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EXPLORING PHENOTYPIC DIVERSITY OF IRIS FEATURES AMONG STUDENTS AT SHAH ABDUL LATIF UNIVERSITY, KHAIRPUR MIR'S, SINDH, PAKISTAN

Majeeda Ruk[1], Naila Parveen Abbasi[2], Zaibun-Nisa Memon[3], Sumbul Mureed Mastoi[4], Tasneem Kousar[*5]

*[1,3,*5]Department of Zoology, Faculty of Natural Sciences, Shah Abdul Latif University, Khairpur Mir's, Sindh, Pakistan*
[2]Department of Geography, Faculty of Physical Sciences, Shah Abdul Latif University, Khairpur Mir's, Sindh, Pakistan
[4]Department of Entomology, Faculty of Crop Protection, Sindh Agriculture University, Tandojam, Pakistan; College of Forestry, Sichuan Agriculture University, Chengdu, China

[*5]tasneemlarik710@gmail.com

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Tasneem Kousar

Abstract

Human iris displays significant phenotypic diversity in color and structural attributes, rendering it a crucial characteristic in the fields of genetics, forensic science, anthropology, and biometric identification. Nevertheless, information is scarce regarding iris phenotypes within the Pakistani population, especially in Sindh. This study aimed to explore the variation of iris features among students at Shah Abdul Latif University, Khairpur, Sindh, Pakistan. A total of 500 healthy, unrelated students (278 males and 222 females), aged between 18 and 25 years, were recruited after obtaining informed consent. Standardized digital images of both eyes were taken and categorized into five eye-color groups according to the Fitzpatrick Phototype Scale. Statistical analyses were conducted using IBM SPSS version 22.0 to assess the distribution of iris phenotypes and their correlation with demographic factors. The results indicated that brown-based eye colors were the most prevalent in the study group. The brownish black phenotype was the most frequently observed (35.6%), followed by dark brown (26.8%) and light brown (25.4%), while hazel/green (7.4%) and blue (4.8%) eyes were relatively uncommon. Males exhibited a higher occurrence of darker eye colors, whereas lighter eye colors were more commonly found among females. In total, brown shades comprised 87.8% of all documented eye colors, reflecting the typical pigmentation pattern of the local population. This research offers foundational data on iris phenotypic diversity among young adults in Sindh and provides essential information for future studies in population genetics, forensic science, and biometric research. The results also underscore the significance of recording regional phenotypic variations to enhance the precision of identification systems and population-based research.

INTRODUCTION

Phenotypic diversity refers to the observable variations in physical characteristics among individuals, including eye color, iris patterns, and other morphological traits. Human iris features are considered one of the most unique phenotypic characteristics because the iris possesses complex patterns such as crypts, furrows, pigment spots, and rings that differ from person to person. These variations are influenced by

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both genetic and environmental factors and are increasingly important in the fields of anthropology, forensic science, biometrics, and population genetics (Bashir et al., 2024; Dario et al., 2015). The human iris develops during early embryonic stages and remains relatively stable throughout life. Iris pigmentation mainly depends on melanin concentration and distribution within the iris tissue (Moazed, 2020). Differences in melanin levels produce various eye colors and iris textures among populations. Researchers have reported that iris color and structural features vary significantly according to ethnicity, geographic location, and genetic background (Jonnalagadda et al., 2019; Rahat et al., 2023). In recent years, iris phenotyping has gained considerable attention because of its applications in biometric identification systems and forensic investigations. Iris recognition technology is considered one of the most accurate biometric methods due to the uniqueness and permanence of iris patterns. Modern forensic studies also use iris traits to assist in personal identification and population-based research (Bhatt et al., 2025). In Pakistan, research related to ocular phenotypes remains limited despite the country's rich ethnic diversity. Sindh is home to populations with diverse ancestral and genetic backgrounds, which may influence iris morphology and pigmentation patterns. Studying iris phenotypes among students at Shah Abdul Latif University can therefore provide important baseline data regarding phenotypic variation within the Sindhi population. Additionally, documenting these characteristics may support future developments in biometric identification systems and forensic investigations in Pakistan (Bashir et al., 2024; Walsh et al., 2012). Another important aspect of phenotypic research is the relationship between iris characteristics and genetic inheritance. Eye color and iris texture are polygenic traits controlled by multiple genes, particularly the OCA2 and HERC2 genes, which influence melanin production and pigmentation. Variations in these genes contribute to differences in iris color and pattern formation among populations. Recent molecular studies have highlighted that iris phenotypes can provide valuable information regarding ancestry, population structure, and biological diversity (Wang et al., 2022). University students represent individuals from different ethnic, cultural, and regional backgrounds, making them an important population for studying phenotypic diversity. Exploring iris characteristics among Shah Abdul Latif University Khairpur students may provide valuable information regarding the prevalence and distribution of iris features in the Sindh population. Furthermore, such studies can contribute to future biometric databases, forensic applications, and genetic research in Pakistan. Therefore, the present study aims to explore the phenotypic diversity of iris features among students at Shah Abdul Latif University, Sindh, Pakistan. The study will focus on identifying variations in iris color, iris texture patterns, and associated phenotypic characteristics among the student population.

MATERIALS AND METHODOLOGY

Sample collection

The study was conducted after obtaining approval from the Institutional Ethical Committee of Shah Abdul Latif University. A total of 500 healthy and unrelated volunteers, including 278 males and 222 females, participated in the research. All participants were students of the university and belonged to the age group of 18–25 years. Before participation, informed consent was obtained from everyone, and a questionnaire was completed to collect demographic information such as gender, age, ethnic background, and ancestral place of birth.

Phenotyping

Digital photographs of the eyes and the inner side of the upper arm of each participant were captured using a 24.2-megapixel Canon EOS 80D camera equipped with an 18–135 mm lens. The photographs were taken from a fixed distance of 10 cm under standardized lighting conditions. Each image was captured three times at a shutter speed of 1/100 and ISO 200 to ensure image consistency and accuracy.

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Sample Binning

Eye color phenotypes were assessed qualitatively according to the Fitzpatrick Phototype Scale (fig. 1) (Edwards et al., 2016a). For easier classification, the eye images were divided into five categories: brownish black, dark brown, light brown, hazel/green, and blue. All digital eye images were independently examined by two investigators under the same environmental conditions. In cases of disagreement, the photographs were re-evaluated carefully until a consensus regarding phenotype classification was achieved.

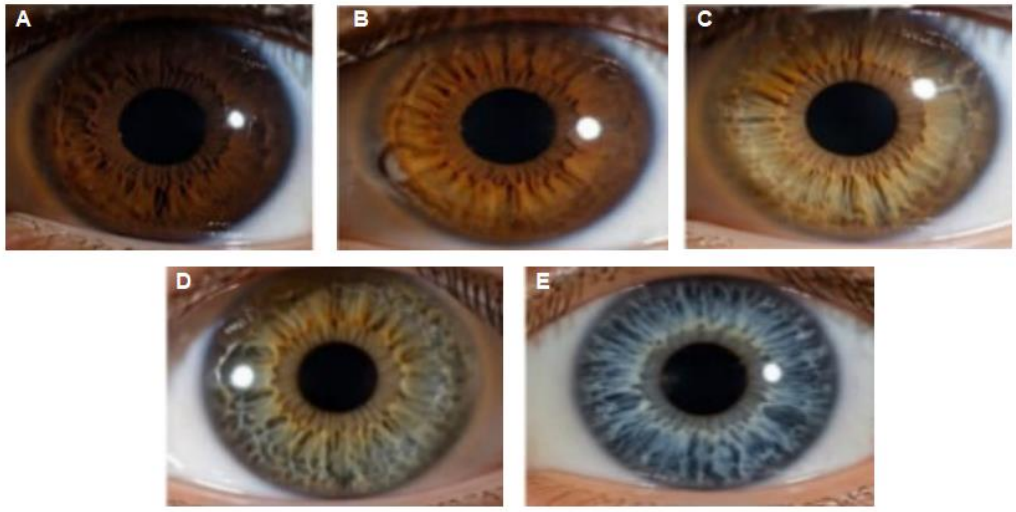


Fig. No. 1. Representative eye color images collected from students of Shah Abdul Latif University and classified according to the Fitzpatrick Phototype Scale (A–E). The scale categorized eye color into five groups: A = brownish black, B = dark brown, C = light brown, D = hazel/green, and E = blue. For easier interpretation and analysis, all collected eye images were organized into these five major categories.

Statistical Analysis

All statistical analyses were performed using IBM SPSS Statistics version 22.0. The Gamma statistic was applied to evaluate the correlation between iris characteristics and eye color phenotypes. For each correlation analysis, both the Gamma (G) value and p-value were recorded to determine the strength and significance of associations. Eye characteristics showing significant statistical relationships were considered correlated with each other or with eye color variation. Ordinal regression analysis was further conducted to investigate the association of iris patterns with factors such as gender, age, iris diameter, and eye color. In addition, goodness-of-fit and proportional odds assumptions were assessed to validate the regression model. To identify significant differences in iris features and eye color among different genders and age groups, the Chi-square test was applied. Variations between provincial populations were also examined using the same method. Furthermore, differences in iris diameter among sample groups were analyzed through one-way ANOVA. Before performing statistical analyses, data normality was assessed using Q–Q plots to ensure the suitability of the applied statistical tests

RESULTS

Data analysis

Figure 2 shows eye color distribution across 500 participants, with 278 males (55.6%) and 222 females (44.4%) with their frequencies (Table 1). Brownish black is the most frequent color overall at 35.6%, with 48.9% of males and 18.9% of females. Dark brown was nearly equal in both genders: 26.6% in male 27.0% in females, 28.8% overall. Light brown was much higher in females at 37.8% compared to only 15.5% in males,

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25.4% overall. Hazel/Green were 7.4% overall, seen in 9.0% of females and 6.1% of males. Blue eyes were the rarest at 4.8% overall, with 7.2% in females and 2.9% in males. Brown shades combined were 87.8% of all people. Males had more dark brown/black; females had more light brown (Fig. 2).

Table 1: Phenotypic Iris features Frequency (Male/Female)

Iris Feature Eye color	Male (n=278)	Females (n=222)	Total (n=500)	Male Frequency	Female Frequency	Overall Frequency
Brownish black	136	42	178	0.50	0.19	0.35
Dark brown	74	60	134	0.27	0.27	0.37
Light brown	43	84	127	0.16	0.37	0.25
Hazel/Green	17	20	37	0.06	0.09	0.074
Blue	8	16	24	0.03	0.07	0.048
Total	278	222	500	1.00	1.00	1.00

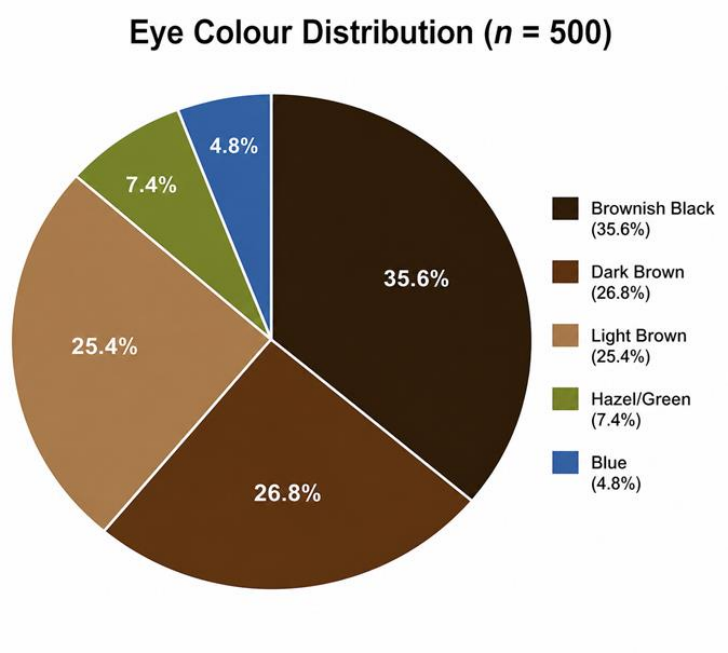


Fig. No 2. Showing percentage of phenotypic iris colour distribution in male and female students.

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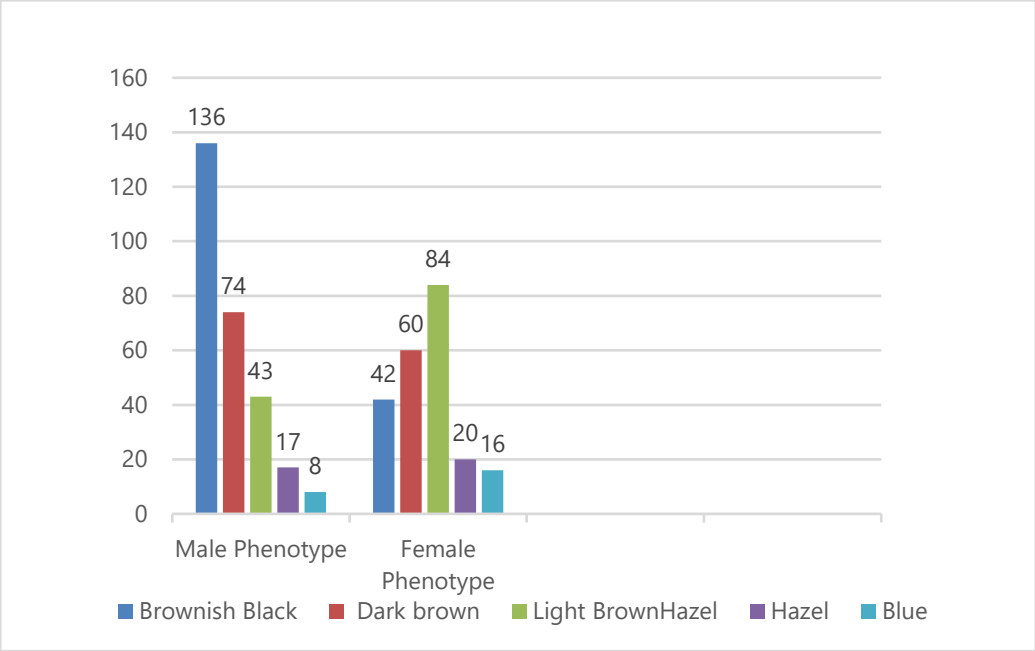


Fig. No. 3 Count of surface features in two quadrants of iris.

The distribution of eye color among the study population shows the brownish black is the most prevalent, accounting for 35.6%, forming the largest segment of the chart. This is followed by dark brown at 26.8% and light brown at 25.4% both representing substantial proportion. Less common eye colors include hazel/green, which makes up 7.4% while blue is the least frequent, comparing only 4.8% of the sample (fig no.1).

DISCUSSION

The present study showed phenotypic versions of iris features among students of Shah Abdul Latif University. The findings showed that brown-based eye colors were most common in this population. Brownish black (35.6%) was the highest, followed by dark brown (26.8%) and light brown (25.4%). Generally, brown shades made up 87.8% of all individuals, indicating a clear dominance of darker eye colors (Kalyani et al., 2023; Katsara&Nothnagel, 2019; Rahat et al., 2020; Soomro et al., 2025; White&Rabago-Smith, 2011). These observations are consistent with previous studies reporting that brown eye color is the most common globally, particularly in South Asian populations. Higher melanin levels in the iris are responsible for darker pigmentation, which is affected by genetic factors and environmental adaptation. Sunlight exposure in tropical regions may also contribute to increased melanin production (Edwards et al., 2016; Sturm&Duffy, 2012). Gender wise differences were recognized in the study. Males have darker eye colors, especially brownish-black, while females had relatively higher proportions of light brown, hazel/green, and blue eyes (Abdalla, 2021; Alghamdi et al., 2019; Jablonski&Chaplin, 2017). These differences may be related to genetic variation or sampling variation; though further studies are needed for confirmation. Less common eye colors such as hazel/green (7.4%) and blue (4.8%) were rare, showing limited phenotypic variation in the population. This suggests that lighter eye colors are uncommon in this area due to genetic background. In the end, the study showed that brown eye color is the dominant phenotype among students and provides useful baseline data for future research in genetics, biometrics, and forensic science (Hu et al., 2009; Wakamatsu et al., 2008; Walsh et al., 2011). In previous studies, environmental factors, particularly UV radiation, have also played a significant role in shaping phenotypic ratios of eye color (MacFarlane et al., 2024; Marro et al., 2022). Regions with high UVR exposure favor increased melanin levels, resulting in darker eye

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phenotypes, whereas regions with lower UVR allow for the persistence and diversification of lighter eye colors (Del Bino et al., 2018; Jablonski&Chaplin, 2017; Kayser&Schneider, 2009).

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