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Cytokine SNPs: Comparison of Allele Frequencies by Race & Implications for Future Studies

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Abstract

The role of inflammation is being considered in chronic diseases. Previous studies have examined SNPs in a few key inflammatory genes and have included small numbers of African American participants. Variation in the frequencies of inflammatory pathway SNPs may help to explain racial disparities in disease risk. Through a population-based study of 103 African American and 380 Caucasian unrelated, healthy women, we examined the relationships between race and allele frequencies of 70 cytokine and cytokine receptor SNPs. The associations between genotypic and haplotype frequencies and race were also analyzed. Allelic frequencies for 52 out of the 70 SNPs meeting criteria for analysis differed significantly by race. Of the 32 pro-inflammatory and 20 anti-inflammatory SNPs for which the allele frequencies varied significantly by race, variant allele frequency differences between Caucasians and African Americans ranged between 6%–37% and 7%–53% for pro-inflammatory SNPs and anti-inflammatory SNPs, respectively. Our findings suggest that while allele frequencies do vary by race, racial groups are not simplistically represented by a pro-inflammatory or anti-inflammatory genetic profile. Given the racial variability in allele frequencies in inflammatory gene SNPs, studies examining the association between these SNPs and disease should at least incorporate self-reported race in their analyses.

Keywords

Cytokines; SNPs; Racial Differences; Women

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1.1. INTRODUCTION

Differences by race in incidence for diseases associated with inflammation suggest that there may be underlying racial differences in inflammatory pathway genes. For example, there is racial variation in the incidence of several autoimmune diseases such as systemic lupus erythematosus (SLE) and multiple sclerosis (MS), and infectious diseases such as tuberculosis, septicemia, and HIV/AIDs [1]. The incidence of several types of cancer associated with inflammation and/or chronic infection, including colorectal, liver and bile, lung and bronchus, prostate, and stomach, is also elevated in African Americans relative to Caucasians, especially among men [2]. Finally, several markers of inflammation, including C-reactive protein and homocysteine, have been shown to be associated with cardiovascular disease and to be elevated in African Americans [3].

One set of markers of inflammatory pathways are cytokines and cytokine receptors. Differences in protein expression of IL-6 SR between healthy, older African Americans and Caucasians have been shown in at least one study [4]. While few studies have examined the relationship between cytokine protein expression and race, studies investigating the association between cytokine single nucleotide polymorphisms (SNPs) and race have been previously published [5–18]. These studies have focused on SNPs in only a few cytokines including tumor necrosis factor (*TNF*), *IL1A*, *IL1B*, *IL6*, *IL2*, *IFNG*, *IL18*, and *IL10*.

The consensus from these published reports is that among African Americans there is an increase in the frequency of alleles associated with high production of pro-inflammatory T helper type I (T_H1) cytokines and low production of anti-inflammatory T helper type II (T_H2) cytokines. These results suggest the potential for systemic inflammatory upregulation in African Americans. Specifically, *IL1A* -889T, *IL1B* -3957C and -511A, *IL6* -174G, *IL18* -137G, *TNFA* -308A, and *IFNG* -874 alleles are associated with an increase in cytokine production and are found more frequently among African Americans [7,9,11,13]. *IL10* -592A, -819T, and -1082A alleles, associated with decreased IL-10 production, are also found more frequently among African Americans [7,9,13,15]. In contrast, another study reported that the pro-inflammatory *IL2* -330G allele, associated with increased IL-2 production, was found less frequently among African Americans than among Caucasians [9]. Furthermore, some studies have found no relationship between allele frequency of the *TNFA* -308 polymorphism and race [7,9,11].

To expand this research, we examined the association between race and 70 cytokine and cytokine receptor polymorphisms in 26 inflammation-related genes among African American and Caucasian women in a large, population-based study.

1.2. MATERIALS & METHODS

1.2.1 Study Participants

Subjects were population-based healthy controls obtained through a study comparing controls frequency matched on age and self-reported race to women with non-small cell lung cancer (NSCLC) [19]. Control participants were women without a history of NSCLC between the ages of 18 and 74 living in the Detroit metropolitan area and were identified through random-digit dialing. Of the households that completed the eligibility screening questionnaire, 69.6% (N=575) participated in the interview. 209 women refused to participate. Excluded from analyses were 11 controls whose self-reported race was not African American or Caucasian. Four-hundred, eighty-three controls provided a blood sample, were genotyped, and are included in the analyses.

1.2.2. Sample Collection & Genotyping

Blood was collected in Vacutainer Plus tubes containing EDTA. DNA was isolated from whole blood with a Qiagen AutoPure LS the Genomic DNA Purification System (Gentra Systems, Minneapolis, MN) following the manufacturer's protocols.

Genomic DNA 250 ng was submitted to the Wayne State University Applied Genomics Technology Center for genotyping. The Illumina GoldenGate assay using the Cancer SNP Panel was utilized. This Panel consists of primers to interrogate 1421 SNPs in 408 genes, including 83 cytokine and cytokine receptor SNPs, selected from the National Cancer Institute's Cancer Genome Anatomy Project SNP500 Cancer Database (http:/snp500cancer.nci.nih.gov/). The GoldenGate assay was run according to the manufacturer's directions. These data were analyzed using Bead Studio software (Illumina).

1.2.3. Statistical Analysis

χ^2 goodness-of-fit test was used to assess whether SNPs were in Hardy-Weinberg Equilibrium (HWE) among African American and Caucasian controls separately. To compare the distributions of allele frequencies by race, Pearson's χ^2 analysis was conducted. Cochran-Mantel-Haenszel test for trend and χ^2_G, or Fisher's Exact test where appropriate, were performed to assess the association between genotype and race. FSTAT V2.9.3.2 was used to calculate the Weir & Cockerham estimations of Wright's fixation index, Fst, which ranges from 0 to 1, with higher numbers indicating greater genetic distance between the two populations [20]. As negative unbiased estimates of Fst have no biological meaning, negative values of Fst were set to zero. PLINK V1.03 (Shaun Purcell, http://pngu.mgh.harvard.edu/purcell/plink/) was used to construct haplotypes separately for Caucasians and African Americans [21]. Haplotypes with at least a 1% frequency in either race are reported. χ^2 analysis was conducted to test for differences in the distribution of haplotype frequencies between African Americans and Caucasians. p-values were corrected for multiple comparisons via the Benjamini & Hochberg False Discovery Rate (FDR) method. Except for haplotype construction, all analyses were conducted in SAS/Genetics v. 9.1 (SAS Institute, Cary, NC). Finally, to validate the allele frequencies obtained in this study, frequencies for Caucasian residents of Utah (CEU) and African American residents of the American Southwest (ASW) were downloaded from HapMap Phase II + III data released in November 2008.

2. RESULTS

2.1.1. Sample

Our sample included 103 population-based African American women (21.3%) and 380 Caucasian (78.7%) women.

2.1.2. Cytokine & Cytokine Receptor SNPs

Five (*IL3*-rs40401; *IL12B*-rs730690; *TNF*-rs3093661; *IL10RA*-rs9610; *TGFβ1*-rs1800471) and two (*IL8RB*-rs1126579; *IFNGR1*-rs11914) out of 83 cytokine and cytokine receptor SNPs on the cancer SNP panel were not in Hardy-Weinberg equilibrium for African Americans and Caucasians, respectively. For an additional six SNPs (*IL8*-rs2227549; *IL8RB*-rs1126579; *IFNGR1*-rs3799488; *TNF*-rs3093661; *LTA*-rs3093546, *IL10*-rs3024509; *IL10*-rs3021094; *IL10RA*-rs2229114) variant allele frequencies were less than 5%, leaving 70 cytokine and cytokine receptor SNPs for analysis.

2.1.3. Allelic Frequencies by Race

Allelic frequencies for 52 out of the 70 SNPs meeting criteria for analyses differed significantly between Caucasians and African Americans (Table 1). For 12 out of the 70 SNPs analyzed,

the minor alleles differed between the two racial groups; therefore, in each table the variant allele is defined as the minor allele among Caucasians. Allelic frequencies obtained from HapMap were strikingly similar to those in our study.

2.2.1. Pro-inflammatory Cytokine & Cytokine Receptor SNP Allelic Distributions by Race

Of the 41 SNPs in pro-inflammatory cytokine and cytokine receptor genes, allele frequencies differed by race for all but nine (Table 1). Three of the SNPs that did not vary by race were the three SNPs in *TNF*. Otherwise, the five remaining SNPs not varying by race were in genes in which other SNPs were genotyped and found to vary by race (including *IL1A*, *IL1B*, *IL15*, *IL15RA*, and *TNFRSF10A*). All SNPs genotyped in *IL2*, *IL6*, *IL6R*, *IL7R*, *IL8*, *IL8RA*, *IL12A*, *IL12B*, *IFNAR2*, *IFNG*, *IFNGR2*, *LTA* and *TNFRSF1A* showed significant variation in allele frequency by race. Of the 32 pro-inflammatory SNPs for which the allele frequencies statistically significantly varied by race, the average absolute difference in variant allele frequencies between Caucasians and African Americans was 0.20 +/− 0.08 with a range from 0.06 to 0.37. Fst estimates for the 32 pro-inflammatory SNPs that varied by race ranged from 0.016 to 0.283. Three of these estimates, for rs4073-*IL8*, rs2854386-*IL8RA*, and rs1059293-*IFNGR2*, exceeded 0.200.

2.2.2. Anti-inflammatory Cytokine & Cytokine Receptor SNP Allelic Distributions by Race

Of the 29 SNPs in anti-inflammatory cytokine and cytokine receptor genes, allele frequencies differed by race in all but nine. All SNPs in *IL1RN*, *IL4* and *IL10* demonstrated significant racial differences in allele frequencies. Six of the nine SNPs in *IL4R* and two of four SNPs in *IL13* also showed variation in allele frequency by race. Of the 20 anti-inflammatory SNPs for which the allele frequencies statistically significantly varied by race, the average absolute difference in variant allele frequency between Caucasians and African Americans was 0.27 +/− 0.12 with a range from 0.07 to 0.53. Estimates of Fst for these 20 anti-inflammatory SNPs ranged from 0.029 to 0.509. Seven of these estimates, in rs380092-*IL1RN*, rs2243250-*IL4*, rs2070874-*IL4*, rs1805011-*IL4R*, rs1801275-*IL4R*, rs1805016-*IL4R*, and rs1295686-*IL13*, exceeded 0.200.

2.3. Genotypic Frequencies by Race

With the following exceptions, the same SNPs with statistically significant differences in allelic frequencies by race also had statistically significant differences in genotype frequencies by race (data not shown). The distribution of genotypes for *IL7R*-rs7737000 was no longer significantly different by race. While there was a statistically significant trend for a higher proportion of the A allele at this SNP among Caucasians (p=0.02), the χ^2 test for a general association between genotype and race was not statistically significant (p=0.07). Similarly, *IFNAR2*-rs2236757 had an increase in allele frequency among Caucasians (p=0.02); however, the χ^2 test for a general association between genotype and race was not statistically significant (p=0.08). χ^2 test for a general association and Fisher's Exact test could not be calculated for one SNP, *IL15RA*-rs2296141, due to sparse data.

2.4. Haplotypes by Race

The 70 SNPs meeting criteria for analysis were located in 26 genes with multiple polymorphisms in 18 of these genes. Within gene haplotypes common to both African American and Caucasian women were discerned for all 18 of these genes including: *IL1A*, *IL1B*, *IL1RN*, *IL2*, *IL6*, *IL7R*, *IL8*, *IL15*, *IL15RA*, *IFNAR2*, *TNF*, *TNFRSF1A*, *TNFRSF10A*, *IL4*, *IL4R*, *IL10*, *IL13*, and *TGFβR1* (Table 2). Two haplotypes were identified per gene in *IL4* and *IL4R*. Of the 20 haplotypes, the distributions of haplotype frequencies differed significantly by race for all haplotypes constructed except for the *TNF* haplotype rs1799964-rs1800630-rs1800629.

Finally, our results remained statistically significant even after adjusting for multiple comparisons.

3. DISCUSSION

We observed allelic frequency differences by race for 52 of the 70 SNPs meeting criteria for analysis. General genotypic frequency differences between African Americans and Caucasians were observed for 49 of these 52 SNPs. Significant trends in genotypic frequency by race were found for all of these 52 SNPs. While the same haplotypes were constructed for African Americans and Caucasians for all genes for which multiple SNPs were analyzed, there was marked variation in haplotype frequency by race.

3.1 Pro-inflammatory Cytokines & Cytokine Receptors

Previous studies that have examined SNPs in *IL1*, *IL2*, *IL6* and *TNF* genes have found differences between African Americans and Caucasians in allelic frequencies for these genes except for *TNF*.

3.1.1. IL1—Secreted primarily by macrophages and epithelial cells, IL-1 activates macrophages and T lymphocytes. In this study, we examined two *IL1A* SNPs, five *IL1B* SNPs, and three SNPs of the IL-1 antagonist gene, *IL1RN*. Allele frequencies for all but one SNP were statistically significantly different by race. Three of the *IL1B* SNPs included in our analyses have been previously studied. For the synonymous SNP, rs1143634, the T allele has been found to be less frequent among African Americans [18], and this finding is consistent with the decreased frequency of the A allele (the complement allele genotyped) in our study. Similar findings were also obtained between the previously published increased C allele frequency of rs1143627 (−31A>G) among African Americans [18]. Complement to the G allele, the C allele is associated with decreased promoter activity and decreased gene transcription, so our observation for this one SNP is inconsistent with the general hypothesis that African Americans have increased frequencies of high producing pro-inflammatory alleles. For the previously studied third *IL1B* SNP, rs16944 (−511 G>A), other researchers have found that the A allele (complement to the T allele genotyped in previous studies), which has been associated with increased transcriptional activity, is increased among African Americans. In our study, the A allele was the major allele among African Americans and the minor allele among Caucasians, supporting these results [13,18].

3.1.2 *IL2*—IL-2 is a T cell proliferation factor that acts in an autocrine fashion on T_H1 cells. The minor allele for both *IL2* SNPs studied in this project was found less frequently among African Americans. At least one of these SNPs, rs2069762 (−330A>C) has been previously studied. The G allele, associated with both an increased IL-2 production *in vitro* and a decreased IL-2 production *in vivo* [22], previously was found more frequently among African Americans [6,8]. Consistent with these results, we observed that the C allele (the complement allele analyzed in our study) was less frequent among African Americans. These findings are consistent with the assertion of increase frequency of high producing pro-inflammatory alleles in African Americans.

3.1.3 IL6—Allelic distributions of one of the two *IL6* SNPs examined in this study previously have been found to differ by race. The *IL6* SNP rs1800795 (−174G>C) is located in the promoter 5′ flanking region. The C allele creates an endonuclease restriction site producing cleaved products with 3′ overhangs; whereas, the G allele, which has been shown to be more frequent among African Americans, is associated with higher IL-6 production [7,9,11,13,16, 17]. In contrast to these findings, the G allele was found less frequently among African Americans in our study.

3.1.4.TNF & TNFRSF—Primarily produced by macrophages, tumor necrosis factor (TNF) plays a role in inflammation partly through activating NFκB, tumorigenesis, apoptosis, and immune cell regulation. The A allele of *TNF*-308 lies in a binding site for the transcription factor AP1 and has been shown to increase TNF-α production *in vitro*. Studies examining the association between *TNF*-308 allele distribution and race have shown either no association [7,13,14,16] or decreased frequency of the A allele among African Americans [6,9,18]. We found no differences between African Americans and Caucasians in the allele frequencies for this SNP and the other *TNF* SNPs. However, we did find a difference by race in three of four *TNF* receptor SNPs, one of which, rs4871857, lies in a coding region of the *TNFRSF10A* gene (also referred to as TRAIL DR4, the "death receptor"). This SNP results in an amino acid substitution from arginine to threonine in the ligand binding domain and subsequently the transition from a basic to a hydroxylic amino acid that bears a phosphorylation site. Our study suggests a decreased frequency of the C allele among African Americans in comparison with Caucasians. The functional consequence of this SNP, its association with disease, and whether allelic variation by race can explain any racial differences in disease susceptibility need to be further explored.

3.1.5. Summary of Pro-inflammatory SNPs & Race—Findings for *IL1*-511 and *IL2*-330 are consistent with the hypothesis that African Americans have an increased pro-inflammatory genetic profile associated with high cytokine production. However, results for *IL1*-31 and *IL6*-174 were contrary to this hypothesis. The relationship between these genes and race are complicated; the effect of the *IL1*-31 polymorphism on IL-1 production has been shown to depend on the *IL1*-511 SNP [23] Additionally, functional studies are not consistent; some studies of the *IL6*-174 polymorphism have indicated that the G allele increases cytokine production [24–26] while others found that the C allele increases IL-6 production [27–29]; no association between genotype and expression has also been reported [30]. Our findings are not uniformly illustrative of African Americans' having a greater likelihood of a pro-inflammatory genetic profile than whites.

3.2. Anti-inflammatory Cytokines & Cytokine Receptors

Studies have previously examined anti-inflammatory cytokine SNPs in *IL4* and *IL10* genes. We also evaluated *IL13* polymorphisms.

3.2.1. IL4—Secreted by T_H2 lymphocytes and mast cells, IL-4 acts to activate B lymphocytes, stimulate IgE class switching, and suppress T_H1 lymphocytes and is thought to play a role in asthma and hypersensitivity reactions. Nguyen et al (2004) found that the A allele of rs2243250 (−590 G>A), which is associated with increased IL-4 production, was more common among African Americans [14]. Our results confirmed this finding as the A allele was the major allele among African Americans but the minor allele among Caucasians. This discrepancy in allele frequencies may contribute to the increased prevalence of asthma among African Americans [31].

3.2.2. IL10—All five of the *IL10* SNPs examined differed by race in terms of allelic frequency, and two of these SNPs have been examined in previous studies. The rs1800896 (−1082A>G) A allele has been associated with lower IL-10 production *in vitro* by stimulated T lymphocytes and *in vivo* [32]. Either no association between allele frequencies at this SNP [7,11,14], or an increase in the A allele among African Americans has been reported [9,13,15,18]. Consistent with these latter findings, we observed an increase in the frequency of the A allele in African Americans versus Caucasians.

Our finding of an increased frequency of the A allele for rs1800871 (−819G>A) among African Americans is consistent with previously published reports [7,9,13,15]; however, studies

including a smaller number of African Americans have observed no relationship between race and allelic distribution of this SNP [11,16]. This SNP sits in an estrogen receptor response element and is part of a haplotype with *IL10*-1082 and *IL10*-592 associated with IL-10 production. The ATA haplotype, associated with lower IL-10 production, has been found to be more frequent among African Americans than Caucasians [9,13,15] or equally as frequent in a smaller study involving 42 African American women [11].

3.2.3 Summary of Anti-inflammatory SNPs & Race—Our findings of an increase in the low IL-10 producing alleles among African Americans supports the conclusion that African Americans have an increased frequency of low producing anti-inflammatory polymorphisms. Contrary to this conclusion is our finding of an increase in the *IL4* A allele associated with high levels of the anti-inflammatory IL-4, which has not been examined in previous studies of the associations between cytokine allelic frequency and race. Again, our findings suggest that while allele frequencies do vary by race, racial groups are not simplistically represented by a pro-inflammatory or anti-inflammatory genetic profile.

3.3. Other Cytokine SNPs

Other cytokine and cytokine receptor gene polymorphisms examined in this study included SNPs in *IL7R*, *IL12A* and *IL12B*, *IL8*, *IL15*, *IL15RA*, *IL4R*, *IL13*, and *TGFβ* and *TGFβR1* genes. IL-7 acts as a hemopoietic growth factor, and IL-12 plays a major role in differentiation of precursor T helper lymphocytes. The chemokine IL-8, which draws immune system cells to the site at which it is produced and is significantly increased in the lungs of chronic smokers, has been implicated in the pathogenesis of COPD. IL-15 functions similarly to IL-2, acting as a T cell and natural killer cell growth factor, and its receptor, IL-15RA has been associated with autoimmune disease. Like IL-4, IL-13 has been shown to play a role in asthma. Under normal conditions, transforming growth factor beta (TGF-β) regulates cellular proliferation and differentiation; however, it is secreted by cancer cells into the tumor environment, resulting in immunosuppression by directly acting on immune cells and converting effector T cells into suppressor T cells. While minor allele frequencies of *TGFβ* and *TGFβR1* SNPs did not differ between African American and Caucasian women, the frequency distributions of a *TGFβR1* haplotype did differ significantly between the two racial groups, supporting the assertions that haplotype analysis needs to be conducted by race and that study of individual SNPs alone may not be adequate to analyze disease-locus associations.

3.4. Strengths & Limitations

This paper has a number of strengths including the population-based design and method of recruitment, which ensure that participants are representative of the population. Our sample included a large number of African Americans. Our use of a standardized panel from the NCI's Cancer Anatomy Genome Project allows for the future comparison of our results to those of other researchers using the same panel. Moreover, we analyzed SNPs in a wider number of cytokine and cytokine receptor genes than have previously been studied in this field.

This study is not without limitations. The relationships between a number of the cytokine and cytokine receptor SNPs included in our analysis and protein expression and function have not been well characterized. Therefore, drawing conclusions about variations in inflammatory pathway function by race cannot be made based only on these data. Another limitation is that the African American women included in this study may not be representative of all African American women in the United States [33].

3.5. Conclusions

We found that cytokine and cytokine receptor SNP allelic, genotypic and haplotype frequencies varied significantly by race. These results suggest that future analyses of the associations

between cytokine and cytokine receptor SNPs and disease risk and prognosis should at least consider self-reported race, if not more complex measures such as race and socioeconomic status or genetic ancestry. Additionally, the conclusion by Ness et al (2004) that African Americans have an increased allele frequency of high producing pro-inflammatory alleles and of low producing anti-inflammatory alleles was not fully supported in this study when a larger number of SNPs were examined than in previous studies, suggesting that this general conclusion may be an oversimplification [13]. It is likely that these differences in cytokine allele frequencies may contribute to racial disparities with regard to risk and mortality of diseases such as cancer, end stage renal disease and transplantation, and preterm labor [7,9, 18,34–36], but the examination of only a handful of polymorphisms and the lack of critical factors confounding such relationships, such as socioeconomic status, has limited this work. Further research should involve a wider array of cytokine genes, as included in this study, and should assess whether these differences in cytokine genes are associated with functional differences in inflammatory pathways between African Americans and Caucasians.

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Table 1

Cytokine & Cytokine Receptor SNP Allele Frequencies by Race

Cytokine	SNP Identifier		Wild Type Allele[a]	Variant Allele[a]	Caucasians Variant Allele Frequencies	Phase III HapMap CEU Allele Frequencies	*African Americans* Variant Allele Frequencies	Phase III HapMap ASW Allele Frequencies	Fst	Comparison of Variant Allele Freq. p-value[c]
Pro-inflammatory Cytokines & Cytokine Receptors										
IL1A	rs17561	$Ala^{114}Ser$	C	A	0.32	0.26	0.17	0.17	0.046	**<0.0001**
IL1A	rs2071374	IVS4−109	A	C	0.28		0.29		0.00	0.71
IL1B	rs1071676	Ex7−97	G	C	0.26		0.14		0.032	**0.0012**
IL1B	rs1143634	$Phe^{105}Phe$	G	A	0.26	0.22	0.13	0.10	0.041	**0.0002**
IL1B	rs3136558	IVS3−123	A	G	0.24	0.22	0.17	0.12	0.009	0.06
IL1B	rs1143627	−31	A	G	0.32	0.35	**0.64**	0.65	0.176	**<0.0001**
IL1B	rs16944	−511	G	A	0.32	0.35	**0.59**	0.57	0.137	**<0.0001**
IL2	rs2069763	$Leu^{38}Leu$	C	A	0.32		0.14		0.072	**<0.0001**
IL2	rs2069762	−330	A	C	0.32		0.10		0.111	**<0.0001**
IL6	rs1800797	−597	G	A	0.37	0.52	0.07	0.08	0.182	**<0.0001**
IL6	rs1800795	−174	C	G	0.38	0.53	0.07	0.10	0.190	**<0.0001**
IL6R	rs8192284	$Asp^{358}Ala$	A	C	0.41	0.34	0.13	0.16	0.153	**<0.0001**
IL7R	rs1494555	$Ile^{138}Val$	A	G	0.29	0.28	0.17	0.16	0.033	**0.0009**
IL7R	rs7737000	$His^{165}His$	G	A	0.12	0.15	0.06	0.13	0.016	**0.02**
IL8	rs4073	−251	A	T	0.43		**0.80**		0.230	**<0.0001**
IL8	rs2227306	IVS1+298	G	A	0.39		0.14		0.126	**<0.0001**
IL8RA	rs2854386	1379 bp 3′ STP	G	C	0.06		0.32		0.283	**<0.0001**
IL12A	rs582537	IVS2−701	C	A	0.45		0.30		0.040	**0.0003**
IL12B	rs3212227	Ex8+159	A	C	0.22	0.19	0.34	0.32	0.034	**0.0008**
IL15	rs1493013	Ex3+163	A	G	0.43		0.39		0.000	0.38
IL15	rs2254514	Ex3−92	G	A	0.28		0.17		0.025	**0.0034**
IL15	rs2857261	IVS3+8	A	G	0.45		0.48		0.000	0.52
IL15	rs1057972	Ex9−181	T	A	0.45		**0.65**		0.068	**<0.0001**
IL15	rs10833	Ex9−66	G	A	0.36	0.37	0.14	0.11	0.095	**<0.0001**
IL15RA	rs2296135	Ex8−361	C	A	0.49	0.53	0.21	0.28	0.141	**<0.0001**
IL15RA	rs2296141	IVS6−242	G	A	0.08		0.10		0.000	0.39

Cytokine	SNP Identifier		Wild Type Allele[a]	Variant Allele[a]	Caucasians Variant Allele Frequencies	Phase III HapMap CEU Allele Frequencies	*African Americans* Variant Allele Frequencies	Phase III HapMap ASW Allele Frequencies	Fst	Comparison of Variant Allele Freq. p-value[c]
IL15RA	rs2228059	$Asn^{146}Thr$	A	C	0.48	0.44	**0.76**	0.68	0.136	**<0.0001**
IL15RA	rs3136614	IVS4+32	A	G	0.22	0.21	0.12	0.16	0.031	**0.0012**
IFNAR2	rs3153	IVS1−4640	G	A	0.30		0.14		0.060	**<0.0001**
IFNAR2	rs7279064	$Phe^{10}Val$	A	C	0.32	0.35	0.19	0.21	0.041	**0.0002**
IFNAR2	rs2236757	IVS6−50	G	A	0.30	0.31	0.21	0.24	0.015	**0.02**
IFNG	rs1861494	IVS2+284	A	G	0.28		0.11		0.075	**<0.0001**
IFNGR2	rs1059293	Ex7−134	G	A	0.45	0.48	**0.80**	0.78	0.207	**<0.0001**
TNF	rs1799964	−1031	A	G	0.18	0.22	0.14	0.18	0.005	0.14
TNF	rs1800630	−863	C	A	0.13	0.15	0.09	0.13	0.004	0.16
TNF	rs1800629	−308	G	A	0.17	0.18	0.14	0.07	0.000	0.38
LTA	rs909253	IVS1+90	A	G	0.35		0.48		0.031	**0.0013**
TNFRSF10A	rs2235126	IVS7+218	G	A	0.27	0.23	0.21	0.19	0.005	0.12
TNFRSF10A	rs4871857	$Arg^{209}Thr$	G	C	0.48		0.34		0.031	**0.0011**
TNFRSF1A	rs1800692	IVS4−33	G	A	0.40		0.10		0.177	**<0.0001**
TNFRSF1A	rs887477	IVS1−2572	C	A	0.48		0.23		0.114	**<0.0001**
Anti-inflammatory Cytokines & Cytokine Receptors										
IL1RN	rs419598	$Ala^{57}Ala$	A	G	0.27	0.28	0.06	0.06	0.110	**<0.0001**
IL1RN	rs454078	IVS6+59	T	A	0.27		0.06		0.108	**<0.0001**
IL1RN	rs380092	IVS6+166	T	A	0.35		**0.72**		0.238	**<0.0001**
IL4	rs2243248	−1098	A	C	0.07	0.09	0.14	0.18	0.029	**0.0018**
IL4	rs2243250	−590	G	A	0.14	0.14	**0.67**	0.60	0.509	**<0.0001**
IL4	rs2070874	Ex1−168	G	A	0.13	0.14	0.45	0.39	0.257	**<0.0001**
IL4	rs2243268	IVS2−1443	A	C	0.13	0.14	0.27	0.27	0.067	**<0.0001**
IL4R	rs2243290	IVS3−9	C	A	0.13	0.14	0.36	0.34	0.158	**<0.0001**
IL4R	rs2057768	−29429	G	A	0.29	0.31	0.30	0.24	0.000	0.67
IL4R	rs3024544	IVS3−85	G	A	0.15	0.17	0.11	0.14	0.002	0.22
IL4R	rs1805011	$Glu^{400}Ala$	A	C	0.13	0.10	**0.53**	0.55	0.369	**<0.0001**
IL4R	rs1805012	$Cys^{431}Arg$	A	G	0.12	0.10	0.11	0.07	0.000	0.75
IL4R	rs1805015	$Ser^{503}Pro$	A	G	0.17	0.16	0.39	0.35	0.130	**<0.0001**

Cytokine	SNP Identifier		Wild Type Allele[a]	Variant Allele[a]	Caucasians Variant Allele Frequencies	Phase III HapMap CEU Allele Frequencies	*African Americans* Variant Allele Frequencies	Phase III HapMap ASW Allele Frequencies	Fst	Comparison of Variant Allele Freq. p-value[c]
IL4R	rs1801275	$Gln^{576}Arg$	A	G	0.22	0.21	**0.66**		0.353	**<0.0001**
IL4R	rs1805016	$Ser^{752}Ala$	A	C	0.05	0.06	0.26	0.43	0.213	**<0.0001**
IL4R	rs8832	Ex12−313	G	A	0.43	0.50	**0.77**	0.81	0.200	**<0.0001**
IL10	rs3024496	Ex5+210	A	G	0.47	0.51	0.28	0.36	0.066	**<0.0001**
IL10	rs3024491	IVS1−286	C	A	0.47	0.51	0.19	0.30	0.149	**<0.0001**
IL10	rs1800871	−819	G	A	0.23		0.44		0.106	**<0.0001**
IL10	rs1800896	−1082	A	G	0.47	0.52	0.28	0.35	0.073	**<0.0001**
IL10	rs1800890	−3575	T	A	0.38		0.18		0.075	**<0.0001**
IL13	rs1881457	−1469	A	C	0.23	0.19	0.16	0.19	0.009	0.07
IL13	rs1800925	−1112	G	A	0.22		0.41		0.093	**<0.0001**
IL13	rs1295686	IVS3−24	G	A	0.22	0.22	**0.68**	0.57	0.371	**<0.0001**
IL13	rs20541	$Arg^{144}Gln$	G	A	0.21	0.22	0.23	0.18	0.000	0.52
TGFβ1	rs1800469	308 bp 3′ STP	G	A	0.31	0.29	0.27	0.18	0.002	0.23
TGFβR1	rs928180	IVS3−2409	A	G	0.09		0.05		0.007	0.08
TGFβR1	rs334358	IVS8+547	C	A	0.22		0.19		0.000	0.40
TGFβR1	rs868	Ex9+195	A	G	0.22	0.20	0.16	0.20	0.005	0.12

[a] Wild type and variant alleles for Caucasians.

[b] African American variant allele frequencies in bold are for SNPs for which the variant allele differed between Caucasians and African Americans.

[c] p-values adjusted by the Benjamini & Hochberg FDR method.

Table 2

Cytokine & Cytokine Receptor Haplotype Frequencies by Race Haplotypes Common to Caucasians and African Americans

		%		
Cytokine	**Haplotype**	**Caucasians**	**African Americans**	**χ^2 p-value[a]**
Pro-inflammatory Cytokines & Cytokine Receptors				
rs17561-rs2071374				
IL1A	C-A	40.4	53.4	
	A-A	31.8	17.5	
	C-C	27.8	29.1	
	Other	0	0	**0.0003**
rs3136558-rs1143627-rs16944				
IL1B	A-A-G	45.2	25.5	
	A-G-A	31.0	52.6	
	G-A-G	22.3	10.4	
	G-G-A	1.3	6.1	
	A-G-G	0.1	4.4	
	Other	0.1	1.0	**<0.0001**
rs2069763-rs2069762				
IL-2	C-A	35.3	75.5	
	A-A	32.0	14.2	
	C-C	32.6	10.3	
	Other	0.1	0	**<0.0001**
rs1800797-rs1800795				
IL6	G-C	61.7	92.7	
	A-G	37.5	7.3	
	Other	0.8	0	**<0.0001**
rs1494555-rs7737000				
IL7R	A-G	59.0	76.9	
	G-G	29.2	17.2	
	A-A	11.8	5.9	
	Other	0	0	**<0.0001**
rs4073-rs2227306				
IL8	A-G	56.7	19.6	
	T-A	39.2	14.1	
	T-G	4.1	66.3	
	Other	0	0	**<0.0001**
rs2245414-rs2857261-rs1057972-rs10833				
IL15	G-G-A-G	44.9	47.0	
	A-A-T-A	20.5	4.3	
	G-A-T-A	14.9	8.6	
	G-A-T-G	11.5	15.9	
	A-A-T-G	7.2	4.7	
	G-A-A-G	0.3	9.7	
	A-A-A-G	0.2	8.3	

		%		
Cytokine	**Haplotype**	**Caucasians**	**African Americans**	**χ^2 p-value[a]**
	Other	0.5	1.5	**<0.0001**
rs2296135-rs2228059-rs3136614				
IL15RA	C-C-A	42.7	70.5	
	A-A-A	33.0	13.4	
	A-A-G	14.0	4.9	
	C-A-G	4.8	3.6	
	C-C-G	3.5	2.2	
	A-C-A	1.9	1.7	
	C-A-A	0	2.4	
	A-C-G	0.1	1.3	
	Other	0	0	**<0.0001**
rs3153-rs7279064				
IFNAR2	G-A	67.7	81.0	
	A-C	29.6	13.6	
	G-C	2.8	5.4	
	Other	0	0	**<0.0001**
rs1799964-rs1800630-rs1800629				
TNF	A-C-G	64.2	71.8	
	A-C-A	17.3	14.6	
	G-A-G	13.0	9.2	
	G-C-G	5.4	4.4	
	Other	0.1	0	0.72
rs1800692-rs887477				
TNFRSF1A	G-C	52.2	77.2	
	A-A	39.6	9.7	
	G-A	7.9	13.1	
	Other	0.3	0	**<0.0001**
rs2235126-rs4871857				
TNFRSF10A	G-G	50.0	63.3	
	A-C	24.7	19.1	
	G-C	22.9	15.4	
	A-G	2.4	2.3	
	Other	0	0	**0.0077**
Anti-inflammatory Cytokines & Cytokine Receptors				
rs419598-rs454078-rs380092				
IL1RN	A-T-A	33.8	72.1	
	A-T-T	39.2	21.1	
	G-A-T	25.6	5.8	
	G-A-A	1.1	0	
	Other	0.3	1.0	**<0.0001**
rs2243248-rs2243250				
IL4	A-G	79.4	23.0	
	A-A	13.6	62.9	

		%		
Cytokine	**Haplotype**	**Caucasians**	**African Americans**	**χ^2 p-value[a]**
	C-G	7.0	9.5	
	C-A	0	4.6	
	Other	0	0	**<0.0001**
rs2243268-rs2243290				
IL4-IL4R	A-C	86.7	63.6	
	C-A	13.0	27.1	
	A-A	0.3	9.3	
	Other	0	0	**<0.0001**
rs2057768-rs1805015-rs8832				
IL4R	G-A-G	43.4	12.4	
	A-A-A	22.5	23.9	
	G-A-A	12.5	22.7	
	G-G-A	7.7	27.6	
	G-G-G	7.6	7.0	
	A-A-G	4.8	2.2	
	A-G-A	0.6	3.2	
	Other	0.9	1.0	**<0.0001**
rs1805012-rs1805015-rs1801275-rs1805016				
IL4R	A-A-A-A	77.7	32.1	
	G-G-G-A	11.4	11.3	
	A-A-G-A	5.2	8.2	
	A-G-G-C	4.3	4.9	
	A-A-G-C	0.4	20.2	
	A-G-G-A	0.1	21.4	
	A-G-A-A	0	1.2	
	Other	0.9	0.7	**<0.0001**
rs1800871-rs1800896-rs1800890				
IL10	G-G-A	37.6	14.6	
	G-A-T	30.0	24.5	
	A-A-T	22.6	44.0	
	G-G-T	9.8	13.1	
	G-A-A	0	3.8	
	Other	0	0	**<0.0001**
rs1295686-rs20541				
IL13	G-G	78.3	32.0	
	A-A	21.0	23.3	
	A-G	0.6	44.7	
	Other	0	0	**<0.0001**
rs928180-rs334358-rs868				
TGFβR1	A-C-A	68.9	76.6	
	A-A-G	21.8	16.0	
	G-C-A	9.3	4.4	
	A-A-A	0	2.1	

Cytokine	Haplotype	% Caucasians	% African Americans	χ^2 p-value[a]
	Other	0	0.9	**0.0017**

[a] p-values adjusted by the Benjamini & Hochberg FDR method.