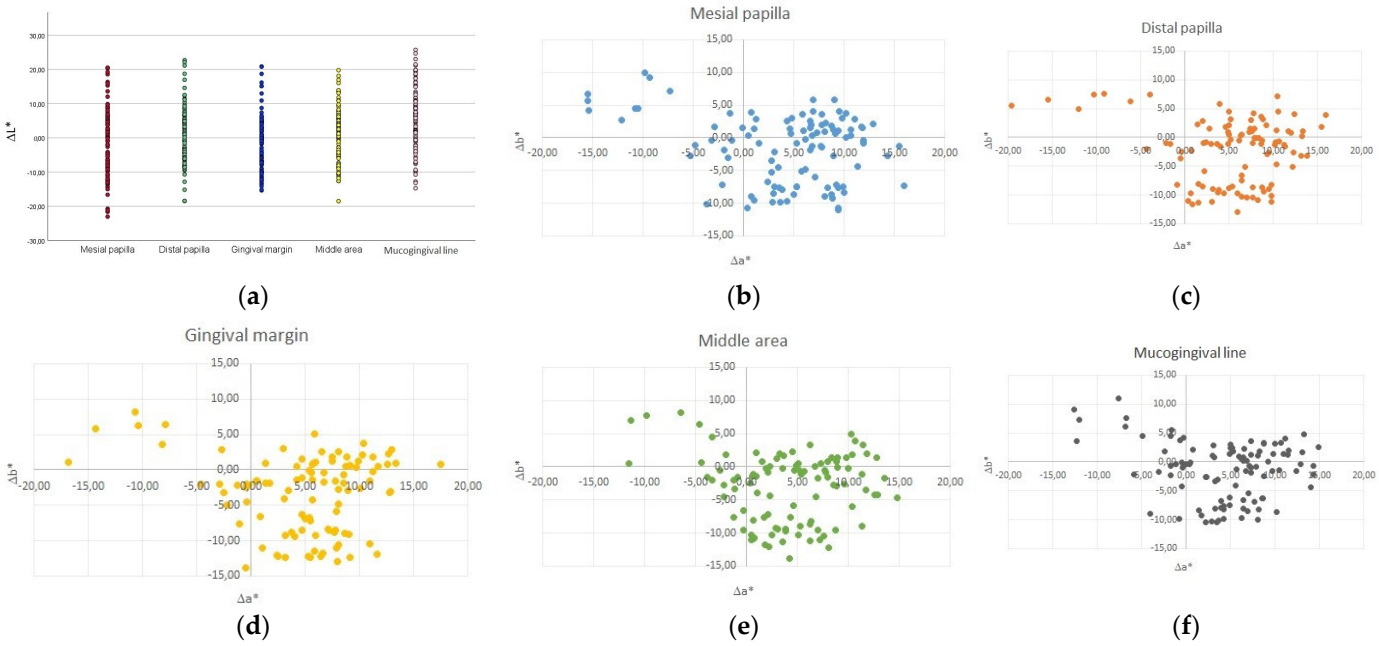

Figure 5. Direction and extent of CIELAB colour coordinates (female observer). (**a**) ΔL* distribution. (**b**–**f**) Δa* − Δb* in the mesial papilla, distal papilla, free gingival margin, middle zone, and mucogingival line, respectively.

Table 6 shows the unweighted Kappa coefficients of agreement between the choices made by the male observer and the female observer for the 47 patients for which both made shade tab selections, with colour readings in the five gingival zones (a total of 235 objective and subjective colour readings). Except for the free gingival margin, where there was a level of alignment between their readings that was not significant, the level of agreement between the observers was low in the other gingival zones [44].

Table 6. Unweighted Kappa coefficients of agreement between the subjective choices of the female and male operator (n = 47).

Mesial Papilla	Distal Papilla	Free Gingival Margin	Middle Zone	Mucogingival Line
0.383	0.338	0.103	0.219	0.242

Of the 235 selections made by the male observer (47 participants × 5 gingival areas), the most frequently chosen tabs were number 11 (77 times, 32.77%) and 12 (62 times, 26.38%). These two tabs were also those chosen most frequently in the 505 selections made by the female observer: number 11 was chosen 136 times (26.93%) and 12 was chosen 121 times (23.96%). If we consider the entire set of gingival shade tabs (n = 21), there were no significant differences in the selection percentages for each shade tab between the male operator and the female operator (χ^2 = 29.714; p = 0.056).

The comparison of the chromatic error in each observer's subjective selections was made for the 47 participants for whom both the male and female operator made shade tab selections (attempting to identify the closest shade to participants' gingival colour). Table 7 shows the means and standard deviations of the colour differences between the gingiva and the shade tab selected, as well as the p value for the comparison of means in paired samples.

The only statistically significant difference (p = 0.042) between the chromatic error made by the male operator and the female operator was at the mucogingival line, using

the CIEDE2000 formula for the difference between the colour coordinates of the gingival shade tabs and the colour readings of the spectrophotometer. No statistically significant differences were found in the other locations.

Table 7. Means (standard deviations) of the colour differences between the gingiva and the shade tab selected by the male observer and the female observer, by gingival zone.

	CIELAB (ΔEab*)			CIEDE2000 (ΔE00)		
	Male	**Female**	***p***	**Male**	**Female**	***p***
Mesial papilla	12.08 (5.30)	12.63 (5.16)	0.386	9.48 (4.99)	9.98 (4.84)	0.400
Distal papilla	10.29 (4.92)	11.56 (4.80)	0.076	8.96 (4.95)	8.74 (4.64)	0.776
Free gingival margin	13.12 (5.53)	11.92 (5.03)	0.146	10.30 (5.09)	9.27 (4.62)	0.183
Middle zone	11.96 (4.10)	11.01 (4.97)	0.234	9.05 (3.69)	8.46 (4.44)	0.410
Mucogingival line	11.87 (4.80)	13.02 (5.34)	0.154	8.64 (5.05)	10.38 (5.71)	**0.042**

4. Discussion

4.1. Null Hypothesis

The considerable chromatic errors made in the subjective selection of gingival colour, calculated using both formulae, did not differ in distinct zones of the gingiva, for either the male observer or the female observer. Therefore, the first null hypothesis cannot be rejected. Nor were differences found between the chromatic errors made by the two observers in any of the gingival zones, except the attached gingiva, although this difference only appeared when evaluating chromatic error using the CIEDE2000 formula. The level of agreement between the observers was low. The second null hypothesis of this study can therefore only be partially rejected.

4.2. Electronic Device

Spectrophotometers have been said to have a precision of over 96% [45] and an accuracy of 66.8% to 92.6% [46] in the dental colour space, since they were initially designed and used to quantify dental colour. These electronic devices may also have the potential to evaluate gingival colour in a reliable manner [47]. Sala L et al. [48] found that the repeatability and reproducibility of colour readings with spectrophotometers on gingival tissue were almost perfect (ICC > 0.9), whereas the results of Staedt and colleagues displayed a limited degree of reproducibility [47]. There are issues—such as gingival curvature, certain characteristics of the gingival surface, the degree of translucency, contact with the gingival tissue, the phenomenon of edge loss, and the failure to adequately maintain electronic devices for colour recording—which can alter the chromatic results obtained [45]. To minimise the impact of these phenomena, a spectrophotometer with a "wide window" was used. Using instrumental methods results in a greater level of agreement in the dental shade match than when colour is recorded using visual methods [49], in addition to a higher level of precision and reproducibility [16]. In quantitative terms, published figures range from 30% to 67% alignment between subjective visual comparisons and spectrophotometric results in the dental context [24,50,51], while other authors have not identified any relationship between the two methods [52,53]. The lack of standardised gingival shade tabs means that the spectrophotometer can only provide colour coordinates: this is why it has not been possible to provide results in the form of percentages of agreement with gingival shade guide terms and why the differences have been shown in terms of the CIEDE2000 and traditional Euclidean formulae. In this study, all the colour differences identified between the results of visual selection and spectrophotometry fall significantly above the published clinical acceptability threshold for the gingival colour space (4.1 units for CIELAB* and 2.9 units for the CIEDE2000 formula) [54], as was the case in Igiel and colleagues' research [51]. Another study, focused on the dental colour space, identified a difference of 7.35 units for ΔEab* between the visual comparison and the spectrophotometric readings [50]. It is important to note that the two formulae (CIELAB* and CIEDE2000) show a higher level of agreement in the gingival colour space [55].

4.3. Subjective versus Objective Metodology

Both observers most frequently selected shade tabs containing more red and less yellow ($\Delta a^* > 0$ and $\Delta b^* < 0$) than the gingival colour of participants in all five zones of the gingiva, which enables us to conclude that visually determining coordinates a* and b* is what presents the greatest challenge to observers. The results obtained show that the female observer more frequently chose gingival shade tabs that were darker than the gingiva at the free gingival margin, while she more often chose tabs that were lighter than the gingiva in the middle zone and the attached gingiva. The subjective values for the a* coordinate were higher than the objective values for this coordinate, for both the male and female observers. Both, therefore, tended to choose tabs that were redder than the gingiva. The opposite was true for the b* coordinate. Subjective values for the b* coordinate were lower than the objective values for both observers, meaning that they tended to select tabs containing less yellow than the gingiva.

Using the Kappa coefficient, Gómez Polo et al. analysed the extent of agreement between visual selection of dental colour by an operator using the Toothguide 3D Master and the objective method of spectrophotometry. Their findings showed that correlation between the objective and subjective methods was greatest for the lightness of colour (Kappa 0.65), followed by hue (Kappa 0.43), and lastly by chroma (Kappa 0.35) [56]. The Kappa values provided by similar studies have also pointed to a moderate level of agreement between three observers in subjective dental colour selection [57]. However, for gingival tissue, the percentages of agreement between the observers under study was lower. This demonstrates that the male and female operators differed in the gingival shade tabs that they subjectively selected (Kappa 0.130 to Kappa 0.383), but neither of them was more "correct" in their choices than the other: each chose as incorrectly as the other, hence the large chromatic errors. This enables us to surmise that subjective colour selection is a very weak method in any area of the gingiva. It is important to remember that the Kappa index is limited by its dichotomous nature (it calculates whether two things are the same or not) and does not identify nuances such as the selection of samples that are chromatically very similar, instead classifying them as completely different. It rates both large and small chromatic errors as exactly the same.

In this study, the observers had the same level of experience and training in colour selection, to ensure that these factors did not have a notable effect. For some authors, experience is not a relevant factor in dental colour selection [11,32,58], while others consider it an important element of the process [59].

The poor results obtained in visual selection of gingival colour—worse than those published in similar studies on dental colour [18,24,50]—could be due to a variety of factors, the most important of which may be the level of training. It is worth considering the potential "subliminal effect" of the theoretical and practical training offered in dentistry degrees, which include more content on the white dental tissue than the pink gingival tissue. Moreover, subjective selection of dental colour is a process that is conducted on almost a daily basis in clinical dental practice—for fillings, monitoring whitening treatments, and prosthetic restorations—while subjective selection of gingival colour is not. It follows that, for dental professionals, their eyes have undergone less training in the gingival colour space. Nor has agreement been reached about any differences related to gender in gingival colour perception: in the present study, there were no differences between the observers, except at the mucogingival line, while in other publications female observers have been shown to be more accurate [33]. It is important to underline that the results of this study are limited to comparing the subjective selections of one man and one woman and cannot be extrapolated to the male and female genders in general. However, no significant differences were found in the chromatic errors made by the two observers in any of the five zones, so it is possible to conclude that the female observer was not more accurate (making better visual colour selections) than the male observer. There are factors that are specific to the gingiva, such as translucency, rugosity, surface characteristics, and fluorescence, which should be taken into consideration. On this basis, a separate line of chromatic training could

be developed, which should be considered to ensure that dentistry professionals receive training in subjective selection of gingival colour, thereby improving results. According to a recent systematic review [31] on the controversial topic of the role of gender in dental colour selection, the lack of methodological homogeneity may explain the discrepancies in the results obtained.

4.4. Future Investigations

The essential role of initial studies in the research process is widely accepted. This initial phase evaluates the methodological and procedural aspects of what will subsequently become a larger-scale study, the planning, execution, and communication of which must be rigorous. Publication of preliminary studies such as this one is important, due to their pedagogical function as regards identifying and overcoming errors in the development of a research project. It would be useful to explore and reproduce the methods that have already been published on dental colour, applying them to the gingival colour space in order to compare results and generate new working hypotheses. However, it should be noted that the inclusion of only one man and one woman in this study prevents generalisation of its findings on gender differences. Future research should aim to include a more diverse and representative sample to provide a more comprehensive understanding of the potential variations in gingival colour selection between the genders. Expanding the sample size for both participants and observers, as well as exploring the consistency of the subjective visual selections at different points in time, may help us learn more about the little-studied phenomenon of gingival colour perception. However, the main limitation of comparisons with previous studies is the fact that the vast majority of the research examined the dental colour space rather than the gingival colour space, and there is no evidence to date that the findings presented can be extrapolated to that setting. Dentists should not use the subjective visual method alone when selecting gingival colour, given that it results in clear chromatic errors.

5. Conclusions

Within the limitations of this preliminary study, we can conclude the following: (1) The "chromatic errors" made in the visual selection of gingival colour are chromatically inacceptable (over 4.1 units for ΔEab* and 2.9 units for ΔE00) for both the female observer and the male observer in the five gingival zones. Lightness is the colour coordinate in which the chromatic fit between subjective and objective results is best for both observers. (2) The level of agreement between the two observers is low, without significantly better visual gingival selections by the female observer having been identified.

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