A Cross-Sectional Assessment of Quantitative Epidermal Melanin and Erythema Indices among North Indians

Abstract

Background: Remarkable diversity of skin tones among Indians ranging from pale pinkish to dark brown appears to be an ideal choice for the assessment of skin pigment variation. **Aim:** The present study was designed to assess the variation observed in melanin and erythema indices among north Indians. **Material and Methods:** Skin reflectance data (n = 574) was collected from six diverse populations of north India using DermaSpectrometer (DSM II ColorMeter) followed by statistical analysis to investigate the impact of geographical location and gender on constitutive skin pigmentation. **Results:** The melanin index (MI) varied between 17.93 and 56.92 (Mean (M) = 35.80 ± 6.26) whereas the erythema index (EI) varied between 4.92 and 18.82 (M = 10.48 ± 2.68). MI and EI of females were found to be significantly lower than males (P < 0.001). Geographical location exhibited a significant association with MI and EI (P < 0.001). Furthermore, we have noted a positive correlation between MI and EI (P < 0.001). Conclusion: The study has refined our understanding of skin pigmentation variation among north Indians in terms of significant association with geographical location {MI: $F_{(5.568)} = 31.07, P < 0.001$; EI: $F_{(5.568)} = 73.37, P < 0.001$ } and gender {MI: $t_{(386)} = -4.06, P < 0.001$; EI: $t_{(386)} = -11.96, P < 0.001$ } and rendered opportunities for further studies.

Keywords: Constitutive skin pigmentation, erythema index, melanin index, pigmentation variation, skin reflectance

Introduction

Skin pigmentation is mainly determined bv melanin. oxygenated/reduced hemoglobin, carotene, bilirubin, and other chromophores.^[1] Melanin, a critical substance in pigmentation, is produced by melanocytes in specialized vesicles called melanosomes located in the stratum basale of the epidermis. Melanin concentration differences are characterized by the size, shape, number, and aggregation of melanosomes along with the relative ratios of its two basic forms - eumelanin and pheomelanin, which can be associated with darker and lighter phenotypes, respectively.^[2,3] The concentration of carotenoids and hemoglobin also contributes significantly to skin pigmentation. Carotenoids give skin a yellow color, whereas red and bluish-purple colors are produced by oxygenated and reduced hemoglobin, respectively.^[4]

Variation in skin pigmentation can be mainly attributed to the function of genetics (constitutive pigmentation)

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stimuli (facultative external or pigmentation).^[4] Skin color assessment has several applications in biomedical sciences like forensic examination of bruises, genotype-phenotype association studies, evaluation of chemotherapy-induced erythema, and other medical conditions.^[5] Evaluation of skin pigmentation using various color scales is a subjective measure based on bias associated with an individual's interpretation of stimuli. The Fitzpatrick scale has been widely used for the categorical assessment of skin sensitivity and reactivity to sun exposure and therefore for skin color assessment.^[6] With the advent of advanced technologies, instruments like the tristimulus colorimeter and spectrophotometer have been developed for precise, quantifiable, rapid, and reproducible measurements of skin color. Tristimulus colorimeter measures the absorbance of red, green, and blue colors (RGB) and provides a readout in RGB or CIEL*a*b* (Commission Internationale de l'Eclairage color space, whereas a spectrophotometer can be used

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to define spectral characteristics of skin color based on reflectance and absorption of visible light (400–700 nm).^[7-9] Reflectance spectrophotometer has been recognized as the "gold standard" for skin pigmentation evaluation. It provides a readout of reflectance indicating the ratio between the intensity of the incident and reflected light from the skin and displays values of melanin index (MI) and erythema index (EI) as a function of absorbance by human skin.^[9,10] The higher the pigment concentration, the higher the light absorption, and subsequently higher the erythema and melanin indices.^[10]

The heterogeneity of skin tones in the Indian subcontinent is quite fascinating with various factors acting along with multilayered endogamy to shape the skin color variation. This study aims to assess the variation in quantitative measurements of erythema and melanin indices in north Indian population using a reflectance spectrophotometer.

Material and Methods

Data collection

A multi-centric, community-based study was conducted by collecting pigmentation data from 574 unrelated healthy individuals (aged 18 to 40 years) belonging to six different geographical locations i.e., Jammu & Kashmir (Sunni Muslim n = 80), Himachal Pradesh (Lahaul-Spiti tribe n = 85), Haryana (Jaat n = 113), Punjab (Punjabi Khatri n = 105), Uttarakhand (Garhwali n = 100), and Uttar Pradesh (Gurjar n = 91) through written informed consent [Figure 1]. Samples were collected from six different centers and participants



Figure 1: Map of the study area and locations of six sampling sites (northern India)

belong to the predominant population residing in the locality or state. For each participant, the last two generations (parents and grandparents) were reported to be born in the same state and belonged to the same community being sampled. Relocated volunteers were excluded. Volunteers with known skin or pigmentary disorders or undergoing topical treatments or supplements that may alter pigmentation, or bearing tattoos on the assessed site were excluded from the study. Participants were allowed to rest for at least 30 minutes before sample collection. Skin reflectance (MI and EI) was measured using DSM II ColorMeter (Cortex Technology, Hadsund, Denmark), a hand-held DermaSpectrometer (consisting of red and green light-emitting diodes). The measurements were performed in triplicates from the volar surface of the upper-left arm (least exposed to ultraviolet (UV) to avoid interference due to facultative pigmentation. The instrument was calibrated before each reading as per the manufacturer's instructions. The study design was approved by the Institutional Ethics Committee.

Statistical analysis

MI and EI measurements were expressed by calculating the mean (M) of triplicate readings. Pigmentation and erythema differences concerning populations and genders were evaluated using ANOVA and unpaired t-test, respectively. In an attempt to assess the linear relationship between melanin and erythema indices, Pearson's correlation coefficient was computed. An alpha level of 0.5 was utilized. We considered P < 0.001 to be significant.

Results

Variation in MI and EI, and effect of geographical location

Analysis of skin pigmentation shows wide variation in MI (Range = 17.93-56.92, M = 35.80, SD = 6.26)



Figure 2: Heatmap Matrix for A. Melanin Index, and B. Erythema Index values obtained in the study population

and EI (Range = 4.92-18.82, M = 10.48, SD = 2.68) among samples from all geographic locations [Figure 2]. Jammu & Kashmir inhabitants exhibited the lowest mean MI (M = 27.52, SD = 5.23), followed by Haryana (M = 34.5, SD = 4.81), Punjab (M = 35.21, SD = 3.68), Himachal (M = 37.27, SD = 4.11), Uttarakhand (M = 39.32, SD = 5.48), and Uttar Pradesh (M = 40.11, SD = 5.40) [Figure 3]. On the other hand, the lowest



Figure 3: Distribution of skin melanin index in North Indian population. Histograms of each geographical location show the frequency distribution of MI. (a) Jammu & Kashmir (17.93 to 43.52), (b) Haryana (27.86 to 52.03), (c) Himachal Pradesh (29.09 to 48.67), (d) Punjab (23.33 to 47.32), (e) Uttarakhand (27.02 to 56.92), and (f) Uttar Pradesh (28.52 to 52.73). Bar color corresponds approximately to the median skin colour observed in that range

mean EI was observed in Punjab (M = 8.97, SD = 1.93), followed by Haryana (M = 9.56, SD = 2.62), Jammu & Kashmir (M = 9.65, SD = 2.26), Himachal (M = 10.75, SD = 2.43), Uttar Pradesh (M = 12, SD = 2.14), and Uttarakhand (M = 12.15, SD = 2.58) [Figure 4]. There is a significant positive correlation between melanin and erythema indices { $r_{(572)} = 0.68$, P < 0.001}. The analysis of variance [Table 1] suggests that geographical location has a statistically significant impact on melanin { $F_{(5.568)} = 73.37$, P < 0.001} and erythema indices { $F_{(5.568)} = 31.07$,



Figure 4: Distribution of skin erythema index in North Indian population. Histograms of each geographical location show the frequency distribution of El. (a) Jammu & Kashmir (6.44 to 16.28), (b) Haryana (5.09 to 14.44), (c) Himachal Pradesh (5.01 to 16.00), (d) Punjab (4.92 to 15.41), (e) Uttarakhand (6.30 to 18.82), and (f) Uttar Pradesh (6.37 to 15.71)

Table 1: Details of Sample, Sampling locations, mean MI and EI and ANOVA results												
Geographic location	Ethnic		Coordinates			Melanin Index variation				Erythema Index variation		
(Groups) background						Count (n)	Sum	Average	Variance	Sum	Average	Variance
Srinagar, Jammu &	Sunni M	luslim	34.0837	° N, 7	4.7973° E	80	2202.08	27.52	27.37	772.33	9.65	5.12
Kashmir												
Sonipat, Haryana	Jaat		28.9931	° N, 7	7.0151° E	113	3899.48	34.5	19.33	1081.33	9.56	6.31
Pauri, Uttarakhand	Garhwali		30.1471	° N, 7	8.7745° E	100	3932.27	39.32	36.1	1215.38	12.15	8.35
Patiala, Punjab	Punjabi	Khatri	30.3398	° N, 7	6.3869° E	105	3697.53	35.21	15.4	941.91	8.97	4
Kullu, Himachal Pradesh	Lahaul S	Spiti tribe	31.9592	° N, 7	7.1089° E	85	3168.03	37.27	16.61	913.77	10.75	5.84
Dankaur, Uttar Pradesh	Gurjar		28.3477	° N, 7	7.5533° E	91	3650.32	40.11	30.58	1092.76	12	4.37
ANOVA	Melanin Index variation						Erythema Index variation					
Source of Variation	SS	df	MS	F	Р	F crit	t SS	df	MS	F	Р	F crit
Between Groups	8819.09	5	1763.8	73.4	3.13E-	59 2.22	886.4	1 5	177.3	31.1	5.48E-28	3 2.22
Within Groups	13654.3	568	24.03				3240.	2 568	5.7			
Total	22473.3	573					4126.	6 573				

df: Degree of freedom, EI: Erythema index, F: F-ratio (ratio of two variances), F crit: F critical value, MI: Melanin index, MS: Mean sum of squares, P: P value (significance level), SS: Sum of squares

P < 0.001}. To test the effect of altitude, the populations were grouped into hilly (Kashmir, Kullu, Pauri) and plain areas (Sonipat, Patiala, Dankaur). T-test exhibited significant difference among hilly and plain areas in terms of EI { $t_{(572)} = 3.89$, P < 0.001} but not with MI { $t_{(572)} = -2.48$, P > 0.001}.

Skin pigmentation differences in male and female

Females (n = 178) in the study group were associated with lower MI (M = 34.31, SD = 5.62) and EI (M = 8.78, SD = 2.15) than males {(n = 396), MI (M = 36.47, SD = 6.42), EI (M = 11.24, SD = 2.54)}. To test the statistical significance of the hypothesis, an independent sample t-test assuming unequal variance was performed. A significant effect of gender was observed with MI { $t_{(386)}$ = -4.06, P < 0.001} and EI { $t_{(386)}$ = -11.96, P < 0.001} with males being more pigmented than females.

Discussion

Variations in skin color across populations have been shaped by several genetic and evolutionary factors. Apart from melanogenesis, human skin color is also regulated by various growth factors, keratinocytes, and cytokines.^[2] The mainland of India extends between 8.0689°N and 37.08586°N with varying ultraviolet radiation (UVR) intensity. Along with other intrinsic and extrinsic factors, the biochemical effects of UV rays and multilayered endogamy regulated by rigid marital practices play a major role in determining India's pigmentation heterogeneity. On comparison with published datasets for MI in India, a lower MI range was noted in the current study. [11-13] The observed significant association of skin melanin { $F_{(5.568)} = 73.37$, P < 0.001 with geographical location can be attributed to the intensity of UVR. Previous studies suggested that geographical stratification of skin color can be correlated with the latitude and intensity of UVR.[13,14] Sarkar and Nandineni reported an inverse correlation between skin pigmentation and latitude, suggesting that UVR has been a driving evolutionary force in shaping skin color variation during the migration of humans from Africa to higher latitudes.^[14] Lower latitudes favor higher melanin production and darker pigmentation to minimize folate depletion and dermal degradation, with melanin functioning as a natural sunscreen. Whereas, individuals with lighter skin had better survival rates in low UV regions due to adequate vitamin D synthesis.[15-17]

In the current study, we report an inverse relationship between skin color and corresponding latitude wherein the study locations with the highest (34.08) and lowest (28.34) latitudes correspond with the lowest (27.52) and highest (40.11) mean MI, respectively [Table 1]. However, no significant correlation was observed between MI and latitude. The reason could be the geographical proximity of Haryana, Punjab, Uttarakhand, and Himachal Pradesh. We would also like to highlight a strong association between EI and geographical location { $F_{(5,568)} = 31.07$, P < 0.001}. The effect of altitude on EI { $t_{(572)} = 3.89$, P < 0.001} can be associated with increased concentration of hemoglobin in hilly areas.

The observed effect of gender on MI { $t_{(386)} = -4.06$, P < 0.001} and EI { $t_{(386)} = -11.96$, P < 0.001} can be answered on account of various hypotheses that have been previously proposed to explain the sexual dimorphism observed in skin pigmentation among humans like hormonal influence on melanocyte activity, the difference in clothing habits, rich blood flow to the skin surface in males, higher calcium needs during pregnancy and lactation, and mate choice of males for lighter than an average skinned sexual partner.^[18,19]

Conclusion

The result of the current study suggests that geographical location and gender have a significant effect on pigmentation and vascularization. The effect of geographical location on melanin { $F_{(5,568)} = 73.37$, P < 0.001} and erythema indices { $F_{(5,568)} = 31.07$, P < 0.001} may be attributed due to underlying genetic variation among populations. Furthermore, it has been observed that variation in erythema levels affects the melanin index and vice versa. The strong positive correlation between MI and EI { $r_{(572)} = 0.68$, P < 0.001} is not surprising since melanin absorbs light in a broad range of wavelengths (including red which primarily contributes to erythema).

Limitations

Though skin vascularization is affected by various factors, we have considered only MI and EI in this study. The latitudinal differences in the geographical distribution of the population included in the study were not significant. Hence, the inclusion of populations from central and south India to study the effect of latitude on skin pigmentation would yield a better result. Finally, the effects of various factors like the working nature of the population, clothing habits, duration, and frequency of UV exposure could not be controlled.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form, the patient(s) has/have given his/her/their consent for his/ her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Conflicts of interest

There are no conflicts of interest.

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