Linkage disequilibrium study on SNPs in Skin Pigmentation Genes: Chromosome specific Markers for Vitamin D Deficiency

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Abstract :

Vitamin D deficiency is one of the pandemic. Serum25(OH)D is inversely proportional to skin pigmentation. So, melanin pigmentation in skin has a pragmatic role in Vitamin D level maintenance. Genes with great relevance for different aspects of skin pigmentation, including melanosomal biogenesis (ATP7A, PMEL, PLDN), melanosomal transport (RAB27A, MLPH), or various melanocyte signaling pathways (MITF, PAX3) may be involved in the pathway. SNPs data for the followings (rs9328342 [EXOC2], rs10830253 [TYR], rs7129973 [TYR], rs4121401 [TYR], rs2000553 [TYR], rs12001326 [PRKACG], rs2150097 [TYRP1], rs13078182 [MITF] and rs17006281 [MITF]) (rs7565264 (MLPH), rs10932949 (PAX3), rs2069408 (PMEL), rs7569427 (MLPH), rs9328451 (BLOC1S5), rs12469812 (MLPH), rs2292881 (MLPH), rs6454677 (CNR1))was retrieved. LD Tag SNP utility was utilized to select SNP list accounting Linkage disequilibrium (LD) from different continental populations (African, European American African, and Asian). Linkage disequilibrium study identifies association of marker SNPs significantly contributing low level of 25(OH)D at chromosomal level for genes engaged in skin pigmentation pathway, highlighting probable genetic basis of Vitamin D synthesis in skin and deficiency in circulation.

Keywords :

Vitamin D, Gene, SNP, LD**.** Vitamin D deficiency is a pandemic in different regions of world. It is a fat-soluble steroid hormone, playing pivotal role in absorption of dietary calcium, crucial mineral metabolite for bone health. It has dual nature of function as endocrine molecule involved in calcium homeostasis and as exocrine molecule regulate nuclear vitamin D receptor expression through genetic transcription (Tsiaras and Weinstock,2011). Most dependent circulating form of Vitamin D is 25-hydroxy Vitamin D (25(OH)D) which one is mainly synthesized in skin through absorption of sunlight (UV-B, 300±5nm) from 7-dehydrocholesterol(Holick et al 1980, Maclaughlin et al 1982, Holick 1995).IOM prescribed the level of 20ng/mL 25(OH)D as standard level for a population (among 97.5%). But serum25(OH)D is inversely proportional to skin pigmentation. Eumelanin (Black Pigment) dominate in dark skin and pheomelanin (Yellow pigment) also in brighter shades. So, melanin

pigmentation in skin has a pragmatic role in Vitamin D level maintenance. Pigmentation related genes are also associated.

A complex network of genes was engaged in skin pigmentation process (Slominski and Postlethwaite,2015). Individual genes regulate different aspects of skin pigmentation, such as melanin synthesis (TYR, TYRP1, and DCT), tyrosine transport (OCA2), melanogenic signaling (PRKAR2B), melanocyte development (FGF2, EDN3, and CTNNB1). The association of several genes related to skin pigmentation, including exocyst complex component 2 (EXOC2), tyrosinase (TYR), tyrosinase-related protein 1 (TYRP1) and dopachrome tautomerase (DCT) with vitamin D status also (Saternus et al. 2015). The significance of these single nucleotide polymorphisms (SNPs) were confirmed (rs9328342 [EXOC2], rs10830253 [TYR], rs7129973 [TYR], rs4121401 [TYR], rs2000553 [TYR], rs12001326 [PRKACG], rs2150097 [TYRP1], rs13078182 [MITF] and rs17006281 [MITF]) through allelic test . SNPs showed a significant association with 25(OH)D serum levels are rs2150097 (TYRP1), rs7356986 (EDN1), and rs12001326 (PRKACG). Seven SNPs are situated within the coding sequence of a gene, rs1042602 (TYR) is associated with low serum 25(OH)D concentration. The SNP rs1042602/A192C (TYR) is a nonsynonymous substitution (Ser to Tyr) (Shriver et al .2003).

To gain further insight into the role of genes involved in skin pigmentation, SNPs in 15 other genes with great relevance for different aspects of skin pigmentation, including melanosomal biogenesis (ATP7A, PMEL, PLDN), melanosomal transport (RAB27A, MLPH), or various melanocyte signaling pathways (MITF, PAX3) and association with serum 25(OH)D concentration were analyzed (Rosenberg et al 2016). However, these genes exert many other functions unrelated to the pigmentation pathway. Using allelic tests, significance of the 8 SNPs (rs7565264 (MLPH), rs10932949 (PAX3), rs2069408 (PMEL), rs7569427 (MLPH), rs9328451 (BLOC1S5), rs12469812 (MLPH), rs2292881 (MLPH), rs6454677 (CNR1) were confirmed.

There are limited data on genetic determinants of serum 25(OH)D concentration. Large Genome Wide Association Studies(GWAS) have already identified 3 genetic loci that are associated with 25(OH)D levels, located in genes encoding for the vitamin D binding protein (GC, group specific component), for an enzyme involved in the formation of the vitamin D precursor 7-dehydrocholesterol and for enzymes catalyzing hydroxylation of vitamin D metabolites (Wang et. al.2010). Genes related to skin pigmentation were not identified in that analyses. Linkage disequilibrium studies of SNPs in different genes may enlighten chromosome spe-

cific linked domain associated with skin pigmentation and vitamin deficiency. Objective of the present study was to identify association between SNPs associated with skin pigmentation and vitamin D level through Linkage disequilibrium study, identify any novel SNP associated with neighbouring domain and any associated chromosomal domain.

Materials and Methods:

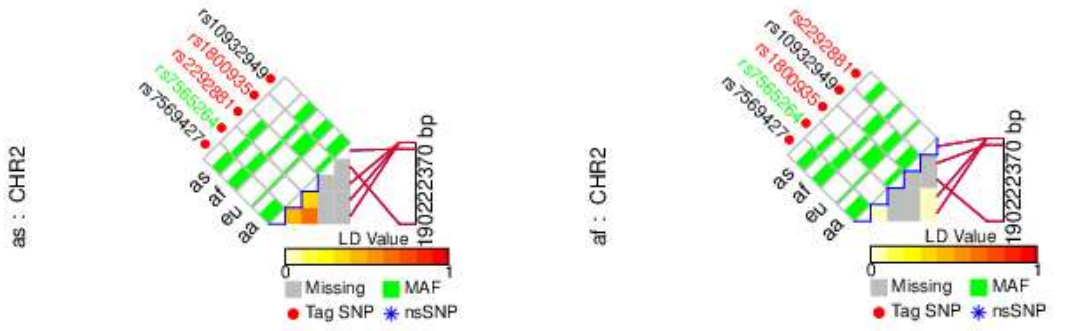
SNPs data for the followings (rs9328342 [EXOC2], rs10830253 [TYR], rs7129973 [TYR], rs4121401 [TYR], rs2000553 [TYR], rs12001326 [PRKACG], rs2150097 [TYRP1], rs13078182 [MITF] and rs17006281 [MITF]) (rs7565264 (MLPH), rs10932949 (PAX3), rs2069408 (PMEL), rs7569427 (MLPH), rs9328451 (BLOC1S5), rs12469812 (MLPH), rs2292881 (MLPH), rs6454677 (CNR1))was retrieved from 1000Genome(http://www.ncbi.nlm.nih.gov/variation/tools/1000genomes/?assm=GCF_000001405.25). SNP selection was done in a web-based SNP selection tool 'SNPinfo' freely available at http://niehs.nih.gov/supinfo (Xu and Taylor,2009). LD Tag SNP utility was utilized to select SNP list accounting Linkage disequilibrium (LD) from different continental populations (African, European American African, and Asian). Here LD was measured by r2 (Xu et.al 2007). All SNPs were organized into chromosome groups and LD structure and genotype array were represented in figures.

Results:

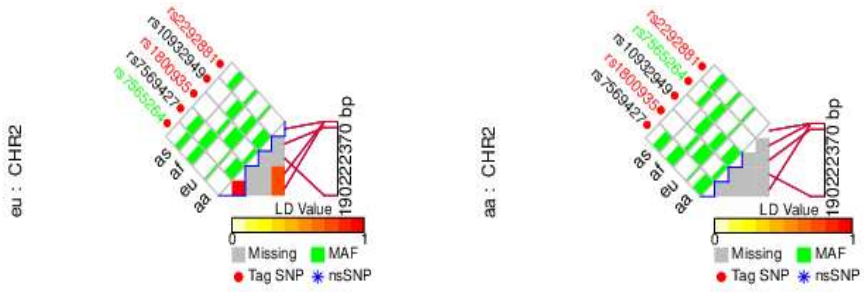
In LD graphs SNP names were coloured as black for intronic region, red in coding region, orange in splicing region, blue in Untranslated Region (UTR) and green in non-genic region. Tag SNPs and non-synonymous SNPs were assigned with flags. Height of the green bar had reflected the minor allele frequency in each population. Different colours in graph indicate Pair wise LD values. Linkage disequilibrium study (LD Taq) had identified positive correlation between two sets of SNPs in two chromosomes (Chromosome 2 and Chromosome 6) within all continental population set. In Chromosome 2, rs10932949 (PAX3), rs1800935(?), rs2292881 (MLPH), and rs7568427 (MLPH) SNPs are in one set, of which rs1800935 are not in query list(Fig1). This novel SNP is associated with MSH6 gene (DNA Mismatch Repair Protein) causing missense variant. In Chromosome 6 rs7356986 (EDN1), rs16872592 (EDN1), rs6454677 (CNR1), rs9328451 (BLOC1S5), rs9328342(EXOC2) and rs16872602 (EDN1) are associated (Fig3). In Chromosome 3, SNP rs17006281(MITF) is present in African and African American population, but rs17006281(MITF) and rs13078182 (MITF) both are in Asian and European population (Fig2). Similarly, in Chromosome 11, rs10830253 (TYR) and rs7129973 (TYR) and in Chromosome 12, rs2069408 (PMEL) are only associated with Asian and European population (Fig5&6). And in Chromosome 9, rs12001326 (PRKACG) and rs2150097(TYRP1) are also associated with all continental populations (Fig 4).

Discussion:

As it is mentioned earlier that a genome-wide association study has identified genetic variants at three loci, namely 4p12 (rs2282679 in GC), 11q12 (rs12785878 near DHCR7), and 11p15 (rs10741657 near CYP2R1) as determinants of vitamin D insufficiency [4].Earlier study confirmed that SNPs rs10932949 (PAX3), rs2292881 (MLPH), and rs7568427 (MLPH) are associated significantly with low 25(OH)D level , present in chromosome 2 among all continental population set (Saternus et. al.2015, Rosenberg et al 2016). Similarly, SNPs rs7356986 (EDN1), rs16872592 (EDN1), rs6454677 (CNR1), rs9328451 (BLOC1S5), rs9328342(EXOC2) and rs16872602 (EDN1) also associated with low 25(OH)D level, residing on chromosome6 (Saternus et. al.2015, Rosenberg et al 2016). In chromosome 9 also rs12001326 (PRKACG) and rs2150097(TYRP1) are associated with low level of 25(OH)D ((Saternus et. al.2015, Rosenberg et al 2016)). But SNPs rs13078182 (MITF) (in chromosome 3), rs10830253 (TYR) and rs7129973 (TYR) (in chromosome 11) and rs2069408 (PMEL) (in chromosome 12) are present only among European and Asian population and associated with low 25(OH)D ((Saternus et. al.2015, Rosenberg et al 2016)). Linkage disequilibrium study identifies association of independent marker SNPs significantly contributing low level of 25(OH)D at chromosomal level for genes engaged in skin pigmentation pathway, highlighting probable genetic basis of Vitamin D synthesis in skin and deficiency in circulation.

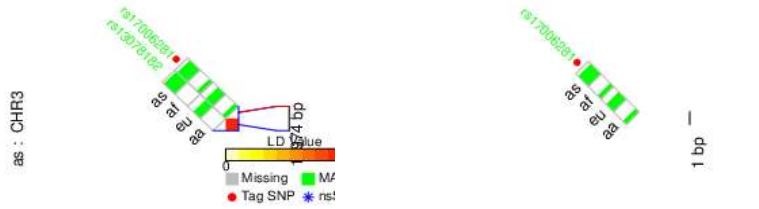


a b

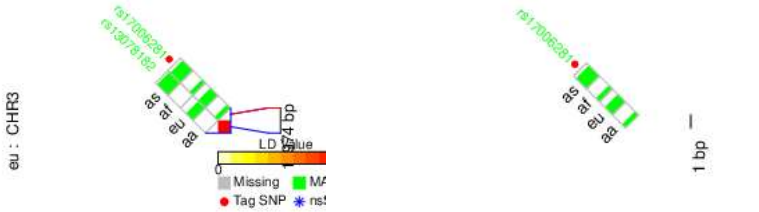


c d

Fig 1. showing LD graph of SNPs in Chromosome 2 in a) Asian; b) African; c) European and d) African American Population.

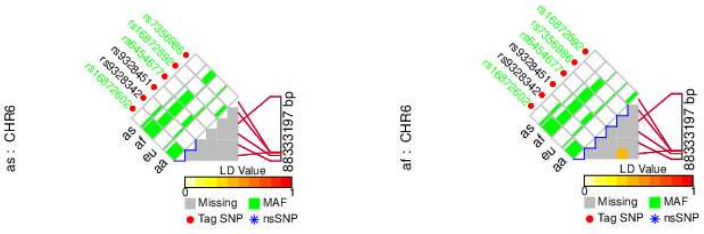


a b

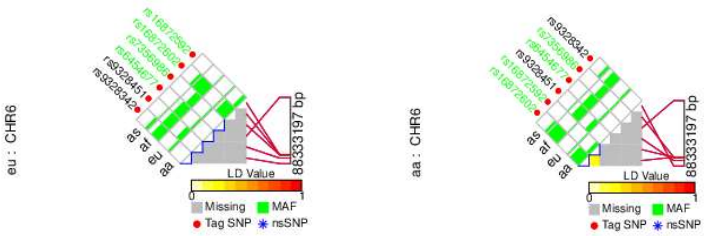


c d

Fig 2 . showing LD graph of SNPs in Chromosome 3 in a) Asian; b) African; c) European and d) African American Population.

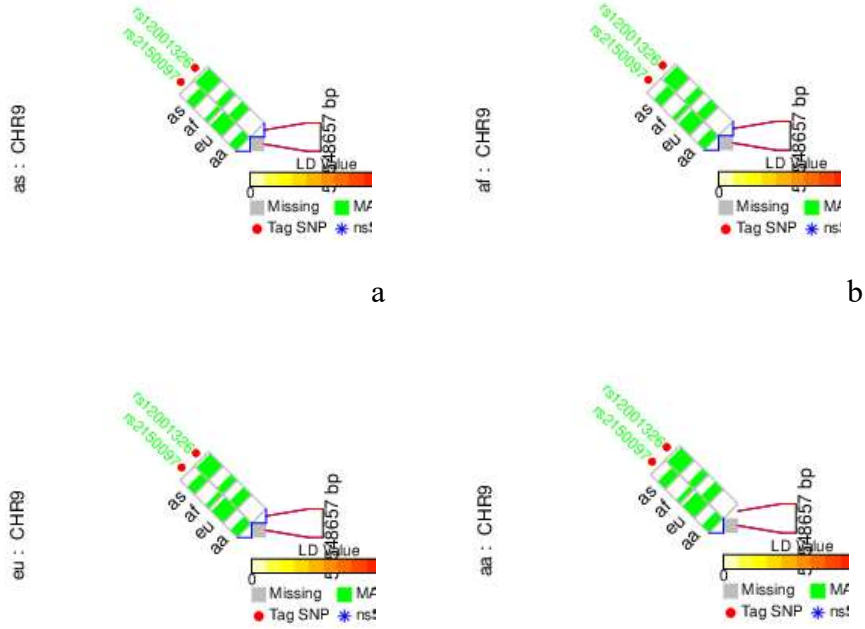


a b



c d

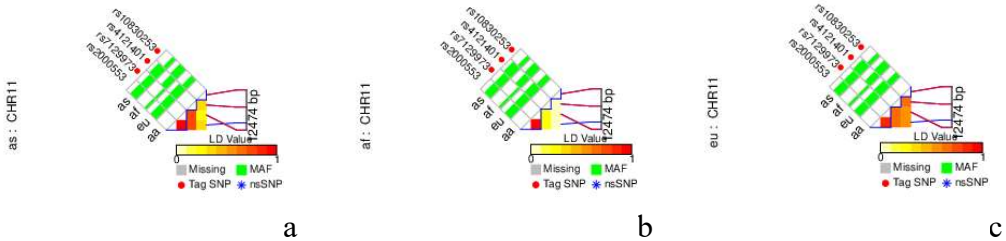
Fig 3 . showing LD graph of SNPs in Chromosome 6 in a) Asian; b) African; c) European and d) African American Population.



a b

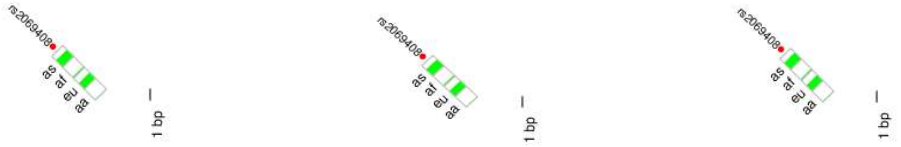
c d

Fig 4. . showing LD graph of SNPs in Chromosome 9 in a) Asian; b) African; c) European and d) African American Population.



a b c

Fig 5 . showing LD graph of SNPs in Chromosome 11 in a) Asian; b) African; and c) European Population



a b c

Fig 6. showing LD graph of SNPs in Chromosome 12 in a) Asian; b) African; and c) European Population.

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