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Posted Date: 13 June 2024

doi: 10.20944/preprints202406.0932.v1

Keywords: Cheiloscopy; Forensic dentistry; Lip prints; Sex.

 doi:10.20944/preprints202406.0932.v1

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Cheiloscopic Evaluation in Forensic Odontostomatology: Exploring Potential and Limitations

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Abstract: Background: The aim of this experimental work was to evaluate cheiloscopy potential: to confirm the individual uniqueness of lip prints, to verify the presence of recurrent patterns according to the Suzuki and Tsuchihashi classification and to search for characteristics that can be used in establishing the sex of a subject, with a focus on assessing the method's limitations. Methods: Lip prints were taken from a sample of 172 volunteers using liquid lipstick and transparent adhesive tape and were digitally analyzed. Results: 59 prints (35.3%) were not considered for Suzuki and Tsuchihashi classification due to quality errors and the impossibility of visualizing lip wrinkles in one or more sextants. All the digitized prints revealed themselves to be unique after having been analyzed using superimposition. Qualitatively valid prints indicated a prevalence of Type II in both male and female. A major problem was that the analysis of the same print by different operators often resulted in a different attribution of a Type. Conclusions: The potential of cheiloscopy lies mainly in the matching of prints, given their uniqueness, but the absence of a database greatly limits its use. Additionally, its use for sex identification does not seem to be reliable. It is necessary to establish an acquisition protocol for taking and analyzing prints that could reduce the errors committed by the operator.

Keywords: cheiloscopy; forensic dentistry; lip prints; sex

1. Introduction

Forensic science is an academic field that investigates legal (and illegal) matters using science, and subsequently applies research results to resolving real crime cases [1].

Conventional approaches to the identification of individuals range from the use of anthropometry to fingerprint analysis, from age and sex determination to blood grouping and DNA analysis, and finally odontology [2], a multidisciplinary field of study combining dental knowledge, forensic medicine and anthropology. It permits the identification of individuals by exploiting the characteristics of the oral cavity such as dental prints and records, palatal rugae and lip prints [5–7,35], which are unique to each person. This branch can be crucial in the victim identification processes or in crime contexts [3].

Forensic odontology plays an essential role when other methods of recognition, such as fingerprints or facial recognition, are not feasible due to the body being compromised, such as in the

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case of charred or decomposed corpses. This is possible because many structures in the oral cavity, such as teeth and bones, are highly resistant to external impacts like putrefaction, fire, explosions, chemicals, etc. [4]. A range of techniques, such as the analysis of bite marks and ante-mortem dental records, rugoscopy and cheiloscopy, can be invaluable aids in forensic odontology [5–7].

Dental prints found at crime scenes have long been regarded as useful or crucial evidence for obtaining convictions. However, over the years, several critical issues have arisen concerning conclusions regarding the identification of bite injuries, which have led to defendants being unfairly convicted [5].

In forensic odontology, palatal rugae represent an important identification marker, especially in edentulous individuals. It has been observed that the hard palate and its wrinkles constitutes an exceptionally stable structure over time, even in cases where there are charred or decaying skulls, and recent studies have shown that there is promising data regarding the identification of the sex of a subject, but this method is also limited by the absence of a database [6].

Forensic odontologists also play a crucial role in collaborating with other experts to ascertain the sex of remains by examining dental and cranial characteristics. Teeth offer a wealth of information, with features like morphology, crown dimensions, and root length serving as key indicators for distinguishing between male and female sexes. Similarly, the skull presents distinct patterns and traits unique to each sex, further aiding in the accurate determination of sex in forensic investigations [7].

Cheiloscopy (from the Greek: 'keilos'= lip and 'skopein'= to see) is a forensic investigation technique that deals with identification of humans based on lips traces, by analyzing the grooves present at the vermilion level [35].

The anthropologist R. Fischer was the first to describe the presence of lip wrinkles in 1902, but without suggesting any practical use [8]. In the early 1930s, Dr. Edmond Locard, a prominent French criminologist, began investigating the potential of lip prints as a means of identification. This represented a turning point in the understanding of the potential of this discipline. Locard recognized the uniqueness of lip prints and their importance in the field of investigation [9]. Recent studies have furthermore proved the uniqueness of lip prints [36–40].

Between 1968 and 1971, two Japanese scholars, Y. Tsuchihashi and T. Suzuki, determined that the arrangements of all the grooves on the red portion of the lips are unique in each individual [10]. Tsuchihashi observed that even homozygotic twins have different lip prints, even though they possess an identical set of chromosomes [10].

Tsuchihashi and Suzuki gave the name 'Sulci Labiorum' to the individual grooves and 'Figura linearum labiorum rubrorum' to the lip prints consisting of these grooves [10].

Lip prints have distinct characteristics, can be found as early as the sixth week of intrauterine life and do not undergo major changes during the course of one's life [11].

It has been observed that the characteristics of labial wrinkles essentially remain stable even in response to minor trauma, such as inflammation or pathological conditions such as herpes, and are not significantly influenced by environmental factors. However, changes in the shape and size of labial wrinkles can be observed following major trauma, such as scarring from surgery [10].

Despite these changes, it is rare for the entire lip structure to be compromised, thus enabling the recognition of lip prints by matching them with lip areas not affected by the alterations due to the traumatic event.

On the other hand, some scarring may be an advantage in the identification process when it pre-exists the taking of lip prints. It adds a further level of uniqueness and specificity to the identification characteristics of the lips [12].

Another observed fact is that there do not seem to be any substantial alterations of lip wrinkles in the presence or absence of parafunctional oral habits such as smoking, vaping, playing a wind instrument or using an asthma inhaler [13].

The aim of this experimental work is to assess the uniqueness of lip wrinkles, identify recurring patterns according to the Suzuki and Tsuchihashi classification and explore potential features identification, building upon previous research in the field [41–43].

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2. Materials and Methods

This experimental study examined a total of 172 lip prints acquired at the University of Palermo and was conducted in accordance with the Declaration of Helsinki and approved by the Institutional Review Board or Ethics Committee University of Palermo Policlinico "Paolo Giaccone", Minutes No. 04/2024, Session of February 8, 2024.

Inclusion criteria:

- Caucasian subjects.
- Lips free of active lesions.
- Absence of previous allergic reactions to lipstick.
- Stable general health condition.
- Indication of the sex of the subject.

Exclusion Criteria:

- Refusal of informed consent.
- Presence of active injuries or recent surgery of the lips (herpes, cheilitis).
- Presence of congenital malformations (cleft lip).

Following the acquisition of informed consent from the volunteers, a meticulous clinical examination of each subject's lips was conducted to identify any recent lesions, active pathologies, or congenital malformations, all of which served as exclusion criteria for the study. Subsequently, the lips were thoroughly cleaned to ensure optimal conditions for the following steps. every impurity, such as cosmetic traces and exfoliated skin cells, were removed using a makeup remover and cotton pads. A thin layer of liquid lipstick was delicately applied to the lips using disposable brushes, and participants were instructed to rub their lips together to achieve an even distribution of the lipstick.

After allowing a brief interval of 30 seconds for the lipstick to partially dry, the upper and lower lip prints were captured simultaneously on strips of adhesive tape measuring 5x10cm. To minimize any potential alterations, the samples were left to dry completely before being transferred onto white cards for ease of storage and handling. In the course of our study on cheiloscopy, we found it necessary to exclude certain lip print tests as invalid tests. This step was crucial to maintaining data integrity and ensuring that our conclusions were based on accurate and representative information. The primary reasons for test invalidity included sample contamination, procedural errors, and issues with data quality. Regarding sample contamination, some samples were exposed to external factors that compromised the accuracy of the prints, such as the intrusion of foreign particles or the presence of non-standard lipstick traces. Procedural errors, such as improper application of lipstick or incorrect use of tape for print collection, resulted in distorted and non-replicable results. Additionally, we encountered issues with data, particularly incomplete or unreliable data. Some prints lacked the necessary information for an accurate analysis, making it impossible to use them for reliable comparative evaluation. These factors made it essential to exclude certain tests, thereby preserving the quality of our study and ensuring that our conclusions were based on authentic and accurate lip prints.

Pertinent information including age, sex and race of each subject, the date of lip print acquisition, and a unique identification number were recorded on the back of each print. This was done to ensure objectivity and mitigate any potential bias during the identification of sex-related patterns.

The identification number was then linked to the respective patient's data in a separate document to uphold privacy protocols.

To facilitate efficient analysis, all lip prints were digitized, ensuring a practical and precise approach to data interpretation and comparison.

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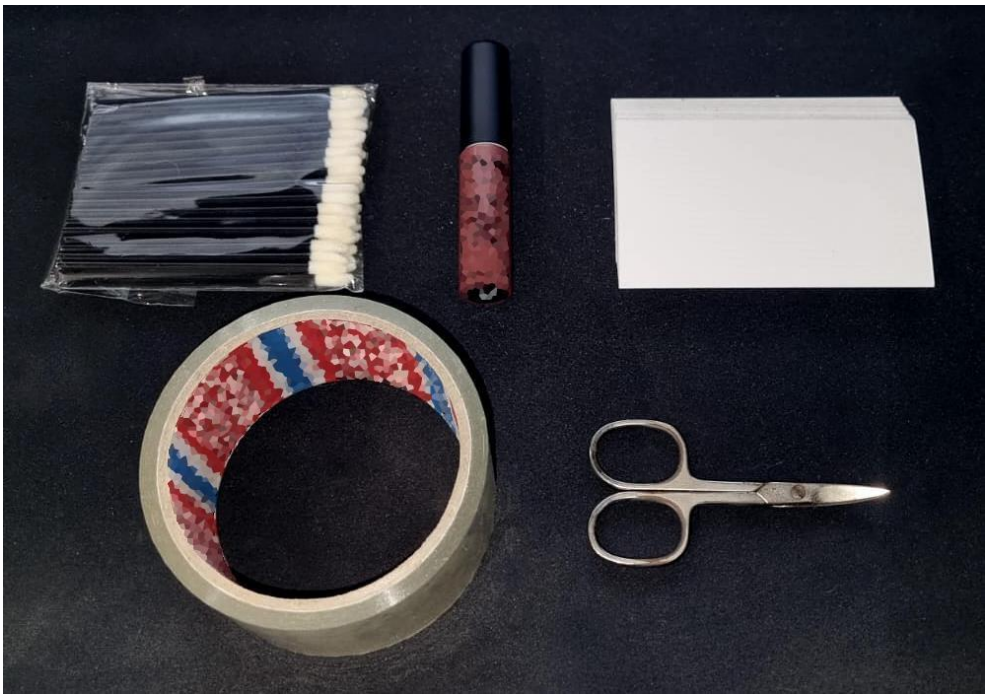

Figure 1. Tools used for lip prints capture.

To better isolate and visualize the lip wrinkles, all digitized prints were edited using Adobe® Photoshop image processing software [Figure 2]:

- Application of greyscale.
- Binarization of the image.
- Outlining the lip contour.
- Manual tracing of labial wrinkles for each print.

Figure 2. Editing steps of digitized prints.

Each print was first analyzed individually and then compared using superimposition through 2D analysis in transparency, also utilizing the Adobe® Photoshop image processing software. This

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process involved examining each print and comparing it one by one with all others to assess its uniqueness.

In this study, lip prints were analyzed using a sextant-based approach, a method that involves dividing the lips into six distinct areas for detailed examination. In literature there're significant variation in the chosen lip area for analysis. Approaches ranged from analyzing the entire lip without any segmental division to dividing the lip into four, six, eight, or twelve segments, most commonly arranged in a clockwise manner. Some methods also focused on more restricted areas, limiting the analysis to specific smaller sections of the lip (47).

Each lip print was divided into sextants , and these segments were evaluated according to the Suzuki and Tsuchihashi classification system that divides lip prints into six different Types [10] [Figure 3]:

- Type I: full vertical.
- Type I': partial vertical.
- Type II: branching.
- Type III: intersection.
- Type IV: reticular.
- Type V: irregular.

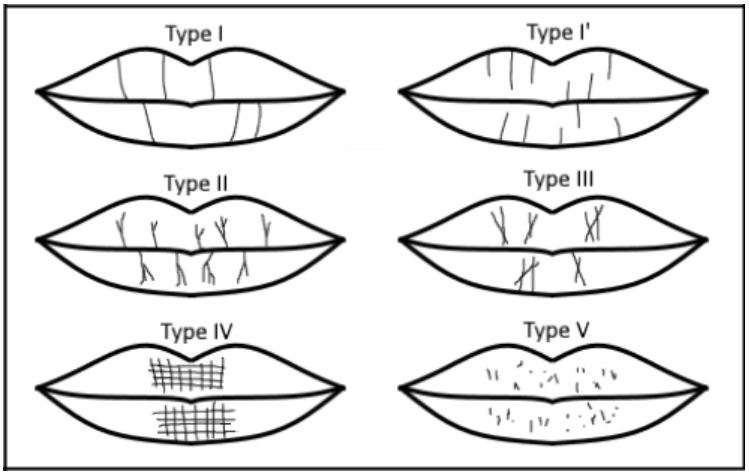

Figure 3. Suzuki and Tsuchihashi classification of lip grooves, adapted from [35].

A predominant Type was attributed to each lip print based on the prevailing pattern observed across the sextants and, in a parallel analysis, the presence of sex-specific features within the lip prints was examined. In this study five operators were included, and each operator independently analyzed all 108 prints and to allow the identification of a specific Type, whenever we encountered a doubtful situation, the print was reanalyzed simultaneously by the 5 operators to establish a common agreement.

The collected data and analyses were then transferred to Microsoft Excel and a Pearson chi-square test was performed regarding the correlation between sex and the quality of lip prints acquisition, the predominant Types, the presence of sex-linked characteristics and the presence of macroscopical cosmetic traces.

3. Results

Of the 172 prints, 1 female and 4 males were excluded as they did not meet the study's inclusion criteria, reducing the impressions to 167, 85 female and 82 males. A further 59 prints (39 male and 20 female) were not included in the Suzuki and Tsuchihashi classification as they presented unreadable sextants due to the poor quality of the sample. These, however, were analyzed to assess their uniqueness and to see which sextants were most affected by technique errors. The final sample consisted of 108 prints, comprising 43 male and 65 female prints, which were eligible for Suzuki and Tsuchihashi classification [10].

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Table 1. Statistical analysis of the quality of affected areas.

	Female prints	Male prints	Age <18	Age ≥18;<30	Age ≥30;<50	Age ≥50	Total
Prints taken	85	82	15	99	43	10	167
Prints exluded	20	39	10	26	16	7	59
Remaining samples	65	43	5	73	27	3	108

This percentage of qualitatively invalid prints (35.3%) underlines how important it is to establish a standardized acquisition protocol that can enable the acquisition of the sharpest and clearest possible prints. A statistical analysis revealed several interesting facts regarding the quality of the prints [Table 1]:

- Male prints proved more difficult to obtain; more than twice as many male prints (47.56%) as female prints (23.53%) were discarded.
- The individual male prints had, on average, more sextants involved than female ones, 3.9 and 2.95 respectively.
- Male prints were more illegible in the upper lip, unlike female prints, where the opposite tendency was noted.
- Of the male prints, 8 were not readable in their entirety, while only 1 of the female prints had the same level of insufficient quality.

The chi-square test showed a correlation between sex and the number of valid and discarded prints (p-value=0.001) and qualitatively affected areas (p-value=0.004)

Table 1. Statistical analysis of the quality of affected areas.

Observed			
N prints	***Female***	***Male***	**Total**
Discarded	20	39	**59**
Valid	65	43	**108**
Total	**85**	**82**	**167**

Expected			
N prints	***Female***	***Male***	**Total**
Discarded	30,03	28,97	**59**
Valid	54,97	53,03	**108**
Total	**85**	**82**	**167**

Test			
N prints	***Female***	***Male***	**Total**
Discarded	3,35	3,47	
Valid	1,83	1,90	
Total			**10,55**

Results	
Degree of freedom	1
Test stats	10,550
p-value	0,001

Invalid Sextant	***Female***	***Male***	**Total**
Sextant I	9	36	**45**
Sextant II	4	28	**32**
Sextant III	9	31	**40**
Sextant IV	13	29	**42**
Sextant V	14	32	**46**
Sextant VI	10	28	**38**
Total	**59**	**184**	**243**

Invalid Sextant	***Female***	***Male***	**Total**
Sextant I	10,93	34,07	**45**
Sextant II	7,77	24,23	**32**
Sextant III	9,71	30,29	**40**
Sextant IV	10,20	31,80	**42**
Sextant V	11,17	34,83	**46**
Sextant VI	9,23	28,77	**38**
Total	**59**	**184**	**243**

Invalid Sextant	***Female***	***Male***	**Total**
Sextant I	0,34	0,11	
Sextant II	1,83	0,59	
Sextant III	0,05	0,02	
Sextant IV	0,77	0,25	
Sextant V	0,72	0,23	
Sextant VI	0,06	0,02	
Total			**4,98**

Degree of freedom	5
Test stats	4,983
p-value	0,418

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Unclassifiable Areas	*Female*	*Male*	Total
Only upper lip	2	12	**14**
Only lower lip	9	3	**12**
Entire print	1	8	**9**
Mixed	8	16	**24**
Total	**20**	**39**	**59**

Unclassifiable Areas	*Female*	*Male*	Total
Only upper lip	4,75	9,25	**14**
Only lower lip	4,07	7,93	**12**
Entire print	3,05	5,95	**9**
Mixed	8,14	15,86	**24**
Total	**20**	**39**	**59**

Unclassifiable Areas	*Female*	*Male*	Total
Only upper lip	1,59	0,81	
Only lower lip	5,98	3,07	
Entire print	1,38	0,71	
Mixed	0,00	0,00	
Total			**13,54**

Degree of freedom	3
Test stats	13,539
p-value	0,004

The analysis of all the prints using superimposition confirmed the uniqueness of the prints; a match between the prints of different individuals was not possible.

In this study five operators were included, and each operator independently analyzed all 108 prints. A relevant factor was that the analysis of the same print by different operators often resulted in a different attribution of a Type. Paying attention to these errors, the greatest inconsistencies were noted between:

- Type I and Type I': Some operators attributed Type I to the presence of at least one complete vertical labial wrinkle, while others did this only if most, if not all, of the vertical wrinkles were complete. For our study, we decided to apply a Type I if 50% or more complete vertical wrinkles were presents.
- Type I/I' and II: In 5 cases, it could not be determined whether one lip wrinkle was the offshoot of another or whether there were two separate wrinkles
- Type II and III: This turned out to be the most frequent error found in 15 prints, with some operators considering branching wrinkles as intersecting and vice versa, mistaking Type II and Type III with each other.
- Type V: In 7 cases, there was the propensity to attribute a Type V when the prints were of an apparently unsuitable quality for cheiloscopy analysis.

We proceeded to initially attribute a single Type per sextant to the 108 qualitatively valid prints, subsequently identifying a single Type per print based on the most frequent Type in the six sextants.

Looking specifically at the data obtained in our study, regarding the individual sextants, in females a prevalence of Type II was found in sextant I (36.9%), III (49.2%), IV (60%) and VI (58.5%), Type IV in sextant II (66.2%) and Type I' in sextant V (29.2%) [Figure 4].

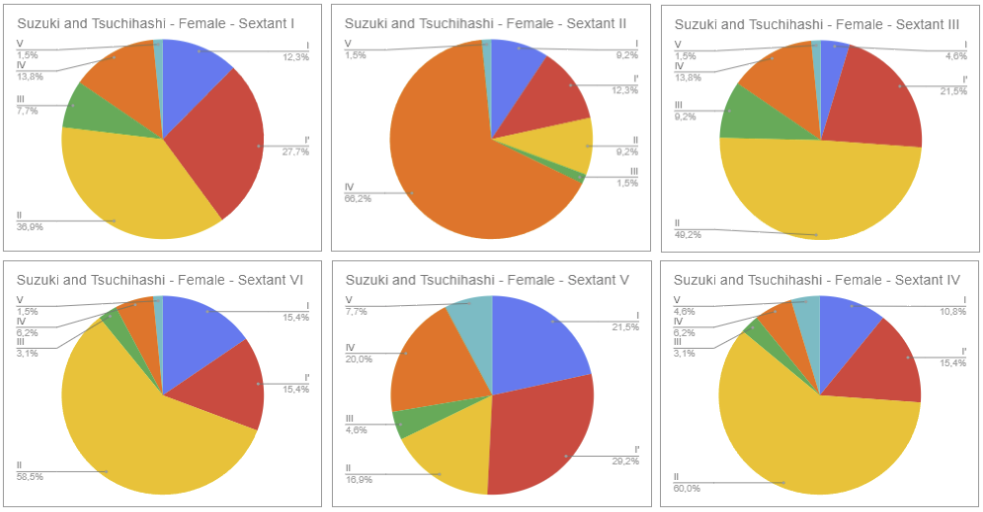

Figure 4. Sextant analysis in female prints.

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Regarding males, the same prevalence of Type II was noted in sextant I (37.2%), III (39.5%), IV (39.5%) and VI (34.9%), while a prevalence of Type IV was observed in both sextant II (34.9%) and sextant V (27.9%) [Figure 5].

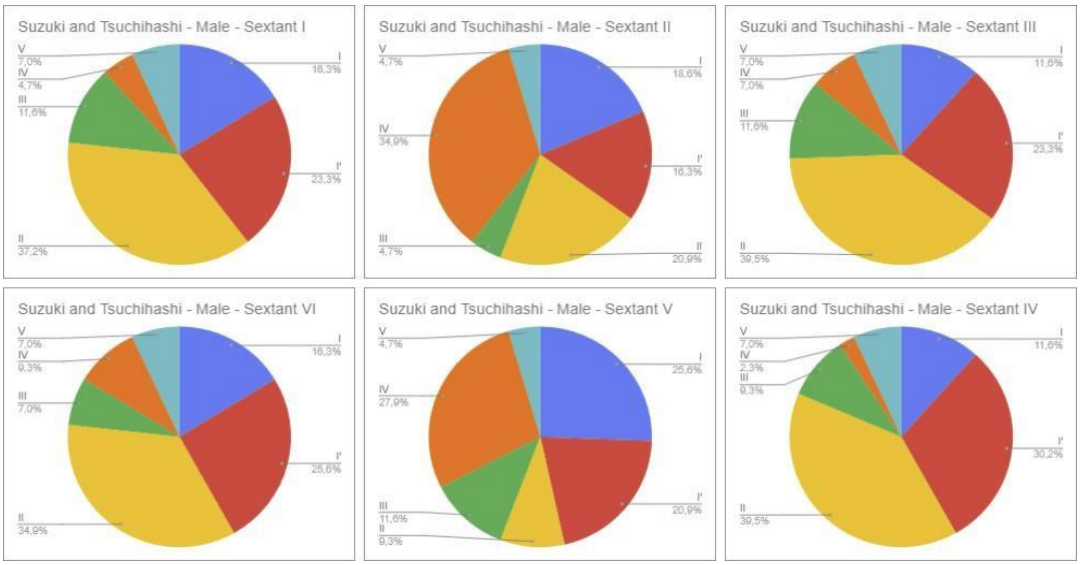

Figure 5. Sextant analysis in male prints.

Analyzing the Type attributed to each print as a result of the predominant Type assessment, we found a higher presence of Type II in both females (50.8%) and males (32.6%) [Figure 6].

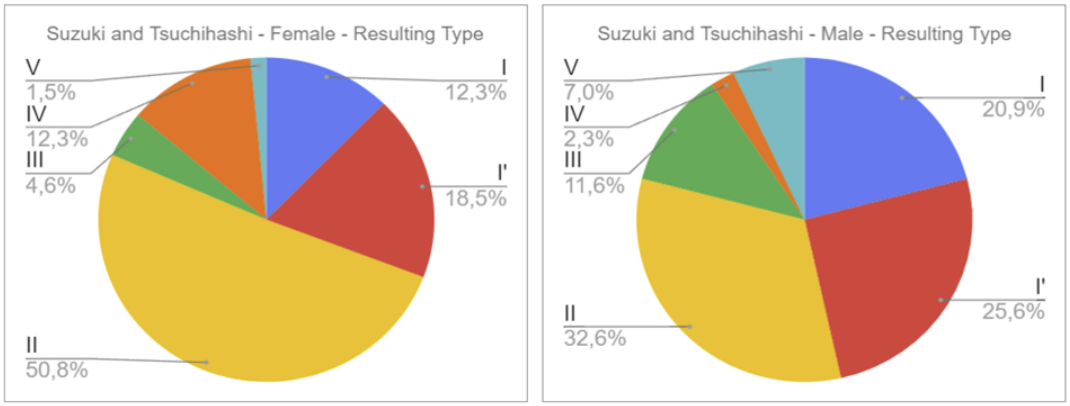

Figure 6. Predominant Type in male and female prints.

After assessing the prevalence of a predominant type in the lip prints taken from males and females, the p-value was calculated to determine the statistical significance of any observed difference. [Table 2].

Table 2. Statistical analysis of predominant Types.

Observed			
Predominant Type	*Female*	*Male*	Total
Type I	8	9	**17**
Type I'	12	11	**23**
Type II	33	14	**47**

Expected			
Predominant Type	*Female*	*Male*	Total
Type I	10,23	6,77	**17**
Type I'	13,84	9,16	**23**
Type II	28,29	18,71	**47**

Test			
Predominant Type	*Female*	*Male*	Total
Type I	0,49	0,74	
Type I'	0,25	0,37	
Type II	0,79	1,19	

Results	
Degree of freedom	5,000
Test stats	10,690
p-value	0,058

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Type III	3	5	**8**
Type IV	8	1	**9**
Type V	1	3	**4**
Total	**65**	**43**	**108**

Type III	4,81	3,19	**8**
Type IV	5,42	3,58	**9**
Type V	2,41	1,59	**4**
Total	**65**	**43**	**108**

Type III	0,68	1,03	
Type IV	1,23	1,86	
Type V	0,82	1,24	
Total			**10,69**

The data also revealed a certain symmetry along the sagittal axis in the Type distribution. In fact, out of all the 108 prints analyzed, 71 (76,68%) had the same Type in sextants I and III, the same being observed in sextants IV and VI in 74 prints (79.92%).It was noted that several of the 108 prints showed a particular concave pattern [Figure 7] for labial wrinkles in the second sextant, concomitant with a Type IV. This same pattern was found in 24 (36.92%) of the female prints and 41 (23.08%) of the male prints, showing a certain sex discrepancy, but with a p-value <0.05. [Table 3]

Table 3. Statistical analysis regarding the presence of concave pattern.

Observed			
Concave pattern	*Female*	*Male*	Total
Present	24	9	**33**
Not present	41	34	**75**
Total	**65**	**43**	**108**

Expected			
Cosmetic traces	*Female*	*Male*	Total
Present	19,86	13,14	**33**
Not present	45,14	29,86	**75**
Total	**65**	**43**	**108**

Test			
Cosmetic traces	*Female*	*Male*	Total
Present	0,86	1,30	
Not present	0,38	0,57	
Total			**3,12**

Results	
Degree of freedom	1
Test stats	3,119
p-value	0,077

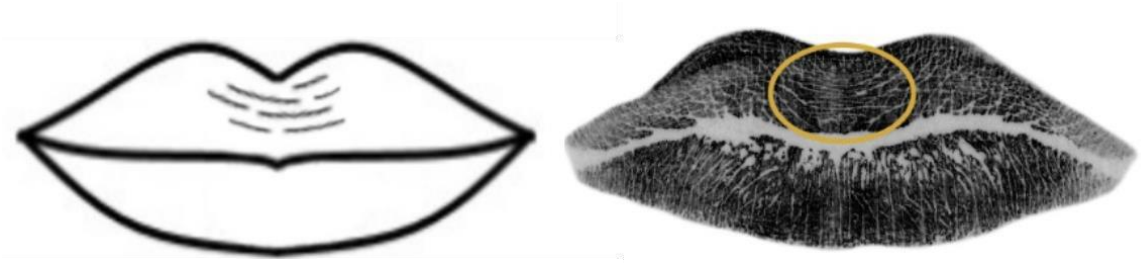

Figure 7. Concave pattern in II sextant.

Another finding was the presence of macroscopically appreciable traces of cosmetic products in the perioral areas. These traces were observed in 30 female prints (35.29%) and 0 male prints. They were looked for in all the 167 prints taken, given that they were detectable regardless of the quality of the print. [Table 4]

Table 4. Statistical analysis regarding the presence of cosmetic traces.

Observed			
Cosmetic traces	*Female*	*Male*	Total
Present	30	0	**30**
Not present	55	82	**137**
Total	**85**	**82**	**167**

Expected			
Cosmetic traces	*Female*	*Male*	Total
Present	15,27	14,73	**30**
Not present	69,73	67,27	**137**
Total	**85**	**82**	**167**

Test			
Cosmetic traces	*Female*	*Male*	Total
Present	14,21	14,73	
Not present	3,11	3,23	
Total			**35,28**

Results	
Degree of freedom	1
Test stats	35,279
p-value	<0,001

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4. Discussion

In our study, we began by analyzing the uniqueness of lip prints and confirmed that each print is distinct from others, consistent with the findings in the literature. In fact literature shows that each lip print is unique and remains unchanged throughout a person's lifetime [16,36–40]. The skin (dry zone) of the lip has all the components of facial skin: sweat and sebaceous glands and hair. The vermilion border, or inner surface of the lip, has neither hair nor sweat glands [17], which could make the detection of latent prints on surfaces inconsistent, as it is not sebum that will be found, but, rather, flaking cells and extremely variable amounts of saliva. This characteristic of lip prints, however, may be of extreme importance, as several studies show that it is possible to extract DNA from latent lip prints from these very residues, creating a synergic effect with cheiloscopy analysis for identification purposes [18–20].

Although our study performed a sextant-based analysis of lip prints, literature reveals various approaches to observing and analyzing these prints. Particular attention must be paid to the way lip prints are observed and analyzed, given that the result of the search for a predominant Type may differ depending on whether the lips are analyzed in their entirety or if attention is paid to a specific portion [47]. Therefore, it is to be expected that a particular Type found when analyzing an entire print may differ from the one found when analyzing a smaller portion [Figure 8].

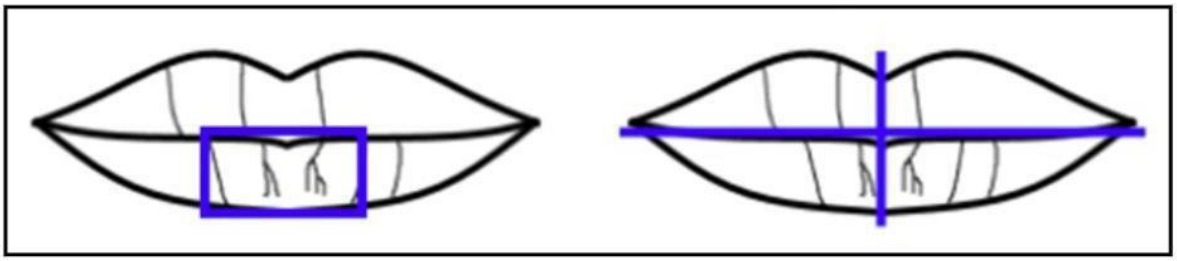

Figure 8. Differences observed based on the subdivision of prints, based on [47].

There is a worldwide inconsistency in the methods used for obtaining and analyzing lip-prints [21], resulting in the need to develop a standardized method.

The method used in our study, that is also the most frequently used in literature, i.e. applying lipstick, and acquiring a print using adhesive tape, would appear to be unreliable as there are many variables [22–24]:

- Lipstick type, viscosity, shade, transferability.
- Amount of lipstick applied.
- Type of adhesive tape.
- Pressure applied and method of application.
- Distortion of the lips during acquisition.
- Setting time.
- Pressure applied with tape.
- Other operator-dependent errors.

The selection of specific materials has emerged as a pivotal aspect in the pursuit of obtaining prints of high quality and reliability. In our study, a variety of lipsticks and adhesive tapes were tested to determine which one was more efficient in capturing accurate impressions. The decision to exclude stick lipstick from consideration was informed by the inherent challenge of achieving uniform coverage across all lip surfaces. Ensuring this uniform coverage is a crucial factor in maintaining the fidelity of obtained prints, an aspect that receives attention in numerous studies [47].

Moreover, our exploration extended to investigating the optimal quantity of lipstick application, it was observed that even minor deviations in application could lead to the obscuration of delicate lip wrinkles, underscoring the need for precision in the printing process. Similarly, the examination of the pressure exerted during detection revealed nuanced insights; the application of gentle pressure

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to the extremities of adhesive tape emerged as the preferred technique, minimizing the risk of distortion while maximizing the retention of crucial details.

With regard to the adhesive tape selection, while differences in thickness may appear trivial, our tests led us to discard tapes with excessively thin profiles. This decision was taken after the realization that such tapes, when applied, tended to form inadvertent folds as they endeavored to conform to the contours of the lips.

Furthermore, it became evident that the distortion observed during acquisitions is not solely attributable to material properties and pressure dynamics. Rather, the very nature of our controlled acquisition environment—a realm characterized by ideal conditions and homogeneous surfaces—introduces a potential disconnect when attempting to extrapolate findings from real-world scenarios. In forensic contexts, for instance, prints may be encountered on diverse surfaces such as straws or cigarettes [25], each presenting unique challenges that must be navigated to ensure accurate analysis and interpretation.

Finally, the impact of variations in lipstick setting time on print quality was noted. Prints taken immediately post-application often manifested themselves as smudged and indistinct, showing the need for careful consideration of temporal factors in the printing process [48,49].

Other studies also observed that the position of the lips during the capture of the print will influence the readings. Qualitatively significant differences were found between prints taken with the lips closed and those taken with the mouth open. Closed lips displayed more defined grooves which were easier to interpret. The absence of front teeth may also cause difficulties in developing prints [26,44].

In summary, this inquiry into materials, techniques, and environmental factors aims not only to refine current methodologies but also to lay the groundwork for future advancements in forensic science and investigative practice.

Our study showed that not only operator-dependent errors are frequently encountered during the process of obtaining prints, but they also persist as well during the subsequent analysis of these prints. The inherent challenge lies in the precise attribution of a specific Type to a print or even a segment of it, thereby rendering the resultant conclusions only partially dependable.

While it was possible to identify a particular type for an entire lip print in some instances, we often found that the Suzuki and Tsuchihashi classification is overly simplistic and not universally applicable, except perhaps in specific areas such as portions of the lips. Moreover, the Suzuki and Tsuchihashi classification system tends to be didactic, meaning it offers broad categorizations that may not fully capture the nuanced variations present in lip prints. While it can be useful as a basic framework for understanding lip print patterns, it may fall short when attempting to provide detailed or comprehensive analysis because the primary challenge lies in the fact that lip prints can exhibit a variety of patterns within the same area [15,16]. Considering this, and given the strict conditions of this study, the reliability of the data obtained regarding the Suzuki and Tsuchihashi classification is uncertain.

In our study we used Suzuki and Tsuchihashi classification, and although this is the most used in the literature [45,46], there are several classifications proposed by other authors such as:

- Dr. Santos, who in 1966 divided the nature of wrinkles and grooves into simple and compound types that were subdivided in four different groups. Dr. Santos also classified lips as thin, medium, thick and mixed type and reported various types of commissures. [27–29].
- A French scientist, Renaud, who studied 4000 prints and described 10 types of lip prints, designated by letters from A to J, capital letters for upper lips and lowercase for bottom lips [30]
- Kasprzak, who determined the pattern based on the numerical superiority of properties of the lines on the fragment. After the patterns of lines had been established, a first catalog of individual features was prepared and 23 types of individual properties were differentiated [31,32].

In our study, we did not find statistically significant data linking sex to the characteristics of lip prints ($p>0.05$), except for the presence of materials in the perilabial areas, such as residues from cosmetic products, which were observed with statistical significance during macroscopic analysis. This suggests that these findings could be further validated through chemical analysis of the samples.

 doi:10.20944/preprints202406.0932.v1

The literature reports highly inconsistent results regarding the potential use of cheiloscopy for sex determination, there is no consensus in the literature about the presence of a predominant Type that could reliably differentiate male from female individuals [47]. Even among the studies that claim statistically significant differences, there is considerable disagreement and lack of univocal results. This underscores the complexity and variability in cheiloscopic analysis and suggests that further research is necessary to clarify these discrepancies.

Authors should discuss the results and how they can be interpreted from the perspective of previous studies and of the working hypotheses. The findings and their implications should be discussed in the broadest context possible. Future research directions may also be highlighted.

A major limitation of the method is also the absence of a database, making only 1-1 matching of prints possible and possibly leading to deductions about the individual's sex.

5. Conclusions

This study highlights how cheiloscopy has great potential in forensics, but the absence of standardized methods to minimize errors in obtaining and interpreting prints makes its application limited. The need to develop a standardized capture protocol is evident. After an analysis of the major problems encountered with the lipstick method, it was concluded that a change of approach was needed. A photographic approach could be used, establishing capture parameters for the lips, thereby minimizing operator-dependent errors. In addition, the application of a digital analysis system is also necessary: a special software that can analyze a print, bypassing human error, would provide more reliable and statistically significant data and would also reduce the time needed to analyze the prints.

We consider it important to pursue a goal of convergence to define a single universal classification. This would represent an essential basis not only for guiding future research but also for facilitating comparison and understanding among scholars in the field.

Although our analyses suggested the presence of some differences between men and women in the morphology of lip prints, these differences were not sufficiently consistent or pronounced to justify the use of cheiloscopy as a reliable method for gender identification. This conclusion is supported by the existing literature, which presents discrepancies and variable results regarding the correlation between lip print characteristics and the individual's sex [47].

In addition, it would be interesting to evaluate the long-term effects of certain cosmetic procedures, such as the use of lip fillers with hyaluronic acid, which could have an impact on lip wrinkles, given that a total restitutio ad integrum of the lips after the treatment period has not been confirmed [14]. Although no studies have yet been conducted regarding this interaction, it could be a promising area for future investigation as it could contribute to a more complete understanding of the variations in lip wrinkles and their determinants.

The results obtained in this study need to be confirmed in future studies. The aim must be to develop a single acquisition protocol and to analyze the prints using digital methods. It would be interesting to call upon the study participants to perform a further survey through photographic means and to analyze the data with dedicated software in order to assess any discrepancies with the previously acquired data.

Author Contributions: Conceptualization, G.A.S. and P.M.; methodology, E.D.V., F.M.S., S.V., L.R., A.T.; software, ; validation, A.A., D.A. and S.Z.; formal analysis, E.C.; investigation, E.D.V., F.M.S., S.V., L.R., A.T. ; data curation, X.X.; writing—original draft preparation, E.D.V., F.M.S., S.V., L.R., A.T.; writing—review and editing, E.C., G.B.; supervision, P.M., G.A.S. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and approved by the Institutional Review Board or Ethics Committee University of Palermo Policlinico “Paolo Giaccone”.

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: Data can be requested to alessandro.scardina@unipa.it.

Conflicts of Interest: The authors declare no conflicts of interest.

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