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Can Lip Prints Change Overnight?: A Study of Lip Print Stability Across Day and Night as a Forensic Identification Tool

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ABSTRACT

Aim

Lip prints have long been considered unique and stable over time, causing a boost of cheiloscopy research to understand the potential of lip prints for forensic identification. While studies looking at lip print stability over time are common, this study investigates the stability based on the day and night phenomenon.

Methodology

Lip prints were taken from 200 participants from the campus population using the standardised paper technique, wherein lip prints were made on A4 papers then digitised using a high- resolution scanner. Lip prints similarity percentage were formed by comparison of the prints collected at the morning and evening, then analysed using Contrastive Language-Image Pre- training (CLIP) image

analysis model. Statistical analysis included repeated-measures ANOVA to compare the lip print similarity percentage obtained at Day 1, Day 7 and Day 14. Intra-class correlation coefficient (ICC) is used to test the reliability of the CLIP model to analyse lip print images.

Results

Repeated measures ANOVA indicated significant variation in lip prints similarity percentage obtained at Day 1, Day 7 and Day 14. The intraclass correlation coefficient (ICC) was rated 0.649, between fair and good.

Conclusion

The study concludes that lip print morphology may not be as stable over short time intervals as previously assumed, and this variability should be considered in forensic evidence collection.

Keywords: Cheiloscopy, Lip Prints, Deep Learning, Digital Analysis, ICC

Abbreviations

CLIP	Contrastive Language-Image Pre-training
ICC	Intra-Class Correlation
SD	Standard Deviation
SPSS	Statistical Package for the Social Sciences

Introduction

Lip print analysis raises questionable potential for forensic identification, known as cheiloscopy, and faced a turning point after the study by Tsuchihashi in 1974.[1] Lip prints are considered relatively stable over time and unique, similar to fingerprints. [2-5] However, the assumption of temporal stability has not been sufficiently challenged, particularly over short-term intervals such as within the day and night of the same day.

Previous studies have primarily assessed long-term consistency of lip prints and their uniqueness among populations over long time intervals. [6, 7] However, these studies are conducted with manual observation and no quantitative analysis was utilised to measure lip print similarity, thus invalidating efforts to ensure lip prints can adhere to the Daubert's standard of evidence. [8]

This study aims to explore the short-term variability of lip prints across different times of day using a novel digital approach. We employed Contrastive Language-Image Pre-training image analysis model (CLIP), a deep learning model capable of analysing images and calculating similarity percentage based on the potential changes of lip print patterns from day to night. Our hypothesis is that the lip print morphology may vary during the morning and evening due to natural biological fluctuations based on the circadian rhythm.

Need for 'this' study

Despite most studies describing lip prints as being stable, there is only a study measuring lip print stability quantitatively using similarity percentage. [9] This study presented new insights to lip

print stability by classifying the similarity rate of lip print patterns into three categories based on their similarity percentage. Based on the results, the lip print similarity percentage was classified as medium (73.8%), meaning there may be potential changes in lip print patterns. The authors would like to provide new insights to these lip print pattern changes.

On the other hand, digital analysis needs to be utilised in lip print analysis to accurately measure potential changes in lip prints. The application of digital analysis methods in forensic science allows better visualization, easier identification, and complete recording of images. [10] Our research findings would be able to provide new insights in measuring lip print similarity percentage.

This study also presents a new perspective to measuring lip print stability. While time is usually the parameter used, the authors believe potential changes of lip print patterns based on the circadian clock system. The circadian clock plays an important role in oral and maxillofacial physiological and pathological processes. [11] In this present work, we studied the similarity of lip prints collected in a university campus population during the day and night phenomenon.

Methodology

This descriptive cross-sectional comparative study of 200 individuals was conducted periodically from November 2024 to March 2025. Every subject underwent a sample collection period of 2 weeks where the prints were collected on Day 1, Day 7 and Day 14. Collection sessions were divided into morning and evening.

The suitable sample size, 200 was calculated based on these factor considerations as suggested by the Cohen's D convention. [12] The factors are as below:

i) Effect size

Set at a value of 0.2, a small effect size magnitude is used to observe the potential small variations in lip print patterns.

ii) Significance level, α

Set at 5% as the majority of studies in this field which has a similar motive of observing lip print changes. [13, 14, 15]

iii) Power

Set at 80% as the majority of studies in this field with similar motives mentioned as above. [13, 14, 15]

The G*Power statistical software is used to calculate an appropriate sample size. The supporting figure from the software is as below where the suggested sample size is 199, and rounded up to 200 subjects **[Figure. 1]**.

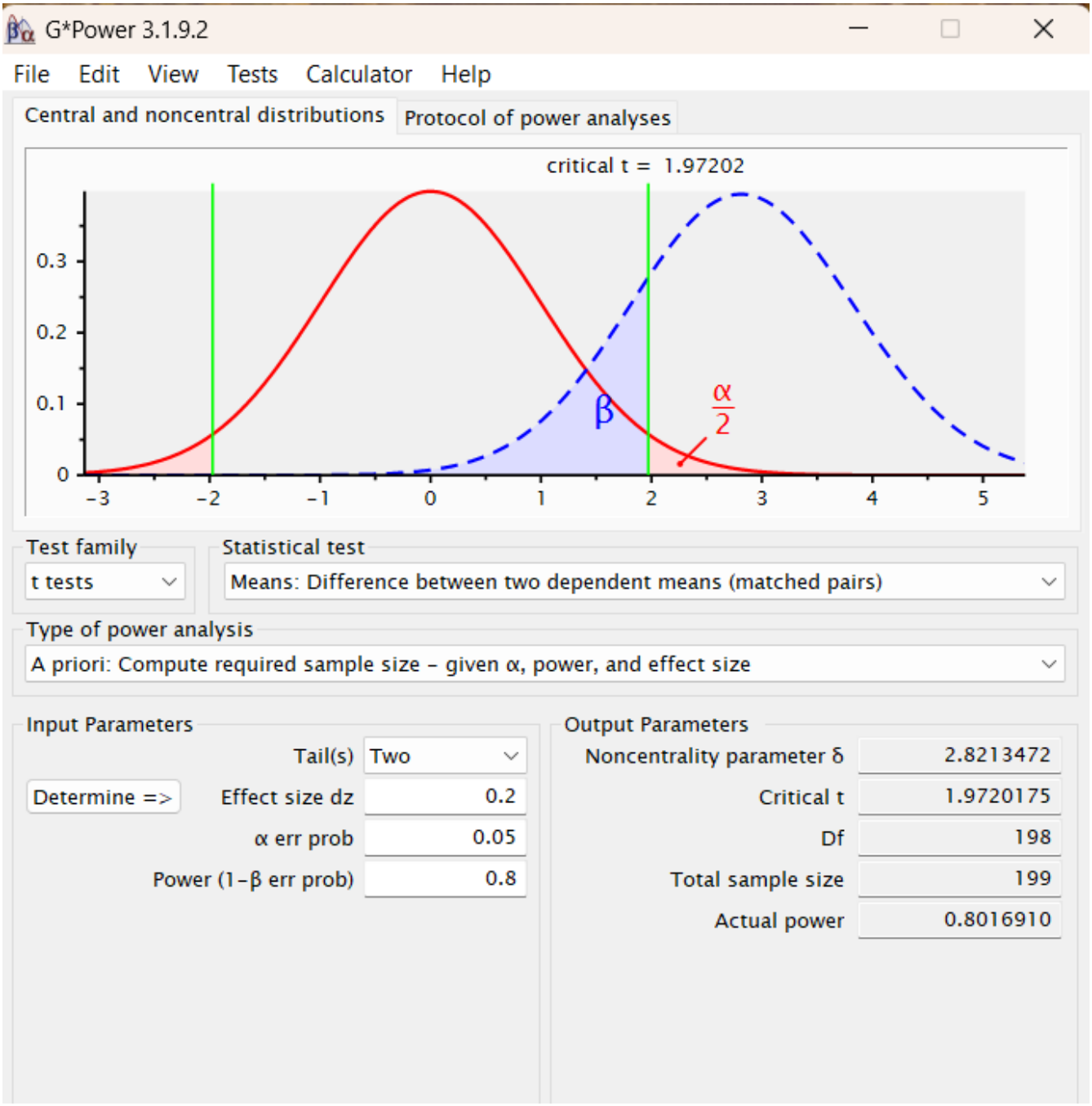

Figure 1. Sample size calculation on G*Power software

The Ethical Committee from Universiti Kebangsaan Malaysia approved this study. It was made sure all subjects were above 18 years old. Written consent from the subjects were taken before collecting their lip print as samples. The subjects consisted of both students and staff. 57 of the subjects were staff while 143 were students. From the 143 students, 50 were 2nd year undergraduate students, 83 were final year undergraduate students while 40 were postgraduate students as mentioned in **[Figure 2]**.

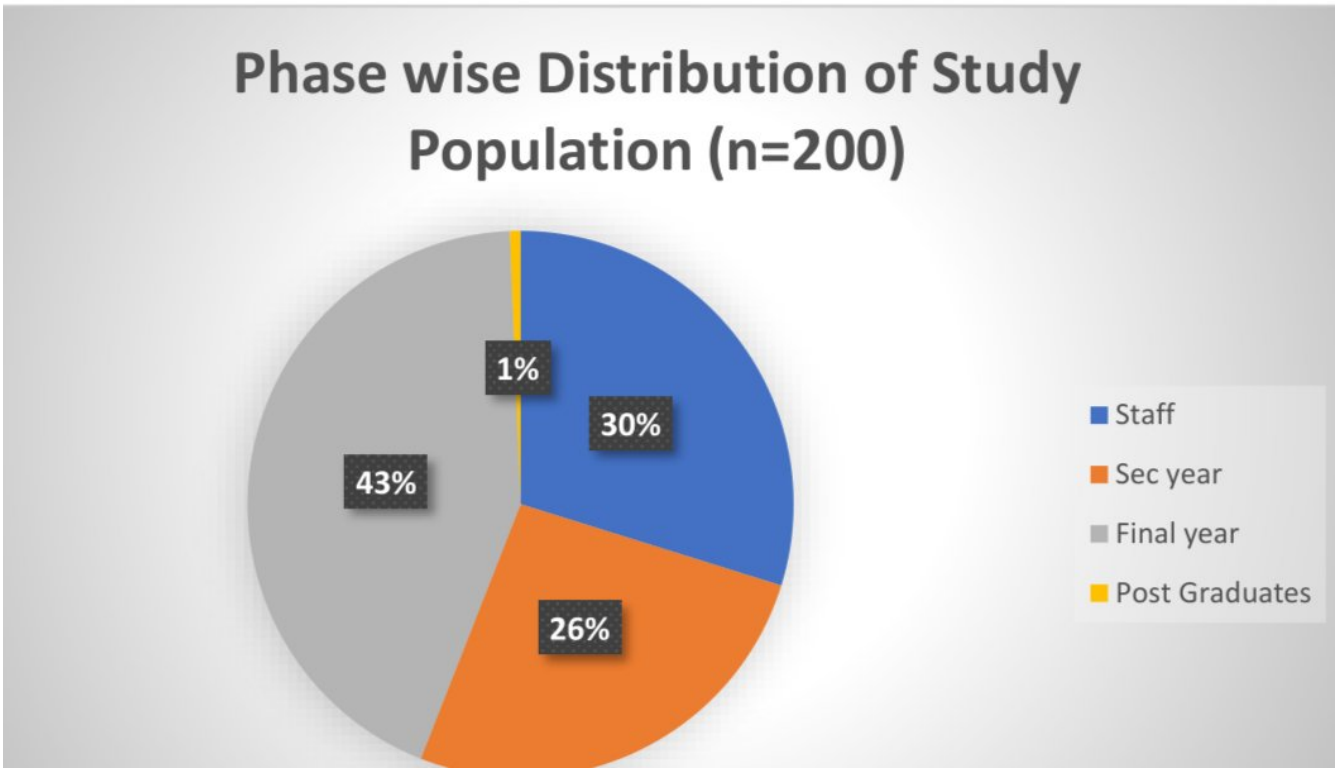

Figure 2. Phase wise Distribution of Study Population

From the consent form, several sociodemographic parameters related to the study such as sex, race, university campus and age are included in a table **[Table 1].**

Sociodemographic profile of participants (n=200)			
Characteristics		**Frequency**	**Percentage (%)**
Sex	Male	75	37.5
	Female	125	62.5
Race	Malay	187	93.5
	Chinese	10	5
	Indian	3	1.5
Age	Young (20-40 years)	145	72.5
	Old (40-60 years)	55	27.5
Campus	Health Science Campus	25	12.5
	Medicine Campus	115	57.5
	Main Campus	60	30

Table 1. Sociodemographic profile of subjects

Smeared lip prints, with unclear structure visualisation, or lip prints which were too dark due to excess lipstick smear were not included in analysis. Excluded lip prints were lips and/or nearby surrounding structures with inflammation/trauma, lips with malformation or deformity, lips with surgical scars, lips with ulcers, lips with wounds, lips with abnormalities, dry and chapped lips, and history of smoking cigarettes/vaping.

Materials

Lipstick (crimson poppy shade, non-glossy; IN2IT brand), white A4 sized papers (Double A brand, 80 gms), facial tissues (Premier brand, 2 Ply), facial wipes (Guardian brand), disposable lip brush (Cleo brand), digital printer (Brother DCP- J100 brand, 600 dpi).

Software

Google Colaboratory, Contrastive Language-Image Pre-training (CLIP) image processing model

Sample collection

1. Sample preparation

The lip prints were compared from the same subject at Day 1, Day 7, and Day 14. The collection sessions were held at the morning and evening of these days. The subjects cleaned their lips with facial wipes. A thin lipstick layer was applied using a clean disposable lip brush to the lips in a singular motion. [16] The subject then rubbed their lips to spread the lipstick more uniformly. [17] A plain white paper was used to take a print while minimal pressure was applied with the index finger (by the researcher). The slightest movement of the lips while recording print can smear the samples hence, the subject was advised to remain still and maintain the position of their lips so they could be adequately traced. Facial wipes were used to clean the lips after the procedure. Three lip prints were collected at each collection period where the best print in terms of clarity was chosen for analysis.

2. Image Digitization

Three replicated lip print samples were taken from each subject at every collection period, where the clearest print would be digitised with a high-resolution printer (600 dpi). All selected lip prints were scanned in grayscale (8-bit) format. The digital images were standardized to a uniform dimension (550 x 232 pixels, 72-point resolution) and saved in Tag Image File Format (TIFF). These images were inputted in the Google Drive folder.

Digital Analysis

A modified Python code is used train the CLIP model for image recognition. The code is run on Google Colaboratory (Colab) platform. This study just uses the CLIP model alone for all image analysis tasks, making it quite different compared to traditional image recognition studies that needs different models for different tasks. [18, 19] In this study, the image encoder feature and the zero-shot feature from the CLIP model are widely used for feature extraction and classification of these lip print images. The zero-shot feature is useful for model training to match up the lip prints between two subjects.

The comparison between the lip print collected in the morning and evening of the same day was quantified as a percentage based on the cosine similarity of their respective feature vectors **[Figure. 3]**. A value closer to 100% indicates a higher probability of a match between both prints.

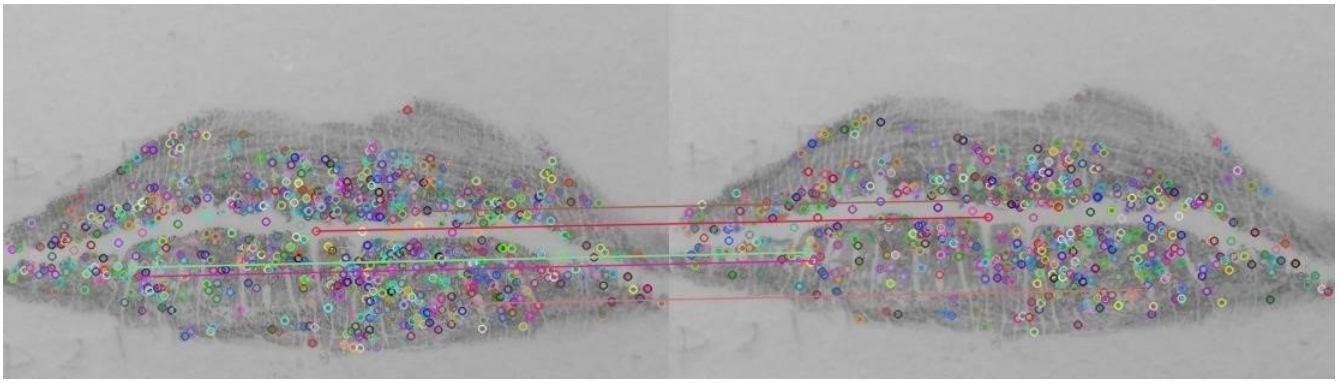

Figure 3. Detection of lip print's unique features with the CLIP image analysis model

Prior to analysis, control samples were processed as a validation and testing step to ensure accuracy. A negative control (clean blank paper) was used to establish a baseline. A positive control (lip print from an existing database) was used to verify script functionality. The workflow is as below **[Figure. 4].**

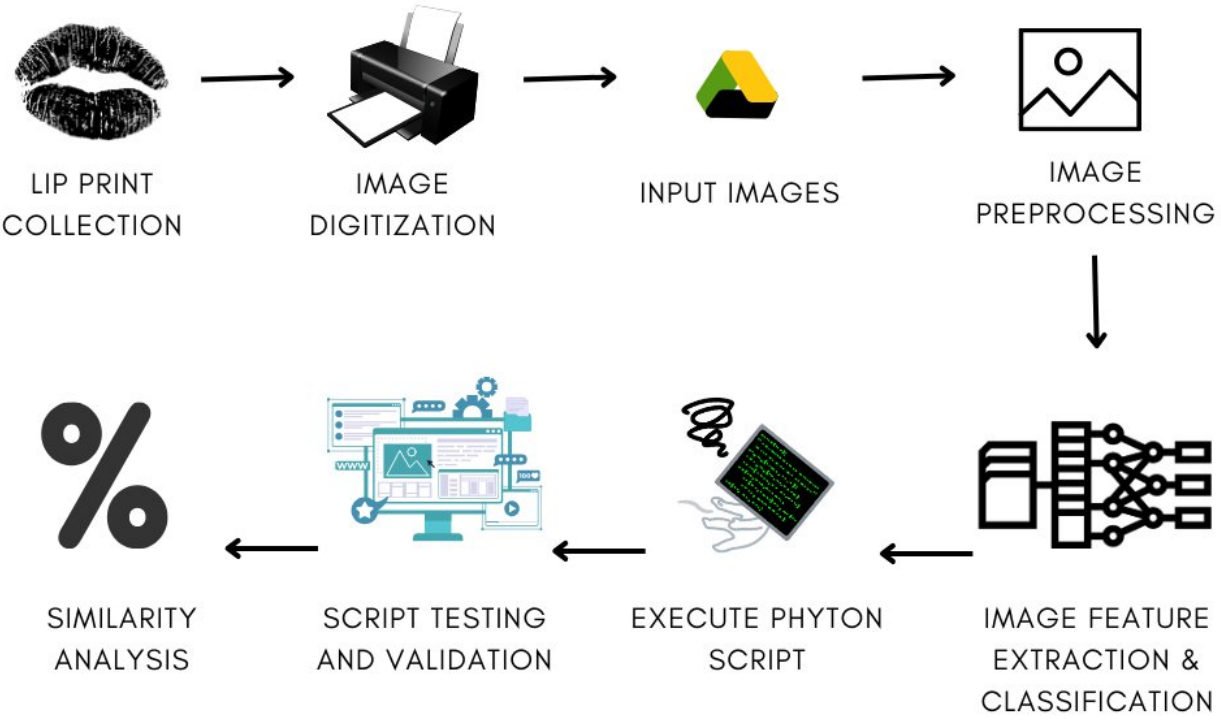

Figure. 4 Methodology Workflow

Statistical Analysis

The software used for statistical purposes was the Statistical Package for Social Sciences (SPSS) software, IBM manufacturer, Chicago, USA, version 29.0. The similarity index of lip prints collected day and night on Day 1, Day 7 and Day 14 was analysed with repeated measures ANOVA. The

reliability of the CLIP model was determined using Interclass Correlation Coefficient (ICC). A threshold of 0.75 was used to define good reliability. [20]

Results

Based on the CLIP analysis model, the mean of lip print similarity percentage obtained after comparing the lip prints collected on the day and night of Day 1, Day 7 and Day 14 were plotted in a graph as below **[Figure 5]**.

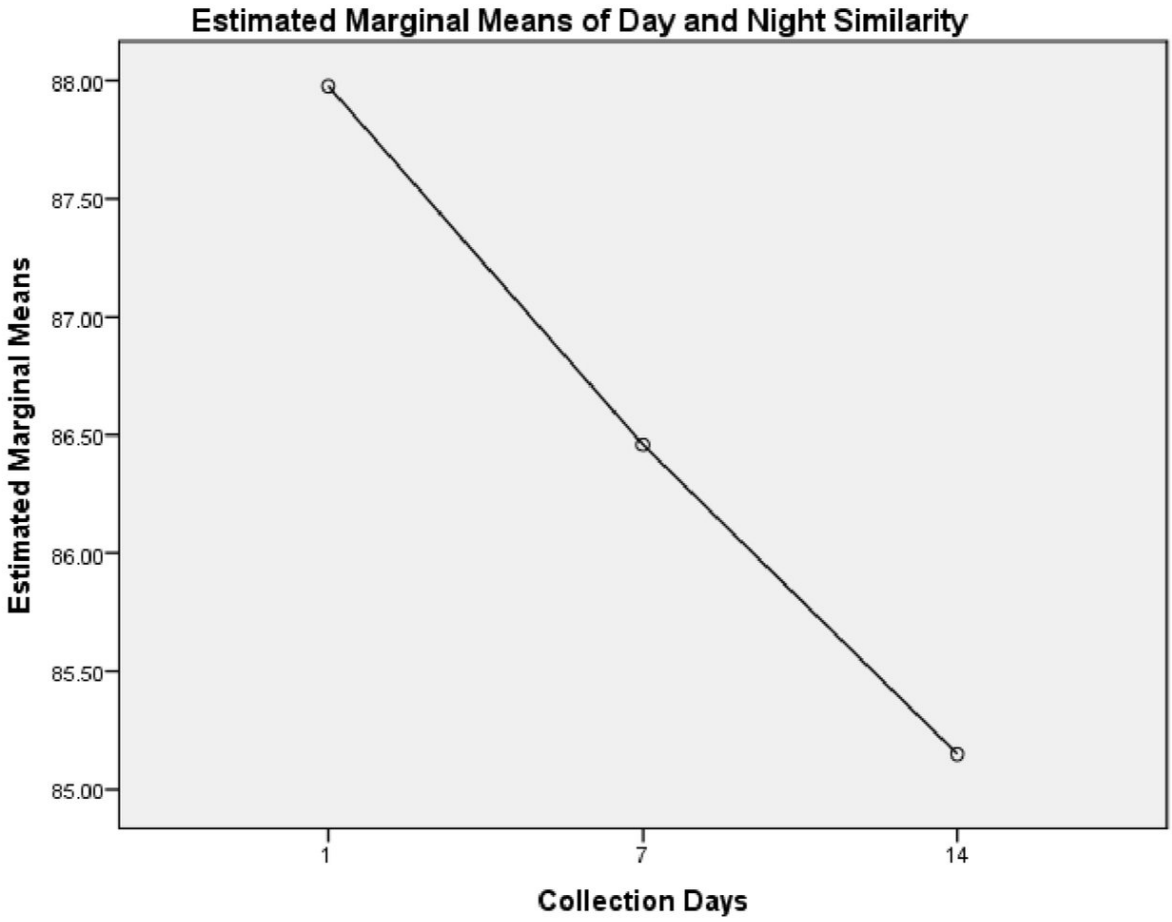

Figure 5. Similarity Scores graph across Day 1, Day 7 and Day 14

The mean ± SD values of day and night similarity percentage values at Day 1 (87.97±7.08), Day 7 (86.46±6.76) and Day 14 (85.15±7.94) shows a slight decrease in similarity as the week progresses.

Repeated Measures ANOVA test evaluated the effect of the day and night phenomenon on lip print similarity. Based on the Mauchly test with rejected sphericity, $\chi^2(2) = 6.795$, $p < 0.05$, the degrees of freedom were corrected using Greenhouse-Geisser, ($\varepsilon = .967$). ANOVA results portrayed a significant difference of the day and night phenomenon towards lip print similarity, $F(2,398) = 9.163$, $p < .05$, partial $\eta^2 = .044$.

Reliability test type used is the test-retest reliability, to assess the reliability of the CLIP image analysis model in analyzing lip prints. The ICC test is conducted using a two-way mixed-way model with a single rating mode. The generated value was 0.649, which is in between the fair and good range (0.556-0.726).

Discussion

The authors aim to challenge the assumption of lip prints temporal stability which has always been considered consistent and does not change over time. [6, 7, 9] We aim to challenge this using digital analysis as a more robust method and by measuring lip print stability, using a different physiological factor such as the circadian rhythm which strongly influences the morning and evening phenomenon.

Our results challenge this assumption of lip prints stability, especially in high- resolution digital contexts. Lip prints were analysed with a customised Python script to train the CLIP image analysis model to measure the similarity percentage of lip prints taken on different collection times over several days. The Python script was run on Google Colab for free of charge. Using this platform, we are not required to install specific modules to run codes and the platform is user friendly to non-technical individuals who do not possess programming knowledge. [21] The computing power is also not dependant on your machine but on Google servers, which ensures performance of your local machine or computer. [22] The data were also obtained in a week. The whole analysis method ensured optimization to lip print analysis as results were obtained in a short period of time with no costs. As a whole, the study also demonstrates the feasibility of using artificial intelligence (AI) tools like CLIP for forensic image analysis.

Our findings suggest that lip print morphology may exhibit changes from day to night in a same day. The reduced similarity observed between the day and night prints supports the hypothesis that circadian or physiological factors may influence lip features. Cortisol, a steroid hormone crucial in the body's metabolic reaction to stress are interconnected to the circadian rhythm and can influence human facial appearance. [23] Cortisol levels normally exhibit a circadian pattern throughout the day, peaking in the early morning and dropping in the late afternoon and night. [24] Therefore, the facial features may appear slightly plumper and more defined in the morning to increase alertness. As for the evening, the skin is more relaxed because of the low level of cortisol. [25, 26] This factor will indirectly influence the perception of the lips, producing different lip print changes throughout the day.

The moderate ICC values suggest that lip prints are not as temporally robust as previously assumed. This has important implications for forensic identification, where the assumption of immutability underpins evidentiary reliability. As there is no standardised procedure in the collection method, the procedure should be explored. In this study, the lip prints were replicated three times after an one-time lipstick application. The application of lipstick for every lip print transfer on paper should be explored to see if it increases the accuracy of lip print similarity.

Limitations of the study include the usage of small sample size, potential artifacts from lipstick application and scanning inconsistencies. Further research with larger, more diverse populations and controlled imaging protocols is warranted. Changes of lip prints during the day and night phenomenon should also be explored with other sociodemographic factors such as sex which are also affected by the circadian rhythm to in future research.

Conclusion

Lip prints may not be entirely stable over short time intervals, particularly between day and night. This finding raises important considerations on the forensic use of lip prints for personal identification. Future studies should investigate biological and environmental factors that may affect lip morphology and explore standardized protocols to enhance reliability.

Ethical committee clearance

Obtained [JEP-2024-947, dated: 21-11-2024]

Conflict of Interest

None

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Faculty of Health Sciences, Universiti Kebangsaan Malaysia

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What's new in our Paper

1. What is already known on this topic?

Lip prints stability are quite known in cheiloscopic research, as being unchanged over long time intervals, usually across several months. Time intervals are usually the condition used to measure temporal stability of lip prints. Most studies conduct lip print stability research using manual observations.

2. What question did this study address?

As specific lip print collection times in previous research are unknown, authors decided to measure lip print stability across day and night. Till date, there are no studies investigating lip print stability over one-day intervals within the morning and evening of the same day. Digital analysis is also used by utilizing the CLIP, a deep learning image analysis model to generate similarity scores of lip prints changes from the morning to the evening of the same collection day.

3. What does this study add to our knowledge?

Authors discovered that lip prints pattern do changes across day and night and believe this is due to the natural biological fluctuations based on the circadian rhythm. We also discovered a novel digital analysis method, using a custom Python code on Google Colaboratory as a language to train the CLIP model to conduct various image analysis tasks such as lip print recognition, lip print feature extraction and lip print similarity match. This whole procedure was conducted at no cost and used a very short period of time for analysis.

4. Suggestions for further development

We suggest future research into identifying a potential correlation among sex and day and night lip print pattern changes as there is much evidence about the different hormonal fluctuations among male and female. More quantitative and robust studies like these would determine the feasibility of lip prints as a potential identification tool that may be accepted by the judicial system.

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