



Research Article

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Exploring the variation association between four head traits

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Abstract

This research work presents a comprehensive overview of four traits related to the head, with the aim of assessing the statistical phenotypic association among them. The traits examined in this study encompass earlobe type, tongue rugosity, cleft chin and tongue rolling. The primary objective was to investigate the potential associations between these traits and understand their interrelationships. The study focused on examining specific traits in a diverse group of 7431 unrelated individuals, where the genders were almost evenly distributed. To facilitate a comprehensive analysis, three distinct groups were created for each characteristic, comprising the total population, as well as male and female subsets. The selection of subjects was carefully done to ensure a fair representation across different geographical regions within Bosnia and Herzegovina, thereby accurately reflecting the nation's national and ethnic diversity. The association among these traits was assessed for statistical significance using the Chi-squared test, with Fisher's exact test used as a supplementary method to examine the connection between each pair of observed traits. Additionally, the Chi-squared test was applied to examine gender-based differences in the frequencies of the phenotypic characteristics of the head. Following traits were shown to have a statistically significant association: tongue rugosity - tongue rolling, tongue rugosity – earlobe type, cleft chin – earlobe type, cleft chin – tongue rolling and earlobe type – tongue rolling. Investigation into the variations in the frequencies of observed phenotypic traits of the head, with respect to gender, revealed statistically significant results for every trait examined.

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Keywords

Earlobe type, Tongue rugosity, Cleft chin, Tongue rolling, Bosnia and Herzegovina

Introduction

Within the field of bioanthropological research, the primary emphasis lies on investigating traits that can be readily measured, leading to their classification into distinct categories: dynamic-morphological, static-morphological, and biochemical-physical traits. These traits typically exhibit an oligogenic or polygenic nature, rendering the assessment of their associations a challenging endeavor. Consequently, one viable approach to comprehend the complexity of such traits involves analyzing phenotypic associations, wherein the evaluation centers around the interconnection of different phenotype variants.

The history of bioanthropological research in Bosnia and Herzegovina traces its origins to the late 19th century, coinciding with the era of the Austro-Hungarian Empire. A comprehensive account of this historical research is available in the publication by Lasic et al. (2016). Subsequently, a series of bioanthropological and genetic studies have been conducted in the region, with the aim of assessing the heterogeneity within local populations and the broader Bosnian and Herzegovinian population as a whole. Notable works include those authored by Ahmic et al. (2018), Berberovic (1969), Dogan et al. (2016), Dzehverovic et al. (2021), Hadziselimovic (1981), Halilovic et al. (2022), Kalajdzic et al. (2021), Kapur-Pojkskic et al. (2014), Kovacevic et al. (2013, 2014), Lasic et al. (2013, 2016, 2020), Marjanovic et al. (2005, 2005a, 2006, 2008), and Pojskic et al. (2013). It is noteworthy, however, that the majority of these investigations rely on molecular genetic markers, with a smaller proportion utilizing phenotypic markers. In 2003, Pojskic conducted an assessment of heterogeneity over temporal periods. Moreover, numerous

studies conducted by Bosnian and Herzegovinian scientists during the last century have focused on evaluating heterogeneity based on the head phenotype and other phenotypic traits (Berberovic 1969, 1978; Berberovic et al. 1976; Hadziselimovic 1977, 1981; Hadziselimovic and Terzic 1985; Kaunic and Grin 1963). However, it is essential to note that none of these studies have explored the associations among different phenotypic traits. Therefore, the purpose of this paper is to provide an overview of four potential traits associated with the head. Utilizing a substantial dataset, we aim to estimate the statistical associations between the phenotypes related to the head.

Through a comprehensive review of relevant literature on the four traits regarding the head (Doraczynska-Kowalik et al., 2017; Eidelman et al., 1976; Jaruga et al., 2022; Redman et al., 1972; Shaffer et al., 2017; Wiener, 1937), the mode of inheritance for these traits was established. These studies have highlighted the presence of variations in the mentioned traits, indicating underlying genetic variability. Notably, the majority of these characteristics are controlled by the influence of multiple genes, categorizing them as polygenic traits. Drawing from the insights gathered above, it becomes evident that investigating the interrelationships among phenotypic traits related to the head holds considerable promise and intrigue within the domain of bioanthropology.

Material and methods

Potential phenotypic associations among four head-related traits were observed, namely earlobe type (L), tongue rugosity (F), cleft chin (Ch) and tongue rolling (R) (Table 1). The research was conducted on a comprehensive sample of 7431

unrelated individuals, selected randomly to ensure a diverse representation across various geographical regions in Bosnia and Herzegovina, effectively capturing the characteristic national and ethnic composition of the population (Figure 1). The gender distribution within the total sample was approximately equal, and out of the 7431 subjects, data on gender distribution were available for 6985 individuals.



Figure 1. Geographical dispersion of the studied local populations in Bosnia and Herzegovina

Hence, for the examination of phenotypic variant frequencies and their associations, only samples with known gender information were included in the analysis. To assess the statistical significance of the relationship among these features, the Chi-squared test was employed. Additionally, Fisher's exact test was used as an additional measure to assess the relationship between each pair of monitored traits. The statistical calculations were performed using MedCalc version 18.10.2, and a significance level of 0.05 was considered. To investigate potential disparities in the frequencies of phenotypic characteristics of the head based on gender, the Chi-squared test was employed with a significance level set at $p < 0.05$. The dataset was divided into three distinct groups for each case: total, male, and female groups.

Results and Discussion

Table 1 provides a comprehensive list of four observed head-related traits along with their respective abbreviations. The outcomes of both Fisher's exact and Chi-squared tests conducted for all three groups are presented in Tables 2, 3, and 4.

Table 1. List of four observed head features

Trait	Abbreviation	Recessive	Variant I	Variant II	Author
Earlobe type	L	Attached	Attached - 0	Free - 1	Hilden (1922)
Tongue rugosity (Fissured tongue) (Geographic tongue)	F	„Normal“ tongue	“Normal” tongue - 0	Geographic tongue - 1	Turpin and Caratzali (1936)
Cleft chin (Dimple in chin)	Ch	Smooth chin	Dimple in chin - 0	Smooth chin - 1	Günther (1939), Meirowsky (1924), Gillin (1959)
Tongue rolling (Tongue curling)	R	Non-roller	Non-roller - 0	Roller - 1	Sturtevant (1940)

Table 2. Fisher's exact test p-value and Chi-squared value for the whole sample regarding the four monitored head features (top section of the table - Chi-squared value; bottom section of the table - Fisher exact p value; * p<0.05; ** p<0.01; *** p<0.001)

	L	F	Ch	R
L		40.237***	11.898***	30.382***
F	0.000000001***		0.997	37.520***
Ch	0.000587899***	0.319294912		3.541
R	0.000000034***	0.000000001***	0.060277203	

Table 3. Fisher's exact test p-value and Chi-squared value for the male samples regarding the four monitored head features (top section of the table - Chi-squared value; bottom section of the table - Fisher exact p value; * p<0.05; ** p<0.01; *** p<0.001)

	L	F	Ch	R
L		24.583***	29.378***	24.045***
F	0.000000905***		0.036	14.437***
Ch	0.000000077***	0.865775718		10.124**
R	0.000000923***	0.000161335***	0.001629648**	

Table 4. Fisher's exact test p-value and Chi-squared value for the female samples regarding the four monitored head features (top section of the table - Chi-squared value; bottom section of the table - Fisher exact p value; * p<0.05; ** p<0.01; *** p<0.001)

	L	F	Ch	R
L		21.858***	2.332	6.419*
F	0.000004462***		0.194	28.985***
Ch	0.132637630	0.673058588		0.114
R	0.011493533*	0.000000074***	0.756790642	

The frequency difference analysis of monitored head phenotypic features by gender revealed statistical significance for L ($\chi^2=39.937$, $p<0.0001$), F ($\chi^2=30.629$, $p<0.0001$), Ch ($\chi^2=73.444$, $p<0.0001$) and R ($\chi^2=11.570$, $p=0.0007$). Our study found a statistically significant difference in the frequency of the phenotype variants L, F, Ch, and R between the genders. In trait R, the male cohort shows a balance between non-roller and roller phenotypes, while the female cohort exhibits a more pronounced presence of roller phenotypes compared to non-rollers. In the case of F, for both genders

“normal” tongue phenotype variant is more pronounced in relation to geographic tongue, with a slightly greater difference noticed in different variants of this trait in female cohort than male cohort, while in the case of L, for both genders attached earlobe variant is more pronounced in relation to free earlobe, with a larger difference observed in male group than female group. In trait Ch, for both genders dimple in chin variant is more pronounced in relation to smooth chin, with a larger difference observed in the female group than the male group.

Statistically significant association was observed between following traits: tongue rugosity - tongue rolling, tongue rugosity – earlobe type, cleft chin –

earlobe type, cleft chin – tongue rolling and earlobe type – tongue rolling (Figure 2A, B, C, D).

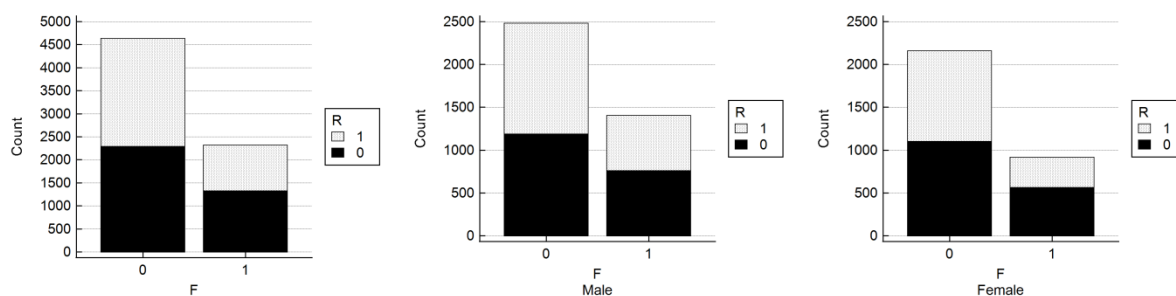


Figure 2A. Comparative visual depiction of the combined frequency of related features showing statistically significant associations

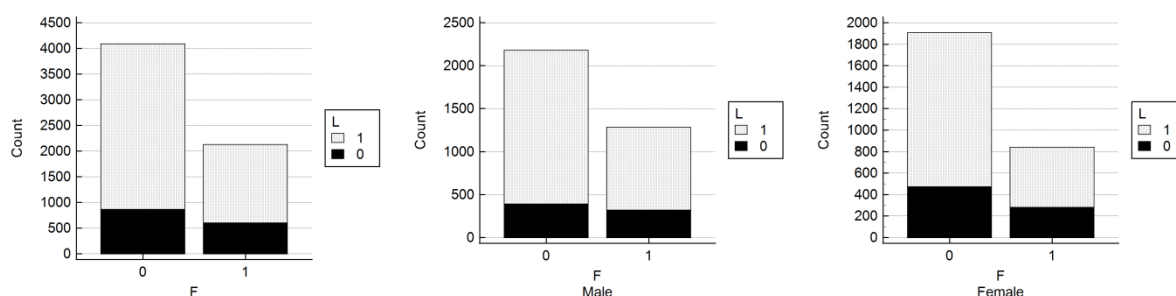


Figure 2B. Comparative visual depiction of the combined frequency of related features showing statistically significant associations

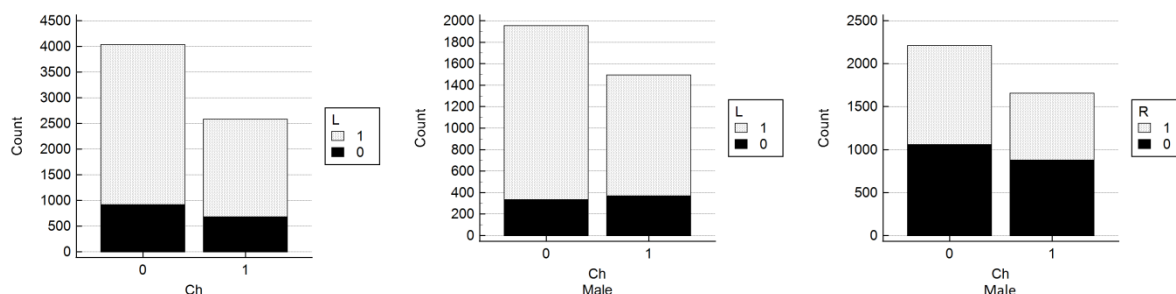


Figure 2C. Comparative visual depiction of the combined frequency of related features showing statistically significant associations

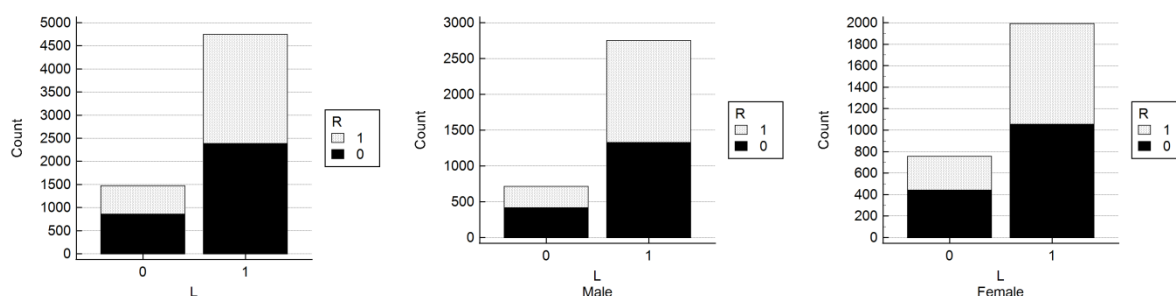


Figure 2D. Comparative visual depiction of the combined frequency of related features showing statistically significant associations

Tongue rugosity and tongue rolling

Regarding association between F and R, highest significance was observed in total sample ($\chi^2 = 37.520$; $p < 0.0001$, Fisher's exact test $p < 0.0001$). Statistical significance was also observed in both gender groups, female ($\chi^2 = 28.985$, $p < 0.0001$, Fisher's exact test $p < 0.0001$) and male ($\chi^2 = 14.437$, $p = 0.0001$, Fisher's exact test $p < 0.0002$). In the overall sample, the highest proportion of individuals (2349) had a "normal" tongue and the ability of tongue rolling, while the smallest proportion (995) had a geographic tongue and the ability of tongue rolling. In female cohort, the highest proportion of individuals (1105) had a "normal" tongue but didn't have the ability of tongue rolling, while the smallest proportion (352) had a geographic tongue and the ability of tongue rolling. In male cohort, the highest proportion of individuals (1293) had a "normal" tongue and the ability of tongue rolling, and smallest proportion (643) had a geographic tongue and the ability of tongue rolling.

Cleft chin and tongue rolling

Regarding the association among Ch and R, statistical significance was found explicitly in the male cohort ($\chi^2 = 10.124$, $p = 0.0015$; Fisher's exact test $p = 0.0016$). In contrast, no statistical significance was found in the female cohort ($\chi^2 = 0.114$, $p = 0.7362$; Fisher's exact test $p < 0.7568$). Among males, the largest proportion of individuals (1151) exhibited both a dimple in the chin and the ability to roll their tongues, while the smallest proportion (778) had a smooth chin along with the ability to roll their tongues.

Tongue rugosity and earlobe type

In case of traits F and L, statistical significance was observed in all 3 groups, total, female and

male. In the total group values are as follows ($\chi^2 = 40.237$, $p < 0.0001$, Fisher's exact test $p < 0.0001$), female group ($\chi^2 = 21.858$, $p < 0.0001$, Fisher's exact test $p < 0.0001$) and male group ($\chi^2 = 24.583$, $p < 0.0001$, Fisher's exact test $p < 0.0001$). In total group, the highest proportion of individuals (3222) had a "normal" tongue and a free earlobe, and smallest proportion (606) had a geographic tongue and an attached earlobe. In female group, the highest proportion of individuals (1433) had a "normal" tongue and a free earlobe, and smallest proportion (283) had a geographic tongue and an attached earlobe. Also in male group, the largest proportion of individuals (1788) had a "normal" tongue and a free earlobe, and smallest proportion (323) had a geographic tongue and an attached earlobe.

Earlobe type and tongue rolling

In case of features L and R, statistical significance was observed in all 3 groups, total, female and male. In the total group values are ($\chi^2 = 30.382$, $p < 0.0001$, Fisher's exact test $p < 0.0001$), female group ($\chi^2 = 6.419$, $p = 0.0113$, Fisher's exact test $p < 0.0115$) and male group ($\chi^2 = 24.045$, $p < 0.0001$, Fisher's exact test $p < 0.0001$). In total cohort, the highest proportion of individuals (2387) had a free earlobe and didn't have the ability of tongue rolling, and smallest proportion (613) had an attached earlobe and ability of tongue rolling. Also, in female cohort, the highest proportion of individuals (1058) had a free earlobe and didn't have the ability of tongue rolling, and smallest proportion (315) had an attached earlobe and ability of tongue rolling. In male cohort, the highest proportion of individuals (1426) had a free earlobe and ability of tongue rolling, and smallest proportion (298) had an attached earlobe and ability of tongue rolling.

Cleft chin and earlobe type

Concerning the relationship between Ch and L, the highest significance was found in the male cohort ($\chi^2 = 29.378$; $p < 0.0001$; Fisher's exact test $p < 0.0001$). Statistical significance was also noted in the overall sample ($\chi^2 = 11.898$, $p = 0.0006$; Fisher's exact test $p < 0.0006$). However, no statistical significance was detected in the female cohort ($\chi^2 = 2.332$, $p = 0.1267$; Fisher's exact test $p = 0.1326$). In the overall sample, the highest proportion of individuals (3121) exhibited a dimpled chin and free earlobes, while the smallest proportion (686) had a smooth chin and attached earlobes. Within the male cohort, the largest proportion of individuals (1620) had a dimpled chin and free earlobes, whereas the smallest proportion (337) had a dimpled chin and attached earlobes.

Cleft chin and tongue rugosity

Statistically significant association was not registered between cleft chin and tongue rugosity. With respect to Ch and F, the results indicate a clear lack of statistical significance across all three cohorts: total ($\chi^2 = 0.997$, $p = 0.3179$, Fisher's exact test $p < 0.3193$); female ($\chi^2 = 0.194$, $p = 0.6600$, Fisher's exact test $p < 0.6731$); male ($\chi^2 = 0.036$, $p = 0.8494$, Fisher's exact test $p < 0.8658$).

Genetic background of the observed traits

The inheritance of cleft chin, also known as a chin dimple (OMIM: 119000), follows an autosomal dominant pattern, though the exact number of genes involved remains unclear. Numerous studies have explored this trait. Guenther (1939) documented 9 cases across 5 generations, while Meirowsky (1924) identified 25 cases in 4

generations. Gorlin (1982) observed it over 4 generations, and McKusick (1988) reported it in 3 generations. According to McKusick (1992), it appears that females are less frequently affected than males. Some evidence suggests that the SNP rs11684042 on chromosome 2 may be linked to cleft chin (AncestryDNA Traits Learning Hub).

Free earlobes (OMIM: 128900) are typically regarded as dominant in contrast to attached earlobes. While there are various theories about the number of genes involved in their inheritance, Dutta and Ganguly (1965) proposed a polygenic model. The term "lobelles" is often preferred over "attached" to describe this trait. El Kollali (2009) suggested categorizing earlobes into three types based on the angle of attachment: acute, right, or obtuse. Lai and Walsh (1966) concluded that earlobe types do not adhere to a simple Mendelian inheritance model, as is commonly assumed. Mohanraju and Mukherjee (1973) observed that in the Indian population, geographical, ethnic, and social factors might influence the frequencies of earlobe types. They recommended using the overall frequencies of attached earlobes for population studies and suggested that quantitative variation should be considered for further genetic analysis. Their study also revealed a stronger association between parents and offspring, but no evidence to support the notion of a single locus with two alleles. Early genetic studies on earlobes by Carrière (1922) and Hilden (1922) produced conflicting results. Carrière (1922), examining 15 families, concluded that attached earlobes were dominant, while Powell and Whitney (1937), studying a single family, found that attached earlobes were recessive.

Turpin and Caratzali (1936) concluded that a single gene is responsible for both geographic tongue and fissured tongue (OMIM:137400).

Dawson and Pielou (1967), in their study of 18 individuals with geographic tongue—some of whom also exhibited fissured tongue across three generations—determined that the inheritance pattern is autosomal dominant. This finding aligns with Seiler's (1936) study, which, based on extensive pedigree data, also supports an autosomal dominant inheritance pattern. Kullaa-Mikkonen (1988), in his genetic study of fissured tongue involving 31 families, found that fissured tongue with smooth-surfaced papillae is inherited as a dominant trait with incomplete penetrance, and often follows the occurrence of geographic tongue.

The inheritance of tongue curling, folding, or rolling (OMIM: 189300) is generally regarded as an autosomal dominant trait following simple Mendelian principles, although some debate exists (Mader, 2000). Sturtevant (1940) categorized individuals into "rollers" and "non-rollers," classifying the roller phenotype as dominant. Hsu (1948) proposed that the ability to fold the tip of the tongue is recessive, a view supported by Liu and Hsu (1949) and Lee (1955), who found these traits to be independent. Matlock (1952), in a study involving monozygotic twins, suggested that there is minimal genetic basis for this trait, as discordance was observed among identical twins. Martin (1975) reached a similar conclusion, noting that concordance rates were comparable between monozygotic and dizygotic twins. Hernandez (1980) found that 63.7% of males and 66.84% of females could roll their tongues, and also identified an association between tongue rolling and the ability to move the ears (OMIM:129100) in males.

If these four traits are considered as markers for a single locus with two alleles, they could be utilized in preliminary screening analyses to investigate the

genetic structure of populations and their interrelationships (Hadžiselimovic, 1981).

The scientific literature contains limited papers that provide estimates of the association of morphological phenotypic traits, particularly concerning the head. Consequently, this study holds additional value as it explores the potential associations of phenotypic traits in a sizable sample of unrelated individuals. The decision to focus on four head-related traits stemmed from the presumption that they might exhibit associations due to their shared location on the body. The outcomes of this investigation reveal the presence of associations among specific phenotypic traits of the head, leading to the categorization of three distinct result types. Firstly, statistically significant associations were observed in both the total population and the gender-specific subgroups. Secondly, there were statistically significant associations observed solely within the total population. Lastly, the third type of result exhibited statistically significant associations exclusively within the gender-differentiated populations. These findings shed light on the interplay of phenotypic traits and provide valuable insights into the complexities of their associations within the head region.

Conclusion

The assumption regarding the phenotypic association among head traits encompasses a total of six trait combinations. Specifically, the phenotypic association of head traits within gender groups (M and F) was observed in three combinations: earlobe type - tongue rugosity, earlobe type - tongue rolling, and tongue rugosity - tongue rolling. The evident polygenic nature of the genetic background underlying these traits suggests the involvement of several genes in their

expression, given that they are polygenic traits. A comprehensive understanding of the inheritance mechanism would further enrich our knowledge of the genetic interplay among specific genes that form the foundation of inheritance.

Authors' contributions

T. Corbo:

Methodology; Writing – original draft; Data curation; Formal analysis.

N. Pojskic:

Conceptualization; Supervision; Writing – original draft; Writing – review & editing.

A. Kalajdzic:

Methodology; Data curation; Formal analysis.

R. Hadziselimovic:

Conceptualization; Resources.

Conflict of interest

Authors declare no conflict of interest.

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