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What makes your "eyes" look different?

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ABSTRACT

Facial morphology is the most distinguishable appearance and represents a promising subfield of Forensic DNA phenotyping. Located in the center of the face, eyes are important for facial recognition. In this study, epicanthal fold and palpebral fissure were selected for a preliminary genotype-phenotype analysis. Two SNPs were genotyped with SNaPshot method in 156 Chinese Han adults. A significant increased incidence of high ratio of palpebral fissure width to height was observed in the populations with AG genotype for SNP rs2074612 on gene HBEGF: AG versus GG, OR = 2.36, 95%CI = 1.08–5.18, $p < 0.05$; AG versus GG/AA, OR = 2.07, 95%CI = 1.03–4.16, $p < 0.05$. No significant genotype difference was detected between epicanthal fold positive/negative populations for both SNPs ($p > 0.05$). Further study including more samples should be conducted to discover SNPs associated with epicanthal fold and palpebral fissure traits.

1. Introduction

Genetic markers have been widely used in forensic fields for individual identification and kinship test. When the DNA extracted from a blood stain of crime scene could not match with known suspect or anyone in the DNA database, Forensic DNA phenotyping (FDP) may be an alternative [1]. FDP aims to predict the appearance traits of unknown sample donors based on DNA from the biological material, providing investigative leads for police in the absence of a suspect. Facial morphology is one of the most significant appearance traits, therefore it represents a promising subfield of FDP. In our previous studies, several SNPs associated with eyelid trait including single/double eyelids were suggested [2,3]. Here, we focused on the phenotypes of epicanthal fold and palpebral fissure, and attempted to identify associated genetic markers through genotype-phenotype analysis.

2. Material and methods

A total of 156 participants were recruited in Sichuan Province of China according to the similar criteria in previous report [3], with informed consent and permission of medical ethical commission. Then peripheral blood samples, photos and information were collected. Genomic DNA was extracted using the phenol–chloroform extraction method.

Based on epicanthal fold, all individuals were classified into two groups: epicanthal fold positive (n = 79) and epicanthal fold negative (n = 77) populations [4]. The height and width of left palpebral fissures were measured from the photos using DICOM Medical Image Viewer software (Ver1.01), followed by calculating the ratio of palpebral fissure width to height (RPF) for each individual. All individuals were divided into two categories based on the RPF, i.e. low RPF (1.8–2.4, n = 92) and high RPF (3.6–5.4, n = 64) populations.

Two SNPs, rs207104714on gene BMP4 and rs2074612 on gene HBEGF were screened and genotyped with SNaPshot® Multiplex Kit (Thermo Fisher Scientific, Warrington, UK) according to the manufactural instruction. Data analysis was performed through GeneMapper ID Software v3.2 (Applied Biosystems). The association analysis was performed using online tool SNPStats (https://www.snpstats.net/start.htm). Probability values of 0.05 or less were regarded as statistically significant between two variables.

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Table 1
Genotype frequencies of two SNPs and the correlation to low/high RPF phenotype (adjusted by sex and age).

Genetic model	Genotype	Low RPF N = 92(%)	High RPF N = 64(%)	OR(95%CI)	P-value
rs2074612					
Codominant	GG	38 (41.3%)	16 (25%)	1.00	**0.027**
	AG	41 (44.6%)	39 (60.9%)	**2.36 (1.08-5.18)**	
	AA	13 (14.1%)	9 (14.1%)	1.52 (0.51-4.51)	
Dominant	GG	38 (41.3%)	16 (25%)	1.00	**0.043**
	AG/AA	54 (58.7%)	48 (75%)	**2.14 (1.01-4.53)**	
Recessive	GG/AG	79 (85.9%)	55 (85.9%)	1.00	0.820
	AA	13 (14.1%)	9 (14.1%)	0.90 (0.34-2.36)	
Overdominant	GG/AA	51 (55.4%)	25 (39.1%)	1.00	**0.039**
	AG	41 (44.6%)	39 (60.9%)	**2.07 (1.03-4.16)**	
rs2071047					
Codominant	CC	40(43.5%)	19(29.7%)	1.00	0.140
	CT	39 (42.4%)	38 (59.4%)	2.12 (0.99-4.53)	
	TT	13 (14.1%)	7 (10.9%)	1.35 (0.43-4.18)	
Dominant	CC	40 (43.5%)	19 (29.7%)	1.00	0.073
	CT/TT	52 (56.5%)	45 (70.3%)	1.93 (0.93-4.02)	
Recessive	CC/CT	79 (85.9%)	57 (89.1%)	1.00	0.780
	TT	13 (14.1%)	7 (10.9%)	0.86 (0.31-2.42)	
Overdominant	CC/TT	53 (57.6%)	26 (40.6%)	1.00	0.057
	CT	39 (42.4%)	38 (59.4%)	1.95 (0.98-3.91)	

N corresponds to the number of individuals.
Boldfaced values indicate a significant difference at the 0.05 level.

3. Results

Two SNPs were successfully genotyped for all samples. Genotype distributions of the two SNPs were consistent with Hardy-Weinberg equilibrium in all groups ($p > 0.05$, data not shown).

As shown in Table 1, significantly increased incidence of high RPF was associated with the AG genotype of rs2074612 compared with GG or GG/AA genotypes ($p < 0.05$). Significantly increased incidence of high RPF was also observed in the rs2074612 AG/AA genotypes compared with GG genotype ($p < 0.05$). No genotype distribution difference was found for rs2071047 between low/high RPF populations ($p > 0.05$). No significant genotype difference was detected for both two SNPs between epicanthal fold positive/negative populations ($p > 0.05$, data not shown).

4. Discussion

In previous studies, facial morphology was suggested of good heredity and ethnic specificity as a complex trait influenced by multiple genetic and environmental factors [5]. Located in the center of the face, eyes and their adjacent areas are presumed to be important for facial recognition. The epicanthal fold is an oblique or vertical lunate fold of skin lying in front of the inner canthus, covering the inner canthus. Also known as Mongoloid fold, it is frequently seen in East Asians and rarely in Caucasians. Palpebral fissure is the gap between upper and lower eyelid edges. A higher RPF indicates a longer and narrower eye, while a lower RPF indicates the opposite. In this study, we tried to seek the probable genetic associations for epicanthal fold and palpebral fissure.

Two SNPs were screened out through massive paralleling sequencing data analysis. The present result indicates that AG genotype of SNP rs2074612 on gene HBEGF was associated with high RPF. No correlation between rs2074612 and common trait or morbid morphology has been reported. Gene HBEGF (heparin binding EGF like growth factor), located on Chr.5 q23, was proved to play a role of the edge extension in epithelial sheet migration during eyelid closure in the development of mouse embryos [6]. Thus, HBEGF could be a genetic factor for the appearance of palpebral fissure.

5. Conclusion

In this preliminary study, we investigated two SNPs for epicanthal fold and palpebral fissure traits in 156 Chinese Han individuals. The rs2074612 on HBEGF was observed to be associated with RPF. Further study including more samples should be conducted to discover SNPs associated with epicanthal fold and palpebral fissure traits.

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Declaration of Competing Interest

The authors declare no conflict of interest.

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References

[1] C.E. MacLean, A. Lamparello, Forensic DNA phenotyping in criminal investigations and criminal courts: assessing and mitigating the dilemmas inherent in the science, Recent Adv. DNA Gene Seq. 8 (2) (2014) 104–112.
[2] Q. Wang, B. Jin, X.Y. Luo, et al., Association between BMP4 gene polymorphisms and eyelid traits in Chinese Han population, Forensic Sci. Int. Genet. Suppl. Ser. 6 (2017) E355–E356.
[3] B. Jin, J. Zhu, H. Wang, et al., A primary investigation on SNPs associated with eyelid traits of Chinese Han Adults, Forensic Sci. Int. Genet. Suppl. Ser. 5 (2015) e669–e670.
[4] J.W. Park, K. Hwang, Anatomy and histology of an epicanthal fold, J. Craniofac. Surg. 27 (4) (2016) 1101–1103.
[5] Y. Li, W. Zhao, D. Li, et al., EDAR, LYPLAL1, PRDM16, PAX3, DKK1, TNFSF12, CACNA2D3, and SUPT3H gene variants influence facial morphology in a Eurasian population, Hum. Genet. 138 (6) (2019) 681–689.
[6] N. Mine, R. Iwamoto, E. Mekada, HB-EGF promotes epithelial cell migration in eyelid development, Development 132 (19) (2005) 4317–4326.