ORIGINAL RESEARCH

Tooth Shade and Blood Type: A Descriptive Cross-sectional Study in Tunisia

Imen Kalghoum[1], Ilhem Ben Othmen[2], Emna Boudabous[3], Leyla Ben Salem[4], Dalenda Hadyaoui[5]

Abstract

Aim: The aim of the study was to assess the prevalence of tooth shade and its correlation with blood type.

Materials and methods: This study analyzed 312 blood donors at the university Hospital in Monastir between November 2021 and June 2022. Both male and female subjects were included, with ages ranging from 18 to 60 years old. Patients with certain dental conditions or habits were excluded from the study. The study recorded various information about blood donors, including gender, age, governorate of origin, tooth shade, and blood type. The tooth shade values were recorded using A–D shade guide. Data input and tabulation were carried out using Microsoft Excel 2016 and SPSS (version 25.0).

Results: The study included 312 participants, with 85.58% males and 14.42% females. Tooth shade value B was the most prevalent (43%), and D was the least prevalent (7%). The statistical analysis showed that there was no significant link between tooth shade and blood type. However, there were three statistically significant categories: Blood type B/Tooth shade B, Blood type O/Tooth shade C, and Blood Type O/Tooth shade D.

Conclusion: The study examined the link between tooth color and blood type but did not find a significant link. However, significant values were found in different subgroups. A wider selection of subjects and a more rigorous measurement equipment might lead to more favorable results.

Clinical significance: By considering the patient's blood type alongside other relevant factors, clinicians can enhance the accuracy and precision of tooth shade selection, resulting in harmonious and natural-looking dental restorations. This approach improves patient satisfaction and acceptance.

Keywords: Age, Blood type, Color, Correlation, Cross-sectional study, Gender, Investigation, Tooth shade.

The Journal of Contemporary Dental Practice (2023): 10.5005/jp-journals-10024-3538

Introduction

In modern society, esthetics has gained significant importance, leading to a growing demand for esthetic dental restorations. As a result, it has become increasingly crucial to provide restorations that are indistinguishable from natural teeth. Among the factors influencing esthetics, tooth shade plays a vital role. The selection of an appropriate tooth shade is a key determinant in patients' perception of esthetics and their acceptance of prosthetic treatments.[1]

The presence of natural teeth greatly facilitates the selection of tooth shade. However, this process is influenced by various factors.[2] Factors such as the clinical expertise of the operator, the lighting conditions under which the shade is evaluated, and the shade guide system used all play significant roles in the shade selection process.[3]

Tooth staining can also significantly alter tooth shades. Stains can originate from various sources, both extrinsic and intrinsic. Extrinsic staining may result from factors, such as diet, smoking, xerostomia (dry mouth), and the presence of restorations. On the other hand, intrinsic staining can be caused by congenital defects of enamel or dentin, such as amelogenesis and dentinogenesis imperfecta, as well as environmental factors, such as tetracycline staining, traumatic injury, dental caries, and the natural aging process.[4-6]

The selection of tooth shade can be challenging for clinicians, particularly when there is no natural tooth available as a reference. In such cases, the process becomes subjective, leading researchers to explore methods to address this issue. Previous studies have investigated alternative characteristics of patients that can be utilized for tooth shade selection in the absence of a reference tooth.[7–9]

[1,2,3,5]Department of Fixed Prosthodontics, Research Laboratory of Occlusodontics and Ceramics, University of Monastir, Tunisia

[4]Dentist, Graduate from the Faculty of Dentistry of Monastir, Tunisia

Corresponding Author: Imen Kalghoum, Department of Fixed Prosthodontics, Research Laboratory of Occlusodontics and Ceramics, University of Monastir, Tunisia, Phone: +21695143268 e-mail: drkalghoumimen@gmail.com

How to cite this article: Kalghoum I, Othmen IB, Boudabous E, *et al*. Tooth Shade and Blood Type: A Descriptive Cross-sectional Study in Tunisia. J Contemp Dent Pract 2023;24(8):560–565.

Source of support: Nil

Conflict of interest: None

Various factors have been considered in previous research, including an individual's eye color, skin tone, hair color, as well as demographic factors such as sex and age. These characteristics have shown potential correlations with tooth shade selection.[7–12] Additionally, since blood type is a hereditary trait, it is possible that certain blood types may be associated with particular tooth shades.

Blood type is determined by specific antigens present on the surface of red blood cells. These antigens are inherited from parents and are part of a person's genetic makeup. There are four main blood types: A, B, AB, and O. Each blood type is determined by the presence or absence of specific antigens.[13,14]

Tooth shade, on the other hand, is influenced by various factors, including genetics, lifestyle, and dental hygiene. The natural color of teeth is determined by the composition and thickness of the enamel, the outer layer of the teeth. Genetics play a role in

Table 1: Example of a filled out investigation grid

Pouch number	*Gender*	*Age*	*Governorate*	*Tooth shade*	*Blood type*
0082xx	F	31	Mahdia	A1	A+
0082xx	M	26	Monastir	B2	O+

determining the thickness and translucency of the enamel, which can affect tooth color.[15–19]

While there have been studies investigating the relationship between genetics and tooth color, the focus has mainly been on genes associated with the production of proteins that affect enamel formation and pigmentation. For example, variations in genes responsible for the production of proteins such as amelogenin and enamelin can potentially impact tooth color. In particular, Amelogenesis Imperfecta (AI) is a group of hereditary enamel defects that affect the formation and structure of tooth enamel. In AI, variations or mutations in genes associated with enamel formation can lead to abnormal enamel development, resulting in tooth color changes and other dental abnormalities.[20,21]

Therefore, the objective of this study was to assess the prevalence of tooth shades and investigate any potential correlations between tooth shade and blood type which allow clinicians to enhance their ability to select appropriate tooth shades and create esthetically pleasing results for their patients.

Materials and Methods

This descriptive cross-sectional study involved the analysis of 312 blood donors. A simple random sampling method was used for data collection. The sample size was calculated using a G Power software considering a power of 90% and a two-sided level of significance of 5%. The minimum sample size required was found to be 255.

The study was conducted at the blood donation center of Fattouma Bourguiba Hospital in Monastir, and data collection lasted for 8 months between November 2021 and June 2022. The study participants were selected through random sampling, encompassing both male and female subjects within the age range of 18 to 60 years. Participants were divided into two groups: group 1 included the individuals in the age 18–35 years and group 2 included individuals from 36 to 60 years old.

Patients were excluded from the study if they had anterior prosthetics, antecedents of whitening, central incisor caries, color or structure anomalies, mediocre hygiene, or if they were heavy smokers.

A detailed grid was created to record various information about blood donors, including gender, age, governorate of origin, tooth shade, and blood type as given in Table 1. The tooth shade values were recorded using the Ivoclar Vivadent A–D shade guide, in natural daylight after each patient had concluded their blood donation. The blood donors each had a corresponding pouch number that was taken down on the day of the investigation, allowing the determination of their corresponding blood type upon later consultation. To ensure accurate tooth shade comparisons, quick assessments (within 5 seconds) were made to avoid visual accommodation. The tooth shade guide was placed under the lip, parallel to the tooth, and in the same plane to avoid distortion. Before comparison, the eye focused on a gray-blue surface to balance color sensors and eliminate afterimages. Female participants removed lipstick, wore neutral-colored attire, and a gray apron to minimize clothing influence. A single researcher conducted data collection for standardization.

Blood type is typically assessed using blood typing tests. There are several blood typing systems, but the ABO system is the most common.

The ABO blood typing system categorizes blood into four types: A, B, AB, and O. This classification is based on the presence or absence of specific antigens on the surface of red blood cells. The blood typing process involves mixing a small sample of blood with antibodies that are specific to the A and B antigens. The reaction between the blood sample and the antibodies determines the blood type. For example, if the blood cells agglutinate (clump) in the presence of anti-A antibodies, the blood type is determined as A.

Statistical Analysis

The study utilized Microsoft Excel 2016 and SPSS (Statistical Package for the Social Sciences) version 25.0 as tools for data input and tabulation.

A level of significance of 5% was retained for this study. In statistical hypothesis testing, the level of significance represents the threshold at which we will reject the null hypothesis. In this study, the null hypothesis would state that there is a link between a person's blood type and their tooth shade value. A 5% significance level means that we are willing to accept a 5% chance of making a type I error (rejecting the null hypothesis when it is true).

The collected data were analyzed using the Chi-square test, which is an inferential statistical analysis method. The Chi-square test is specifically designed to assess the presence of a significant association between two categorical variables, in this case, tooth shade and blood type. It compares the observed frequencies of the variables across different categories with the expected frequencies, assuming that the variables are independent.

By calculating the Chi-square statistic and comparing it to a critical value, the test determines if the variables are independent or if there is a significant association between them.

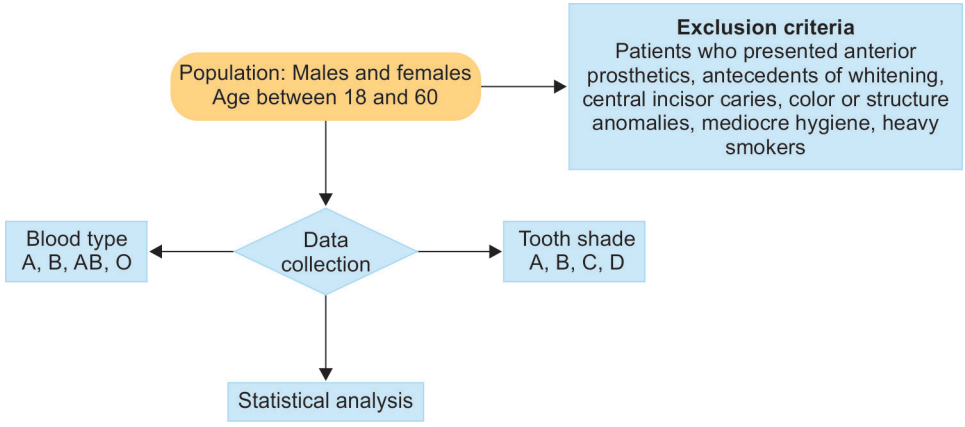
The results were displayed using a table in order to highlight the level of significance of each category independently as well as the level of significance for the overall analysis and visualized via a histogram and pie charts.

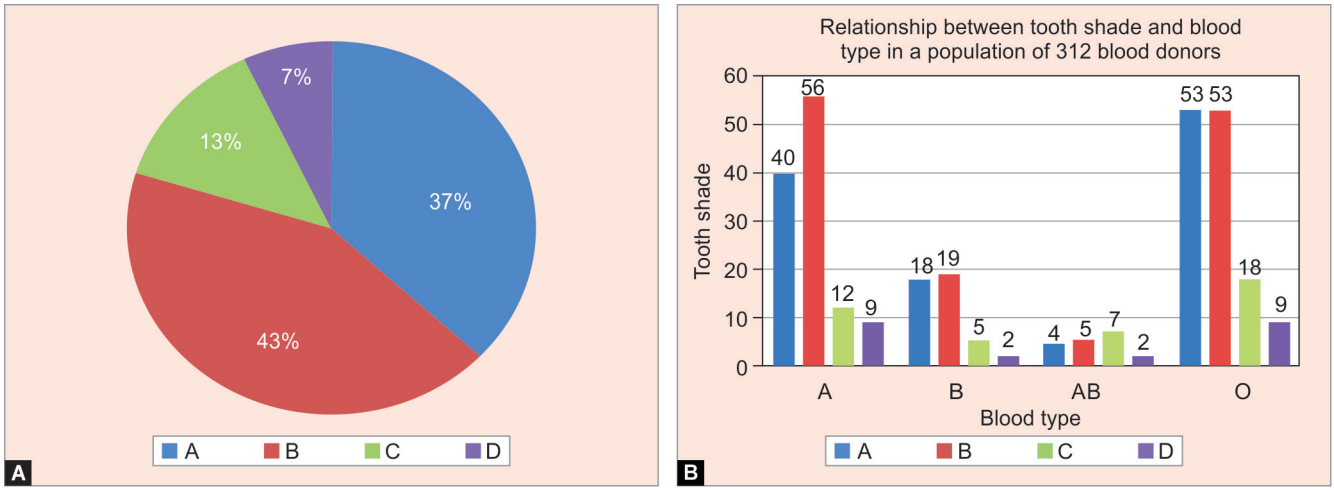
A flow diagram was provided for ease of understanding as shown in Flowchart 1.

Results

In this study, a total of 312 participants were examined, with 267 (85.58%) being male and 45 (14.42%) being female. As shown in Figure 1A, most prevalent tooth shade value was B, with 43% of the population, while the least prevalent tooth shade value was D, with only 7% of the population falling within its spectrum. According to Figure 1B and Table 2, statistical analysis revealed that the overall *p*-value was 0.112139 >0.05, indicating that there is no significant link between tooth shade and blood type. However, the *p*-values for the categories [Blood type B/Tooth shade B], [Blood type O/Tooth shade C], and [Blood type O/Tooth shade D] were <0.001, <0.001, and 0.02, respectively. All three *p*-values were <0.05, indicating that they were statistically significant. According to Table 3A, of the 158 subjects examined, those with tooth shade B were the most prevalent with 70 subjects (44%), while those with tooth shade D were the least prevalent with only 5 subjects (3%). Of the 154 subjects examined, those with tooth

Flowchart 1: Flow diagram





Figs 1A and B: (A) Visual representation of tooth shade distribution in a population of 312 blood donors; (B) Visual representation of tooth shade distribution according to blood type

Table 2: Statistical analysis of the study results

Blood type *Tooth shade*	*A*	*B*	*AB*	*O*	*Row Totals (%)*
A	40 (12.82%) [0.20]	18 (5.76%) [0.20]	4 (1.28%) [1.05]	53 (16.98%) [0.32]	115 (36.85%)
B	56 (17.94%) [0.75]	19 (6%) [<0.001]	5 (1.6%) [0.93]	53 (16.98%) [0.24]	133 (42.62%)
C	12 (3.84%) [0.89]	5 (1.6%) [0.14]	7 (2.24%) [8.65]	18 (5.76%) [<0.001]	42 (13.46%)
D	9 (2.88%) [0.07]	2 (0.64%) [0.39]	2 (0.64%) [0.42]	9 (2.88%) [0.02]	22 (7%)
Column totals (%)	117 (56.73%)	44 (14.1%)	18 (5.76%)	133 (42.62%)	312 (100%)

shade B were the most prevalent with 63 subjects (41%), while those with tooth shade D were the least prevalent with only 17 subjects (11%) as given in Table 3B. According to Figure 2A, of the 45 females examined, subjects with tooth shade A were the most prevalent with 26 subjects (58%), while subjects with tooth shade D were the least prevalent with 0 subjects falling into this category. As per Figure 2B, of the 267 males examined, subjects with tooth shade B were the most prevalent with 116 subjects (44%), whereas subjects with tooth shade D are the least prevalent with 22 subjects (8%).

Discussion

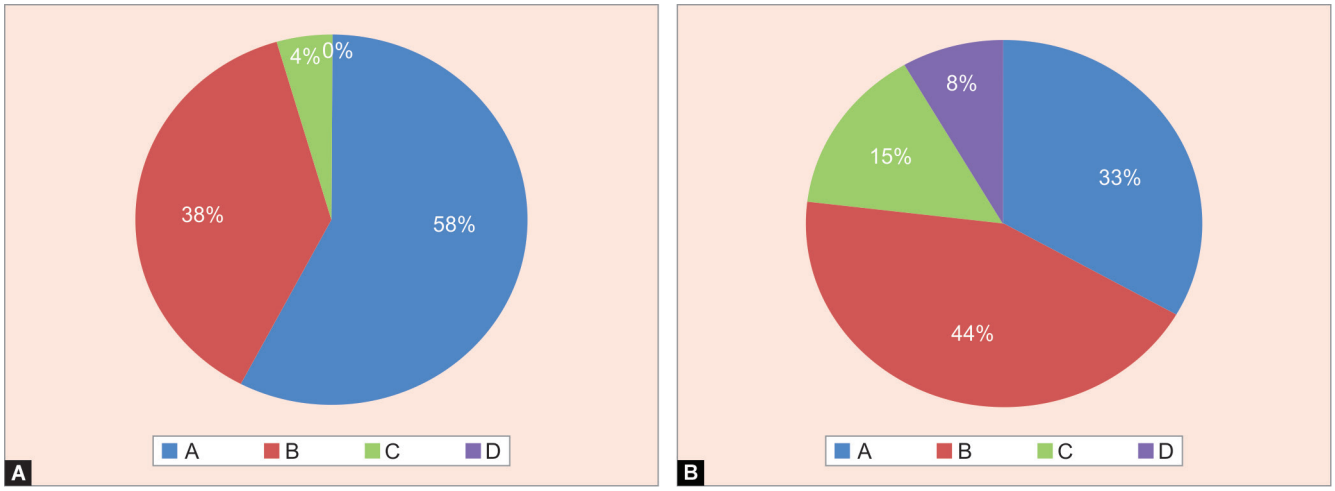
Tooth color plays a significant role in esthetics and is influenced by various factors. The complexity of color encompasses both subjective and objective aspects.[22] Currently, multiple methods are employed to assess tooth color, ranging from visual subjective comparisons using paper, colored porcelain, or acrylic resin shade guides to instrumental objective measurements utilizing spectrophotometers, colorimeters, and image analysis techniques.[23]

Table 3A: Tooth shade distribution within subjects aged under 35

	A	*B*	*C*	*D*	*Total*
<35	67	70	16	5	158
Percentage	43%	44%	10%	3%	100%

Table 3B: Tooth shade distribution within subjects aged over 35

	A	*B*	*C*	*D*	*Total*
>35	48	63	26	17	154
Percentage	31%	41%	17%	11%	100%



Figs 2A and B: (A) Visual representation of tooth shade distribution within the female population; (B) Visual representation of tooth shade distribution within the male population

However, clinicians may encounter difficulties when there is no tooth available as a reference. Overcoming this challenge has been the subject of past studies, with various methods proposed and published. In these situations, alternative patient characteristics can be utilized to aid in tooth shade selection.

While a specific gene responsible for regulating tooth color remains unidentified, in 2009, Golonzhka et al. discovered a gene that controls enamel formation. However, it is possible to genetically determine the presence of dentin protein chromophores, which are aromatic amino acids contributing to tooth color.[24]

Several factors have been suggested as guidelines for selecting tooth shade, including age, sex, and inherited characteristics, such as the color of skin, hair, and eyes. Some studies propose that skin tone provides a more predictable reference for artificial tooth selection, suggesting that individuals with darker skin tones tend to have lighter shades of teeth.[25–27] However, other studies have found no significant relationship between skin tone and tooth shade.[28,29]

In light of these considerations, this study aimed to provide a scientific basis for understanding the relationship between tooth shade and blood types as it is also an inherited characteristic. This knowledge could ultimately enhance the accuracy and reliability of tooth shade selection in clinical practice.

The Ivoclar Vivadent A–D shade guide has been widely chosen for investigating tooth shade due to its regular utilization in dental practices. This shade guide comprises 16 tabs that are organized into four groups according to hue: A (reddish-brown), B (orange-yellow), C (greenish-gray), and D (pinkish-gray). Each subgroup further classifies tabs based on chroma and value. The selection of this shade guide is attributed to its reputation as a reliable, consistent, and user-friendly tool, which has become the standard in dental shade matching.[23]

In the realm of scientific research, a study conducted on the Ivoclar Vivadent A–D shade guide demonstrated its remarkable repeatability, achieving an accuracy rate of nearly 100%. This finding highlights the consistency and precision of the shade guide, further establishing its credibility and reliability for dental shade analysis.[30]

Shade comparison was made at the start of the patient's visit to avoid dehydration of the tooth; as dehydrated teeth appear lighter.[31,32] Shade comparison should be made quickly without staring at a tooth for more than 5 seconds, since our eyes might become accommodated to different colors, thus affecting the tooth color selection procedure. The tooth shade guide was placed under the lip directly, next, and parallel to the tooth being matched, and if possible, it should be in the same plane with the tooth, not behind it or it will appear darker or in front of it or it will appear lighter. Immediately before shade comparison, the eye was focused on a gray-blue surface in order to balance all the color sensors of the retina and resolve the afterimage if it occurred.[33–35]

Specific considerations were implemented for female participants, who were instructed to remove lipstick or lip gloss during the clinical examination and data collection process. Patients were also requested to wear neutral-colored attire and a gray apron was put over their clothes at the time of tooth shade selection to minimize the potential influence of colorful clothing on the examiner's perception of tooth color, thus ensuring the accuracy of the tooth color selection procedure. To maintain the standardization, data collection was carried out by a single researcher.[33]

No previous study has ever explored a possible link between tooth shade and blood type. Even though the overall result of this investigation has concluded the non-existence of a correlation between these two factors, significant results within a few subgroups suggest that further exploration is needed in order to obtain more definite results. It is highly likely that a study with a wider selection of subjects and better tooth shade measurement equipment would lead to a more favorable conclusion.

In our study of tooth shade among Tunisian adults, the predominant shade for females was A (reddish-brown), observed

in 58% of the female population, while the predominant shade for males was B (orange-ish yellow), observed in 44% of the male population. Demirel et al. objectively measured the color of 302 maxillary central incisors using a spectrophotometer and recorded the L, a, and b* parameters. They found a significant difference in a* and b* parameters between genders: Male teeth exhibited higher values of yellowness (b) and redness (a) compared with female teeth, indicating stronger tones of yellow and red in male teeth.[36] In a similar study conducted by Esan et al., it was founded that gender is significantly associated with tooth shades, in that men are more likely to present with darker tooth shades whereas women of the same age-group were more likely to show lighter tooth shades. The reason could be that females have greater esthetic concern and better oral hygiene practices.[28] This discrepancy in the results would be due to the small number of individuals examined, especially to the extremely low number of female individuals (only 45 individuals).

We divided the age range into two groups: 18–35 years and above 35 years because the secondary dentin starts to form at around this age and the enamel layer begins to wear off. Our study found that for both age-groups, the most common tooth shade was B: 44% for those under 35 years and 41% for those over 35 years. Dilesh Pradhan conducted a study that showed that teeth tend to become darker with increasing age (>35 years).[37] Other studies have also reported similar findings.[27,28,38] As Hartmann explained, the darkening of the dentin leads to an altered color in aged teeth.[38] Veeraganta et al. conducted a study comprising 100 subjects aged between 16 and 55, which found that younger subjects have lighter tooth shade values.[1]

Many authors have suggested eye color and skin color as influencing factors in selecting teeth. A few studies only have investigated the relationship between teeth shades and skin tones, and only in one of them, a significant but inverse relationship was found. Persons with medium to dark skin tones were more likely to have teeth with lighter shades than those with lighter skin tones, regardless of their gender or age. Hassel et al., in their study in two urban regions in Germany, found that facial skin or eye color was not associated with L*(Lightness), C*(Chroma), or h*(Hue) color parameters of teeth. However, they also showed that there was an association between hair color (except black) and h* parameter of teeth color. Hassel et al. found in a univariate analysis no association between eye and teeth color, but in a multivariate analysis, higher L* values of teeth were associated with subjects having lighter eye color.[7,10,29,39–41]

Main limitations of the study are: the subject of this study has never been explored prior to this investigation. It follows therefore that there is a severe lack of points of reference, reliable literature, or predetermined schematics that might have saved time and effort, increased the accuracy of results, and better informed this discussion. The selected population was restricted exclusively to blood donors in the eastern coastal region (mainly Monastir) with a few expansions. This restriction automatically excludes anyone who is not eligible to donate blood, such as people in age-groups lower than 18 years and higher than 60 years, people with chronic illnesses, people who weigh less than 60 kg, pregnant people, and people from the rest of the Tunisian governorates. Many researchers, Gigola et al. among them, place classical shade guides on the low end of the spectrum when it comes to accuracy, compared with the 3D-Master system, spectrophotometers, or colorimeters which present a much higher level of accuracy.[42–45] Tooth shade measurement in this study was performed using the Ivoclar Vivadent A–D shade guide, which is a classical shade guide containing 16 tabs arranged according to hue. It is important as well to mention that lighting conditions varied depending on the season and time of day, which might have had an impact on the color matching.

By considering the patient's blood type alongside other relevant factors, clinicians can enhance the accuracy and precision of tooth shade selection, ensuring a more harmonious and natural esthetic outcome. This knowledge aids in overcoming the limitations posed by the absence of historical records, allowing clinicians to tailor their approach based on the patient's specific blood type characteristics.

Further scientific research and investigations into the relationship between tooth shade and blood type will contribute to expanding our understanding of this correlation. Such insights will empower clinicians to employ evidence-based approaches and personalized treatment strategies, ultimately leading to improved clinical outcomes in the field of aesthetic dentistry.

Conclusion

A link between tooth color and blood type, were it to exist, would provide immeasurable advantages. Especially for prosthodontists, it could even allow the reconstruction of the authentic tooth shade of the patient simply based on their blood type. And while the result of this study refutes the existence of this link, the statistical analysis still showcased significant values within different subgroups of the population. A wider selection of subjects, more rigorous tooth shade measurement equipment and conditions, and a more diverse population might well lead to more favorable results.

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