


Morphological analysis of the bristles from toothbrushes sold in the Peruvian market, using scanning electron microscopy

Análise morfológica das cerdas de escovas de dentes vendidas no mercado peruano, utilizando microscopia eletrônica de varredura

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How to cite: Lopez Beraun PA, Alegria Carhuanambo EA, Soto Amante AJ, Paredes Zelada E. Morphological analysis of the bristles from toothbrushes sold in the Peruvian market, using scanning electron microscopy. *Braz Dent Sci.* 2024;27(4):e4503. <https://doi.org/10.4322/bds.2024.e4503>

ABSTRACT

Objective: To analyze, using scanning electron microscopy, the morphological characteristics of the bristle filaments of toothbrushes marketed in Peru. **Material and Methods:** Images obtained through scanning electron microscopy of 23 adult toothbrushes with nylon bristles (621 bristles) were used, and the morphology of their filament tips was evaluated using the Silverstone and Featherstone classification modified by Reiter and Wetzell and classified as Acceptable or Non-acceptable. **Results:** Of the bristles evaluated, 169 (27.2%) were classified as acceptable, while 452 (72.8%) were non-acceptable. Among the toothbrushes analyzed, four exhibited more than 50% acceptable bristles, including one with 100% acceptability. Conversely, another four brushes displayed 100% non-acceptable bristles. **Conclusion:** The results demonstrate a high frequency of non-acceptable bristles, with a ratio 2.5 times greater than the acceptable ones. This variability indicates a lack of uniformity in manufacturing standards across brands, emphasizing the urgency to implement stricter regulations to ensure the quality and safety of these crucial oral hygiene instruments.

KEYWORDS

Dental and oral hygiene products; Oral hygiene; Oral self-care devices; Personal hygiene products; Toothbrushing.

Resumo

Objetivo: Analisar, por meio de microscopia eletrônica de varredura, as características morfológicas dos filamentos das cerdas de escovas de dentes comercializadas no Peru. **Material e Métodos:** Foram utilizadas imagens obtidas por microscopia eletrônica de varredura de 23 escovas de dentes para adultos com cerdas de náilon (621 cerdas), avaliando-se a morfologia das pontas dos filamentos com base na classificação de Silverstone e Featherstone, modificada por Reiter e Wetzell, e categorizadas como Aceitáveis ou Não aceitáveis. **Resultados:** Das cerdas avaliadas, 169 (27,2%) foram classificadas como aceitáveis, enquanto 452 (72,8%) foram consideradas não aceitáveis. Entre as escovas analisadas, quatro apresentaram mais de 50% de cerdas aceitáveis, incluindo uma com 100% de aceitabilidade. Por outro lado, outras quatro escovas apresentaram 100% de cerdas não aceitáveis. **Conclusão:** Os resultados demonstram uma alta frequência de cerdas não aceitáveis, com uma proporção 2,5 vezes maior que a de cerdas aceitáveis. Essa variabilidade indica a falta de uniformidade nos padrões de fabricação entre as marcas, destacando a urgência de implementar regulamentações mais rigorosas para garantir a qualidade e a segurança desses instrumentos essenciais para a higiene bucal.

PALAVRAS-CHAVE

Dispositivos de autocuidado oral; Higiene bucal; Produtos de higiene dental e oral; Produtos de higiene pessoal; Escovação dos dentes.

INTRODUCTION

Dental caries and periodontal diseases are highly prevalent pathologies among humans and are the leading causes of tooth loss. Both conditions, chronic and complex in nature, share several risk factors, with the presence of bacterial plaque being the most notable. This biofilm is a crucial element that poses a significant threat to oral health [1-8].

Effective control of biofilm, both professionally and personally, is essential to ensure and maintain oral health, highlighting the removal of bacterial plaque as a critical strategy in the prevention of oral diseases, using two main methods: mechanical, such as brushing and flossing, and chemical, through mouth rinses. Dental hygiene, particularly through daily and regular toothbrushing, is the most popular and widely adopted method for this purpose. This approach is the most important component of prophylaxis against dental caries and periodontal diseases [3,9-15].

Among the various instruments designed for oral hygiene, the toothbrush stands out due to its remarkable efficacy in removing bacterial plaque, achieving a level of performance that other devices have not matched [16]. From ancient Babylon around 3500 BC, where chewing sticks were used for their antimicrobial properties, to the invention of the first toothbrushes with pig bristles and bamboo or bone handles during the Tang Dynasty (618-907 AD) in China, the toothbrush has undergone significant evolution. These early designs spread to Europe, where pig bristles were replaced with softer bristles, such as horsehair, to suit local preferences. The mass production of the modern toothbrush began with William Addis in England in 1780, followed by industrialization in the United States starting in 1885 [17,18]. The 20th century marked a turning point with the replacement of bone handles by celluloid and later other plastics, and the introduction of nylon bristles and synthetic fibers by DuPont, which improved the hygiene and durability of toothbrushes. The innovative design by Dr. Robert Hutson in the 1950s, featuring multi-tufted nylon filaments with rounded tips and a flat cut, known as the Oral-B manual toothbrush, set a standard for modern toothbrush design [13,19].

The contemporary market offers a diverse range of toothbrushes, each claiming superior plaque reduction capabilities [10], incorporating design improvements that include the number

and distribution of bristles, as well as the size and shape of the head, aimed at overcoming limitations associated with brushing duration and technique [11,19]. These products are equipped with unique features, designed to meet precise standards that facilitate effective dental biofilm management while preserving the integrity of oral tissues [20].

In the development of new toothbrushes, it is crucial to assess not only their efficacy in plaque reduction and improvement of gingival health but also their safety [19,21,22], which is directly related to the functional characteristics of the bristles, often imperceptible to the public, thus necessitating studies that evaluate this aspect [21,23].

Bristles constitute a critical component of toothbrushes, being the only part that comes into direct contact with dental surfaces. For this reason, a deep understanding of the characteristics of these components is essential for selecting the appropriate brush. Factors such as length, diameter, material, and color of the bristles significantly influence the brush's effectiveness in removing plaque and cleaning teeth effectively [17].

The American Dental Association (ADA) recommends that bristle tips be rounded to minimize the risk of dental and gingival abrasions [12,21,24,25]. In 1988, Silverstone and Featherstone evaluated the tips of various toothbrush bristles using a scanning electron microscope (SEM) and classified them into two groups: acceptable (nearly rounded) and non-acceptable (not rounded) [26].

In the toothbrush market, there is a notable lack of information regarding the rounding of bristle tips in many available products. This omission leads to a wide variability in filament morphology, with tips ranging from completely rounded to notably sharp [21]. This variability is also reflected in the Peruvian market, where a wide range of toothbrushes is sold in different establishments at various prices. Consequently, the present study aims to analyze, through SEM, the morphological characteristics of toothbrush bristle filaments marketed in Peru.

MATERIAL AND METHODS

Selection of toothbrushes

This study was conducted at the SEM Laboratory of the Universidad de Huánuco

during the year 2024. Prior to the experiment, a survey of commercially available toothbrushes was conducted in various pharmacies, wholesale stores, and supermarkets in the city of Huánuco. Nylon toothbrushes with cylindrical filaments of varying stiffness (hard, medium, and soft) were selected, excluding those with conical bristles that are chemically treated rather than mechanically processed. The sample was limited to adult toothbrushes to reduce variability. After the initial evaluation, 23 toothbrushes met the established criteria. One unit of each selected model was purchased, totaling 621 bristles for analysis. The details of the models, including toothbrush names, manufacturers, country of origin, and bristle stiffness, were documented in Table I, based on the specifications provided by the manufacturers.

Sample preparation

An examiner prepared the samples from the selected toothbrush brands, assigning

numerical codes to each according to the previous Table. A tuft was randomly selected from each toothbrush to cut the filaments using a No. 15 scalpel. To standardize the counting of filaments to be observed, the 10 samples with the fewest filaments per tuft were selected, and an average was conveniently established across all observations, resulting in 27 filaments ($\mu = 26.8$) selected for each toothbrush brand. The cut filaments were mounted on aluminum slides using carbon adhesive discs and examined under a Thermo Scientific Prisma E SEM, operating under low vacuum conditions at 75 Torr and 30 kV. The microscope was set to a 2 mm magnification and a 90° viewing angle relative to the longitudinal axis of the filaments.

Analysis of toothbrush filament morphology

Based on the images obtained, the percentages of rounded bristles were evaluated in relation to the total number of bristles. All determinations

Table I - Distribution of toothbrushes by name and manufacturer

ID	Name	Manufacturer	Bristle stiffness	No. of tufts/ Filaments per tuft	Origin
ID_001	Family Care	-	-	40 / 26	China
ID_002	Pro® Múltiple acción	PROCTER & GAMBLE mfg. México	Soft	45 / 52	Mexico
ID_003	Pro® Doble acción mayor alcance	PROCTER & GAMBLE mfg. México	Medium	50 / 35	Mexico
ID_004	Pro® Doble acción profile	PROCTER & GAMBLE mfg. México	Hard	50 / 18	Mexico
ID_005	Oral B® Pro-Salud 4 beneficios	PROCTER & GAMBLE mfg. México	Soft	38 / 32	Mexico
ID_006	Colgate® Extra clean	COLGATE SANXIAO Co.	Hard	43 / 16	China
ID_007	Colgate® Extra clean	COLGATE SANXIAO Co.	Medium	43 / 36	China
ID_008	Connert Djimba	-	Soft	35 / 34	-
ID_009	Colgate® Premier Clean	COLGATE PALMOLIVE	Medium	38 / 42	Vietnam
ID_010	New biofresh	-	Soft	42 / 28	China
ID_011	Kolynos® Master Plus	COLGATE SANXIAO Co.	Medium	39 / 32	China
ID_012	Dento®	INTRADEVCO INDUSTRIAL	Medium	40 / 24	Peru
ID_013	Oral B® 1,2,3	RIALTO ENTERPRISES PVT. Ltda	Medium	38 / 50	China
ID_014	Vitis® Medio	DENTAID S.L.	Medium	29 / 35	Spain
ID_015	Vitis® Suave	DENTAID S.L.	Soft	29 / 42	Spain
ID_016	Vitis® Duro	DENTAID S.L.	Hard	40 / 30	Spain
ID_017	Colgate® 360° Advanced Luminous	COLGATE SANXIAO Co.	Soft	26 / 52	China
ID_018	Balanzé BÁSICOS PROACTIVE	HUBEI CROWN HOUSEWARES Co. Ltd	Medium	37 / 30	China
ID_019	Oral B® Pro-Salud 7 beneficios	RIALTO ENTERPRISES PVT. Ltda	Soft	35 / 72	India
ID_020	Oral B® Complete 4 beneficios	PROCTER & GAMBLE mfg. México	Medium	38 / 42	Mexico
ID_021	Daily Caristop®	MAVER PERÚ S.A.C.	Soft	41 / 50	Peru
ID_022	Colgate® Recy Clean	COLGATE SANXIAO Co.	Soft	39 / 45	China
ID_023	Total Dent	-	n.s.	41 / 32	China

n.s. = ...

of the rounding patterns of toothbrush bristles were performed by two blind, duly calibrated investigators. To mitigate the risk of bias related to the prior knowledge of the toothbrush brands by the investigators responsible for sample preparation, rigorous measures were adopted to ensure the impartiality of the analysis. The samples were exclusively evaluated by two independent researchers who did not participate in the preparation phase or in the capture of images using SEM. This strategy ensured complete blinding of the evaluators, avoiding any influence derived from familiarity with the toothbrush brands.

For the evaluation of the bristle tip morphology, the Silverstone and Featherstone classification [26], modified by Reiter and Wetzel [27], was used, as shown in Figure 1, taken from the study by Lee et al. [23]. In this classification, bristles with adequately rounded ends were considered acceptable and were subdivided into three categories (A1–A3). On the other hand, bristles with undesirable characteristics, such as sharp edges, sharp rectangular tips, sharp oblique tips, triangular tips, and tips with protruding plastic material, were classified as non-acceptable. These were further grouped into five subcategories (N1–N5) [23].

Due to the microscope's magnification settings, it was necessary to capture multiple views to observe the 27 filaments for each brush ID.

Analysis of the obtained data

After data collection, the cumulative frequency distribution by characteristics and

pattern type was analyzed according to the Silverstone and Featherstone classification [26], modified by Reiter and Wetzel [27]. Subsequently, the ratio between the total bristles per pattern and the total number of bristles in the study was calculated to determine the relative frequency of the characteristics and pattern types of the bristles for each toothbrush ID.

RESULTS

The characteristics of the bristles of the toothbrushes evaluated in this study are shown in Table II. According to the total number of bristles analyzed by SEM ($n=621$), it was observed that the number of acceptable bristles was 169 (27.2%). Of the total toothbrushes analyzed, only four codes (ID 001, 002, 006, and 023) had a percentage of over 50% acceptable bristles based on the number of filaments per sample. Additionally, four codes (ID 010, 011, 012, and 022) were observed with a 100% percentage of non-acceptable bristles. The remaining observations ranged from 7.4% to 48.1% for acceptable bristles. ID 002 had an observation equal to 100% acceptable bristles.

On the other hand, Table III shows the distribution of bristle characteristics by pattern type according to the classification of Silverstone and Featherstone [26], modified by Reiter and Wetzel [27]. In the case of patterns with Acceptable tips, ordered from highest to lowest, category A3 presented a percentage of 12.6%, followed by A1 with 8.4% and A2 with 6.3%.

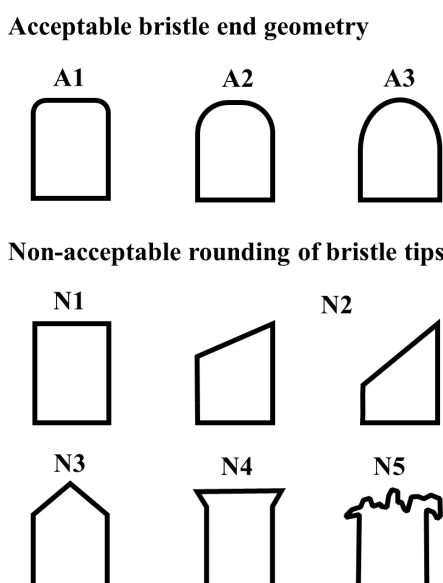


Figure 1 - Classification of toothbrush bristle tip morphology according to Silverstone and Featherstone [26], modified by Reiter and Wetzel [27].

Table II - Distribution of bristle characteristics according to SEM observation

ID	Acceptable	%	Non-Acceptable	%	Total bristles per tuft	%
ID_001	19	70.4%	8	29.6%	27	4.3%
ID_002	27	100.0%	0	0.0%	27	4.3%
ID_003	11	40.7%	16	59.3%	27	4.3%
ID_004	4	14.8%	23	85.2%	27	4.3%
ID_005	5	18.5%	22	81.5%	27	4.3%
ID_006	18	66.7%	9	33.3%	27	4.3%
ID_007	6	22.2%	21	77.8%	27	4.3%
ID_008	3	11.1%	24	88.9%	27	4.3%
ID_009	5	18.5%	22	81.5%	27	4.3%
ID_010	0	0.0%	27	100.0%	27	4.3%
ID_011	0	0.0%	27	100.0%	27	4.3%
ID_012	0	0.0%	27	100.0%	27	4.3%
ID_013	3	11.1%	24	88.9%	27	4.3%
ID_014	8	29.6%	19	70.4%	27	4.3%
ID_015	4	14.8%	23	85.2%	27	4.3%
ID_016	4	14.8%	23	85.2%	27	4.3%
ID_017	10	37.0%	17	63.0%	27	4.3%
ID_018	13	48.1%	14	51.9%	27	4.3%
ID_019	7	25.9%	20	74.1%	27	4.3%
ID_020	2	7.4%	25	92.6%	27	4.3%
ID_021	5	18.5%	22	81.5%	27	4.3%
ID_022	0	0.0%	27	100.0%	27	4.3%
ID_023	15	55.6%	12	44.4%	27	4.3%
Total	169	27.2%	452	72.8%	621	100%

Table III - Distribution of bristles by pattern type

ID	Acceptable			Non-Acceptable					Number of bristles analyzed	%
	A1	A2	A3	N1	N2	N3	N4	N5		
D_001	7	-	12	-	4	4	-	-	27	4.30%
ID_002	3	6	18	-	-	-	-	-	27	4.30%
ID_003	-	4	7	-	9	7	-	-	27	4.30%
ID_004	1	3	-	-	23	-	-	-	27	4.30%
ID_005	5	-	-	3	19	-	-	-	27	4.30%
ID_006	-	-	18	-	8	1	-	-	27	4.30%
D_007	-	-	6	-	4	17	-	-	27	4.30%
ID_008	-	3	-	5	-	-	9	10	27	4.30%
ID_009	-	-	5	-	5	17	-	-	27	4.30%
ID_010	-	-	-	2	7	-	18	-	27	4.30%
ID_011	-	-	-	-	17	10	-	-	27	4.30%
ID_012	-	-	-	2	9	16	-	-	27	4.30%
ID_013	1	2	-	1	7	5	5	6	27	4.30%
ID_014	8	-	-	4	12	3	-	-	27	4.30%
ID_015	4	-	-	9	10	3	-	1	27	4.30%
ID_016	1	3	-	-	11	12	-	-	27	4.30%
ID_017	3	7	-	9	2	-	6	-	27	4.30%
ID_018	7	2	4	9	1	4	-	-	27	4.30%
ID_019	2	2	3	3	11	5	1	-	27	4.30%
ID_020	-	-	2	9	11	5	-	-	27	4.30%
ID_021	5	-	-	7	9	4	2	-	27	4.30%
ID_022	-	-	-	6	-	21	-	-	27	4.30%
ID_024	5	7	3	1	3	8	-	-	27	4.30%
Subtotal	52	39	78	70	182	142	41	17	621	100%
fr (%)	8.40%	6.30%	12.60%	11.30%	29.30%	22.90%	6.60%	2.70%	1215	100%

Likewise, in the Non-Acceptable pattern group, the category that presented the highest frequency was N2 with 29.3%, followed by N3, N1, N4, and N5 with frequencies of 22.9%, 11.3%, 6.6%, and 2.7%, respectively.

Figure 2 presents SEM images showing the tips of toothbrush bristles. Images labeled I and II represent bristles classified within acceptable quality standards. In contrast, images labeled III and IV show bristles that do not meet the established quality criteria and are considered non-acceptable.

DISCUSSION

The purpose of this study was to analyze, through SEM, the morphological characteristics of the bristle filaments from toothbrushes sold in the Peruvian market.

Good oral hygiene practiced regularly can help maintain functional dentition throughout life. However, it is also known that good oral hygiene standards have often been associated with soft tissue complications, such as

gingival abrasions [1,10,19,28,29], which are characterized as localized, reversible epithelial lesions that can range from superficial wounds to erosions exposing the underlying connective tissue. This complication can be caused by various factors, including excessive use of the toothbrush, brush design, bristle stiffness and quality, as well as brushing frequency and the characteristics of the bristle material [21]. Specifically, the use of brushes with non-rounded filament tips has been linked to a higher incidence of these lesions [12,21,24,25]. Hennequin-Hoenderdos et al. [21] have demonstrated that a rounding of bristle tips between 40% and 50% can significantly reduce the risk of gingival abrasions. It has been emphasized that repeated gingival abrasions constitute a significant factor in the development of gingival recession [1,30]. However, there is no clear evidence that gingival abrasion caused by toothbrushing leads to recession [1], as the etiology of gingival recession is multifactorial, involving anatomical and iatrogenic factors, in addition to pathological conditions associated with dental plaque, such as periodontitis [28].

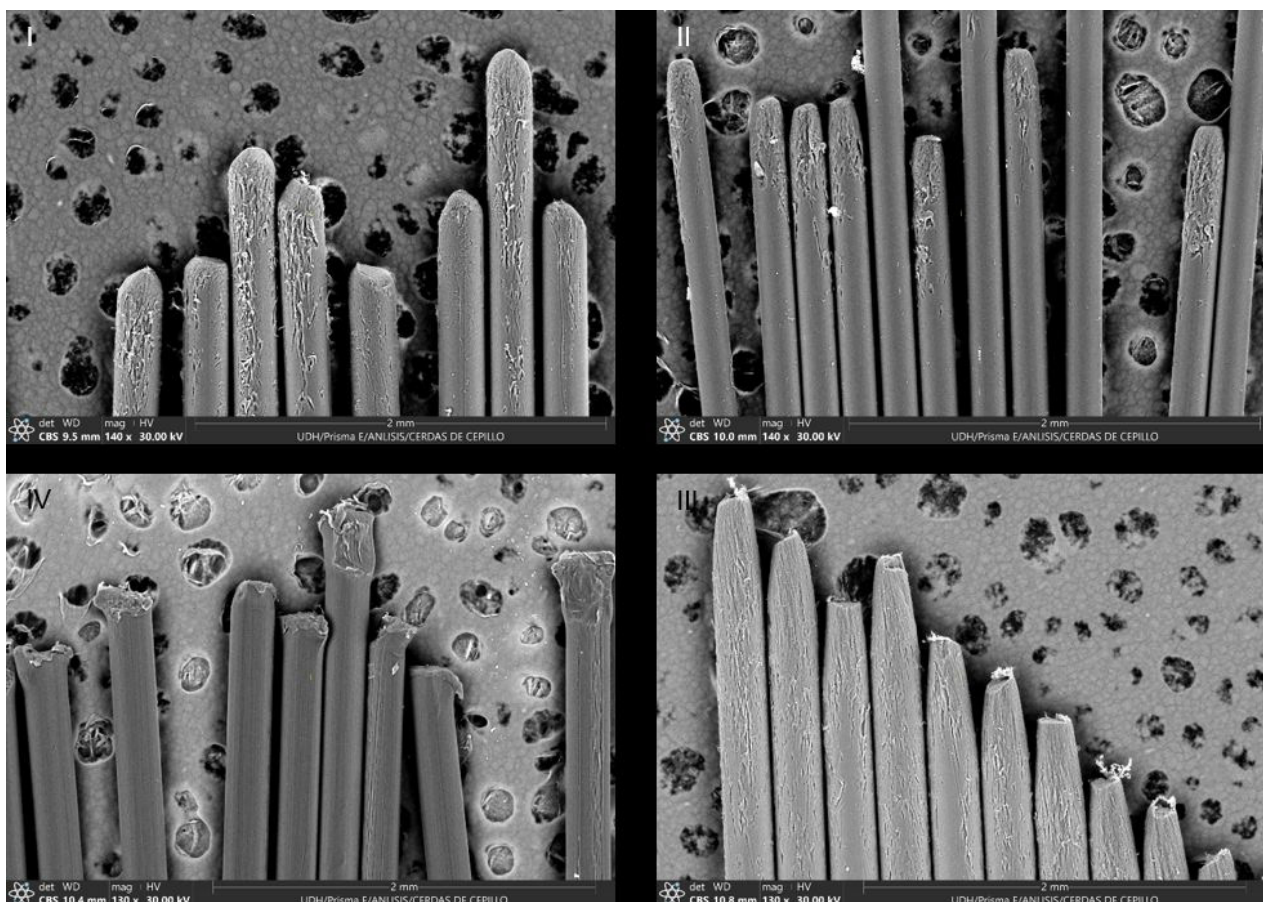


Figure 2 - SEM images showing the tips of toothbrush bristles. Images labeled I and II were classified as acceptable, while labels III and IV correspond to bristles considered non-acceptable.

The ADA also mentions dental abrasions as a pathology that can be prevented by using toothbrushes with rounded bristles [12,21,24,25]. These non-carious lesions are common among patients who suffer from dental sensitivity and discomfort resulting from dentin exposure [31], and are associated with gingival recession [1]. However, it has been established that nylon toothbrushes alone have insignificant effects on hard dental tissues and could only indirectly influence the abrasion process by modulating the action of toothpaste abrasives carried over the dental surface, which varies depending on the design of each brush [32].

According to the guidelines of the “European Workshop on Mechanical Plaque Control,” the optimal toothbrush should be tailored to the user’s individual needs. It is essential that the brush head is proportionate to the oral cavity of the person using it, considering their age and motor skills. Moreover, the brush filaments should be soft and comply with ISO standards, with materials such as nylon or polyester being preferred. These filaments should have rounded ends to prevent injuries to the gums and soft tissues, and their diameter should not exceed 0.25 mm to facilitate effective and safe brushing [12,24]. It is crucial that the proportion of bristles with an acceptable morphology exceeds those considered non-acceptable according to Silverstone and Featherstone’s definition [12].

A wide range of toothbrushes shows variability in the degree of bristle tip rounding, and a significant percentage of these do not meet acceptable quality standards [21]. The results of the present study revealed a notable variability in the quality of the analyzed toothbrushes, with some samples presenting 100% non-acceptable bristles, while others exhibited entirely acceptable bristles. This heterogeneity is consistent with findings reported in previous literature, reflecting the variability among different brands and models. Rehman et al. [10] found that between 57.6% and 91.7% of the bristles met the acceptability criteria, whereas studies like that of Yildiz Telatar et al. [12] reported lower compliance percentages, ranging from 8.9% to 41.3%. These differences may be influenced by variability in toothbrush design and manufacturing processes used by different producers. Additionally, Lee et al. [23] observed even lower percentages of acceptable bristles (1.4% to 20.2%) in children’s toothbrushes,

which is particularly concerning given that children are more vulnerable to harm caused by poorly designed bristles. On the other hand, Jung et al. [29] identified greater variability in children’s electric toothbrushes, with an acceptability percentage ranging from 18.9% to 94.3%; for adult toothbrushes, they found that between 21.7% and over 80% of the bristles were acceptable. This suggests that electric toothbrushes also suffer from inconsistencies in quality standards. These findings align with those of Meyer-Lueckel et al. [33] and Checchi et al. [24], who reported a wide variation in bristle quality (34% to 98% and 0% to 70%, respectively). The results of this research contribute to the growing body of evidence highlighting inconsistencies in toothbrush manufacturing standards. This underscores the need for more rigorous regulations at both local and global levels to ensure the quality and safety of these essential products.

Studies that evaluate the morphology of bristle tips are generally conducted using optical microscopy, stereomicroscopy, or SEM [12,29,33]. The latter method is preferred due to its high resolution, which facilitates detailed visualization of bristle tips. The SEM’s ability to capture images with superior depth of focus allows the researcher to discern variations in the surface texture of multiple filaments within a single tuft simultaneously [33]. Consequently, this study used images from SEM to achieve precise representations of the filaments. However, it is important to consider that the three-dimensional representation of a filament tip is reduced to a two-dimensional image in the capture process, limiting the analysis to only the visible parts of the filament tip, omitting any hidden aspects. As a result, some judgments or measurements may be erroneously positive, following the evaluation instrument proposed by Silverstone and Featherstone [33].

Another limitation identified in research related to SEM is the potential morphological alteration of toothbrush bristle tips. These changes are attributed to temperature increases during preparatory procedures for analysis, which can significantly compromise the integrity of the samples and, consequently, affect the reliability of the results obtained [23,29]. Franchi and Checchi [34], in a study, revealed that the preparatory conditions for analyzing toothbrush bristles using SEM affect their original

morphology. Factors such as the distance to the cathode in a sputter coater and the coating time with gold-palladium influence the temperature within the vacuum chamber, which in turn modifies the morphology of the bristles. They found that higher temperatures, around 59 °C, near the cathode can alter the shapes of the bristles, making them appear acceptable when they are not, while lower temperatures, around 38 °C, tend to preserve their original shape. This is why some of the studies mentioned in the previous paragraph opted to use the stereomicroscope [12,24]. In the present study, a low-vacuum SEM method was used, where the aforementioned preliminary coating was not necessary, maintaining the integrity of the filaments throughout the process.

A significant limitation of the present study was the use of a single tuft per toothbrush, selected at random. This methodology did not allow for determining whether variations exist between individual tufts or among different toothbrushes of the same model. Therefore, it is recommended that future research include the evaluation of multiple toothbrushes per brand to obtain more generalizable and accurate results.

CONCLUSIONS

The Comparative observation between acceptable and non-acceptable bristles revealed that the prevalence of non-acceptable bristles was significantly higher, surpassing the acceptable ones by a ratio of 2.5 to 1.

Author's Contributions

PALB: Conceptualization, Investigation, Writing – Original Draft Preparation, Writing – Review & Editing, Project Administration, Funding Acquisition. EAAC: Investigation, Writing – Original Draft Preparation, Writing – Review & Editing. AJSA: Writing – Original Draft Preparation, Writing – Review & Editing. EPZ: Conceptualization, Investigation, Writing – Original Draft Preparation, Writing – Review & Editing.

Conflict of Interest

The authors have no personal, financial, or proprietary interests in any product, service, and/or company presented in this article.

Funding

This study was fully funded by the Universidad de Huánuco, under Resolution No. 857-2023-P-CD-UDH.

Regulatory Statement

This study was exempted from review by the local ethics committee, since it did not involve the participation of any volunteers, or the use of any human material.

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Date submitted: 2024 Aug 30
Accept submission: 2024 Nov 21