

Developing the Latent Lip Print Using Hibiscus Sabdariffa (Roselle) Extract for Morphological Pattern Analysis in Thai Population

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Abstract

Background: A latent lip print can be important evidence, and specific substances are needed for the development of a latent lip print. However, collecting the lip print may be difficult due to people who have sensitive skin with lipsticks and almost men who do not wear lipstick. Moreover, the enhancement methods of latent lip print are still lack of standardization and consistency.

Aim: To develop a new staining method for the study of the lip print patterns Thai population.

Method: The latent lip prints were collected from 24 males and 24 females. The ability of Roselle extract was investigated for developing latent lip prints on cigarette paper. The developed lip print was used for the morphological pattern analysis using Suzuki and Tsuchihashi's classification criteria combined with Autodesk SketchBook.

Result: The results revealed that Roselle extract exhibited potential as a staining dye for the development of latent lip prints on cigarette paper. Among the 24 males and 24 females, Type I was found to be most common and was present in 54.55% of the volunteers, followed by Types I', II and IV (21.12%, 14.58%, and 9.03%, respectively). The least common patterns were Types III and V, each represented in 0.35% of the total volunteers.

Conclusion: latent lip print could be used for study of lip print pattern by stained with Roselle extract. Moreover, a digital method using Autodesk SketchBook serves as an alternative method that provides ease in storing and identification of lip print pattern.

Keywords: Forensic Sciences; Natural dye, Latent lip print, Digital method, Cheiloscopy.

Introduction

The analysis of lip prints is referred to as cheiloscopy. Individual lip prints can be identified by differences in the pattern of grooves that develop during the sixth week of intrauterine life¹. Moreover, individual lip prints are unique which possible use for personal identification²⁻³.

A lip prints can be visible or latent which may serve as a physical evidence. A good quality lip print requires a staining dye for the development of the latent lip print. When a sprinkle of black fingerprint powder was used, some latent lip prints developed lip print patterns⁴. Aluminum powder, silver metallic have also been used to develop latent lip prints². The lysosome dyes (Sudan III, Sudan black, and oil red O) dissolve with lipids

produced from the sebaceous glands of the lips². These dyes are also effective on both, non-porous and porous surfaces⁵.

Several studies have reported that synthetic dyes can cause allergies and cancer. In contrast, natural dyes are not only safe for humans, but also environment friendly⁶. Interestingly, indigo, a plant-derived natural dye, has been used previously to develop latent lip prints⁷. However, sprinkling of the powdered dye could destroy the groove patterns of the lip print. Moreover, collecting the lip print may difficult due to people who have sensitive skin with lipsticks and almost men who do not wear lipstick. Thus, it is important to develop a new method for enhancing the latent lip print.

Hibiscus sabdariffa, also known as roselle which can be used as food, traditional medicine and fiber⁸. The most Roselle quality is from Thailand and Sudan. However, the main world suppliers are China and Thailand⁸. Besides its advantage for a food or traditional medicine, several studies have been used the Roselle extract as staining solution for histological study^{6, 9-12}.

This study aimed to use a Roselle extract as a solution for the development of latent lip print and their application for study of the lip groove pattern. Moreover, this study also aimed to assess a digital technology to manage and analyze lip print patterns.

Materials and Methods

Preparation of staining solutions

A fresh Roselle were purchased from the Iyara Market, Pathumthani province, Thailand. 10 grams of roselle dye were dissolved in 100 mL of 70% ethanol and sonicated for 30 minutes at 60°C. Finally, the

extracted solution was filtered using Whatman filter paper No. 1. The extracted Roselle solution was retained in glass bottles at 25-28° C. Crystal violet stain was used as a reference dye.

The morphological pattern study of the latent lip print.

Lip prints were collected from 24 males and 24 females from Department of Medical Technology, Faculty of Allied Health Sciences, Thammasat University, Thailand, whose aged between 18 - 35 years.

For latent lip print collection, the lips were cleaned with normal saline solution and air-dried. Next, Vaseline petroleum jelly was applied uniformly on the lips and a lip impression (as three replicates) was taken on a 7 × 5.5 cm cigarette paper for 15 s. The time interval between the first and subsequent prints was 1 min. The latent lip prints were stained by dipping for 25 s and air-dried.

The stained latent lip prints were scanned using Epson V39 at a resolution of 1200 dpi. The scanned pictures were viewed and analyzed by Autodesk SketchBook. Each latent lip print picture was divided into six quadrants (**Fig.1**). A color code was used for represent each type of lip print pattern as orange, red, green, yellow, white and gray for lip print type I, I', II, III, IV and V, respectively (**Fig.1**). The stained latent lip prints pictures were classified according to the morphological pattern of the lip print using Suzuki and Tsuchihashi's classification criteria. Pearson's chi-squared test was used to compare the frequency of the lip groove types between the Thai population and previously reported populations^{2, 13, 15, 19, 23-26}. Statistical analyses were conducted using SPSS version 22.0 (IBM Corp., Armonk, NY, USA).

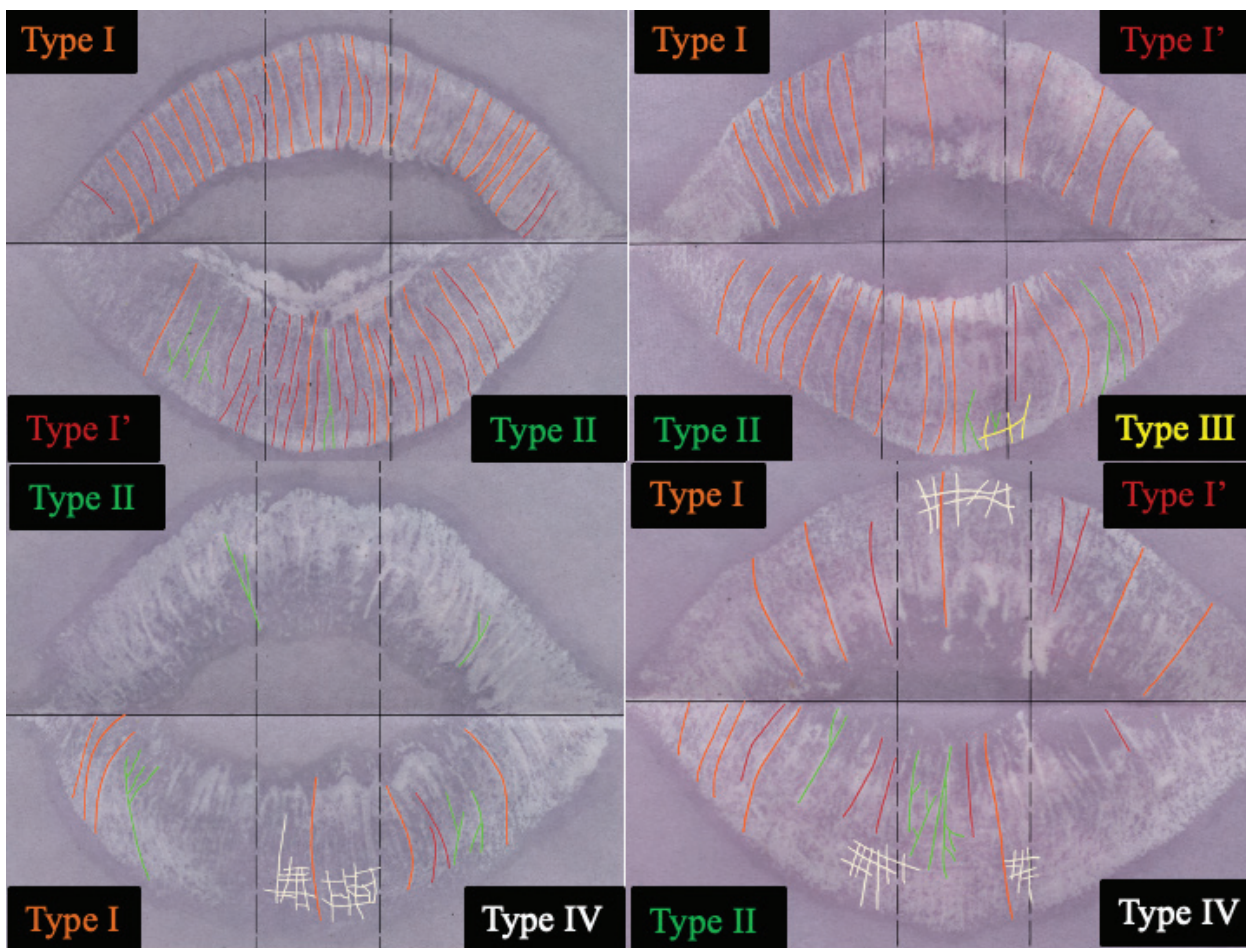


Fig. 1 Analysis of Lip print pattern with color code using Autodesk SketchBook.

Results

The morphological pattern study of the latent lip print

A total of 48 latent lip prints were collected on cigarette paper (Figs. 2A, 2C, 2E and 2G) and stained with the Roselle extract (Figs. 2B and 4F) or crystal violet stain (Figs. 2D and 2H). The results showed similar color intensity and groove clarity of the developed lip prints following staining with Roselle extract and crystal violet.

Each lip print collected from the volunteer was divided into six quadrants, and each quadrant was analyzed independently by drawing a color line in Autodesk SketchBook. Among the 24 males and 24 females, Type I was found to be most common and was present in 54.55% of the volunteers, followed by Types I', II and IV (21.12%, 14.58%, and 9.03%, respectively). The least common patterns were Types III and V, each represented in 0.35% of the total volunteers (Table 1).

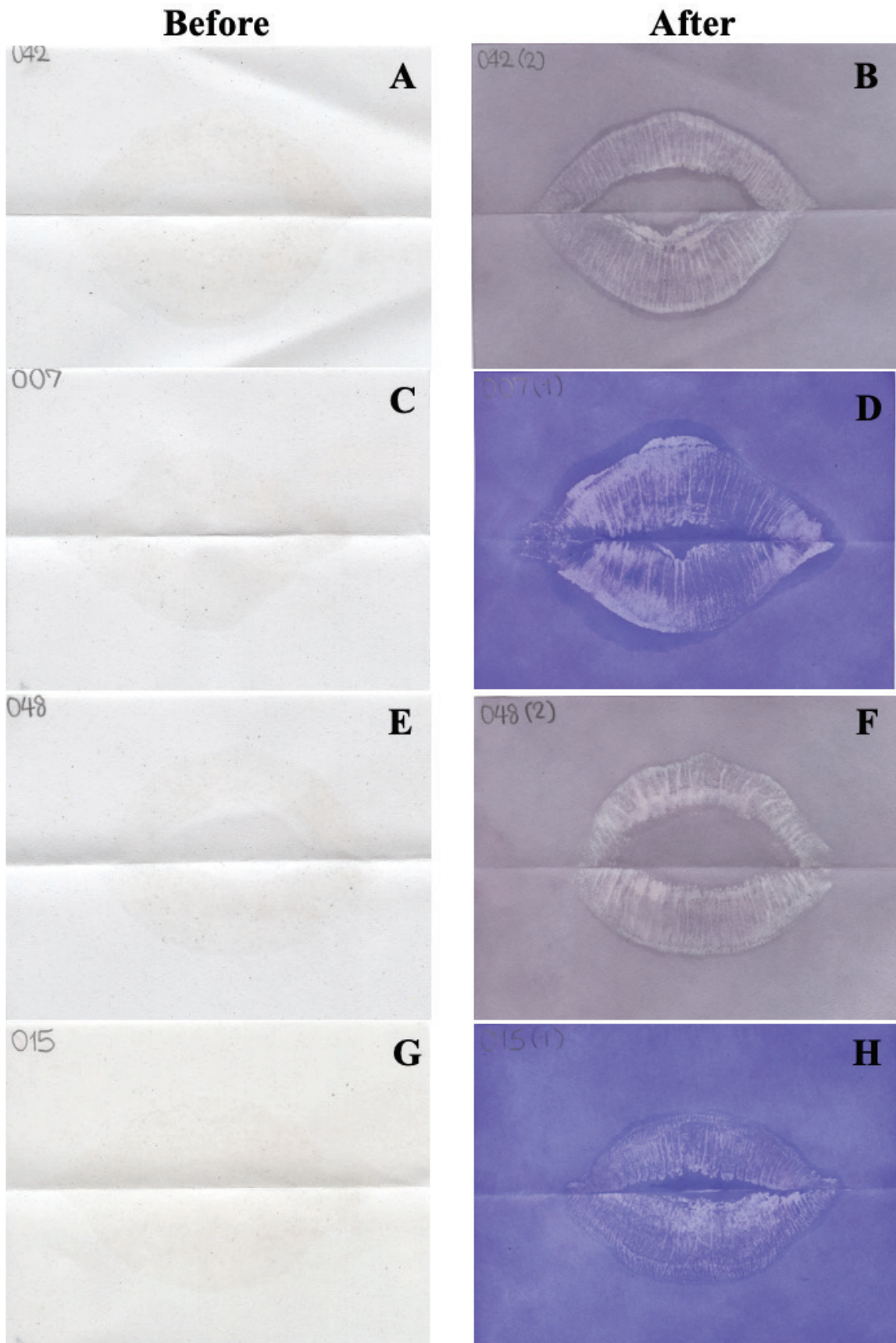


Fig 2. Four examples from 48 lip prints; latent lip prints detected on cigarette paper by the roselle extract staining method (B and F) and crystal violet staining method (D and H).

Table 1 Lip prints pattern distribution (n)

Types	n	%
I: complete vertical groove	157	54.55
I': incomplete vertical groove	61	21.12
II: branched vertical groove	42	14.58
III: intersected groove	1	0.35
IV: reticular groove	26	9.03
V: undetermined vertical groove	1	0.35

In the lip prints from the male volunteers, the order of the most common patterns was Types I > II > IV > I'. Types II and V were not found in the male population. In the lip prints from the female volunteers, the order of appearance of the patterns was Types I > I' > II > IV > III and V. The significant difference in Lip print I', II, and IV was observed between both sexes ($\chi^2=19.31, p<0.001, \chi^2=19.08, p<0.001, \chi^2=10.68, p<0.001$, respectively) (**Table 2 and Fig.3**)

Table 2 Lip prints pattern distribution regarding gender (%)

Types	Male	Female	χ^2	p-value
I: complete vertical groove	50.00	59.03	0.74	0.380
I': incomplete vertical groove	7.64	34.72	17.31	<0.001*
II: branched vertical groove	26.39	2.79	19.08	<0.001*
III: intersected groove	0.00	0.69	0.69	0.406
IV: reticular groove	15.97	2.08	10.68	<0.001*
V: undetermined vertical groove	0.00	0.69	0.69	0.406

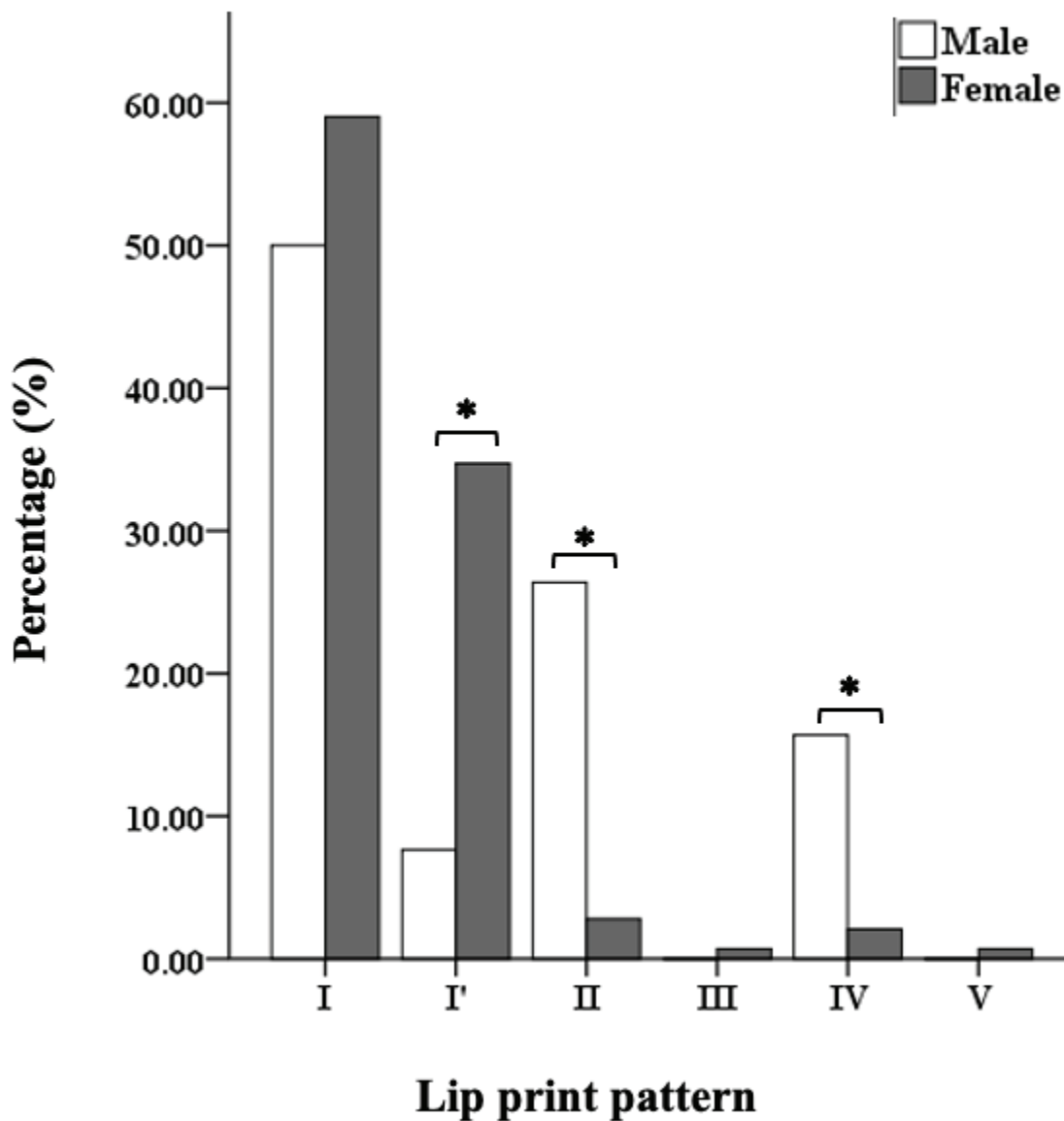


Fig 3. Distribution of the lip print pattern using Suzuki regarding gender after the roselle extract staining.

The frequencies of the lip print patterns were compared among this study and other ethnic groups reported earlier (Table 3). Type I was the most common pattern in Malaysian (Melaka, Malaysia)¹³, Indian (Maharashtra state, India)¹⁹, Libya²³ and this study with 29.84%, 46.50%, 56.72% and 54.51% respectively. On

the contrary, Type II was the most frequent pattern in the Portuguese population¹⁵ with 35.50%, the Type III lip print was found in the Brazilian², Portuguese¹⁵, Egyptian (Alexandria University, Egypt)²⁴ and Malaysian (Alexandria University, Egypt)²⁴ with 44.00%, 34.00%, 37.50% and 35.45%, respectively. Type V was the most common pattern in Iranian²⁵ and Goan population²⁶ with 35.35% and 53.42%, respectively. Overall, the lip print pattern frequency of the Thai group differed significantly from the other ethnic groups ($p = 0.000$).

Table 3 Lip prints pattern frequencies among population.

Population	Lip print patterns						χ^2	p-value
	Type I	Type I'	Type II	Type III	Type IV	Type V		
Thai (This study)	54.51	21.18	14.58	0.35	9.03	0.35		
Brazilian ²	30.00	12.00	4.00	44.00	1.00	0.00	65.13	<0.001
Malaysian ¹³	29.84	13.44	23.12	22.45	9.64	1.61	33.13	<0.001
Portuguese ¹⁵	22.50	2.50	35.50	34.00	3.00	2.50	74.38	<0.001
Indian ¹⁹	30.63	1.88	16.50	25.38	8.63	17	63.30	<0.001
Libya ²³	56.72	7.07	18.49	9.40	8.17	0.00	16.34	0.005
Egyptian ²⁴	4.60	10.80	24.50	37.50	21.25	1.65	90.26	<0.001
Malaysian ²⁴	5.00	10.80	29.55	35.45	15.00	4.15	88.74	<0.001
Iranian ²⁵	9.24	24.17	16.69	3.30	10.74	35.35	69.33	<0.001
Goun ²⁶	16.33	17.28	10.42	0.59	2.05	53.42	78.5	<0.001

Discussion

The objective of the present study was to use the Roselle dye extract for developing latent lip prints and to use the developed lip prints for study of morphological analysis. In this study, soaking of the latent lip print was used as the staining method instead of the brushing technique. However, collecting lip prints is a highly sensitive technique that depends on the lip's impression or pressure during collection¹⁷. As recommended by previous studies^{14-15,24,26}, the lips should be cleaned with a napkin before collecting the prints, and lipstick should be applied uniformly.

Generally, a manual method was used for lip print pattern analysis. Hence, it is necessary to apply digital methods for managing and analyzing lip print pattern. In this study, Autodesk SketchBook was used for lip print pattern analysis. Result revealed that a digital method provide ease in storing and identification of lip print images. Likewise, the study conducted by Rachana V. Prabhu *et al.* Adobe Photoshop 7.0 was introduced for the lip print analysis in Goan dental students.

In this study, latent lip prints were detected from 48 volunteers using the Roselle staining method. The results from the Roselle dye were similar to those with the crystal violet, indicating their reliability. Thereafter, the frequencies of the lip print pattern were performed using Autodesk SketchBook. The 48 lip groove patterns from the Thai volunteers revealed statistically significant results and among them no two lip prints were identical. The unique arrangement of the lip groove patterns confirmed to the findings by Suzuki *et al.*¹⁶. The Type I lip print pattern was found to be the most common (54.55%) followed by Types I', II, and IV. These results are similar to the findings for Type I in Libya populations (56.72%). In the present study, the frequency of Type I' was 21.18%. This is in accordance with the study conducted by Nazanin *et al.* (Iranian populations, Type I' = 24.17%)²⁵.

The frequency of Type II was 14.58% in the current study. Similar results were found in Indian¹⁹, Libyan²³ and Iranian populations²⁵. In the Brazilian population², Type III pattern was most common, and was present in 44.00% of volunteers. In contrary, only 0.35% of Type

III was observed in this study which coincide with the findings in Goun populations²⁶.

For Type IV lip pattern, results of the present study agree with Malaysian¹³, Indian¹⁹, Libyan²³ and Iranian²⁵. While Type V was the least observed in current study. This is in contrast to Iranian populations²⁵ and Goun populations²⁶. They reported that Type V was the most frequent lip groove patterns (35.35 % and 53.42%, respectively).

Nevertheless, the arrangement and the frequencies of the lip groove patterns are differ among populations²⁴. This variation in the lip print pattern could be explained that geographical origin and ethnicity of volunteers^{24,26}.

Conclusion

The current study demonstrated the efficacy of roselle dye as a staining agent to develop latent lip prints. The stained lip print could be used for study of lip print pattern. Moreover, a digital method using Autodesk SketchBook serves as an alternative method that provides ease in storing and identification of lip print pattern. However, studies with larger sample sizes should be performed to confirm these results.

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Conflict of Interest: The authors declare that there was no conflict of interest.

Ethical Clearance: Signed informed consent was obtained from each volunteer, and the study was approved by the third ethics committee of the Thammasat University Rangsit Campus (No. 194/2561).

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