Introduction

Acute coronary syndrome (ACS) is one among several conditions of cardiovascular disease. ACS is believed to determine clearly the disease progression associated with myocardial ischemia and includes unstable angina and myocardial infarction (MI) under its ambit. Symptomatically ACS comprises a continuous intensity from non-ST segment elevation MI (NSTEMI) to ST-segment elevation MI (STEMI). The Indian origin from the Asian subcontinent population accounts for a fifth of all worldwide deaths from CHD that includes ACS and overall 7 million deaths occur as per the estimates each year together due to CAD and ACS. ACS comprising about half of the global burden is now considered to be the prime cause of deaths in the Asia-Pacific region. ACS is a multi-faceted problem that results due to genetic variations, environmental effect, dietary habits, and lifestyle. It is known that ACS often results from the compound effects of altered multiple genes where in most cases it is not clear whether ACS is inherited in a dominant or recessive manner. Currently the LDL receptor (LDLR) gene, ApoB-100 gene, ARH gene and ABCA1 gene have been identified as disease causing genes for familial hypercholesterolemia and Tangier disease, a risk factor for ACS.

The APOA1 -75 G/A and +83 C/T gene polymorphic variations are comprehensively investigated and found to be associated with other disease as well. Bi Hong et al investigated the role of APOA1 -75 G/A and +83 C/T SNPs and found APOA1 -75 A allele to have a lower risk of CAD. Also, a pilot study in North India found association of APOA1 +83 C/T in patient with MI. In fact, it is strongly believed that above APOA1 polymorphic sequence variations have an important role in a host of other diseases like Alzheimer’s, breast cancer and schizophrenia etc. The genetic nature of variability accounts for nearly 50% in the plasma HDL cholesterol concentration and among many one such sequence variants is within the APOA1 gene, where a guanine to adenine transition occurs 75 base pairs upstream from the start of transcription (-75G/A).
and other site (first intron) of the \textit{APOA1} gene results with transition of cytosine to thymine at +83bp (+83 C/T). Although ACS is the most prevalent disease throughout the world yet our population (Kashmir province, North India) is also heavily burdened with the same disease. The overall prevalence of coronary artery disease in our region as calculated by different diagnostic procedures stands at 7.54% where its frequency in males is higher 7.80% versus 6.63% in females.\textsuperscript{13} Keeping the well-recognized role of \textit{APOA1} gene in ACS in many populations of the world but no study till date has been done on \textit{APOA1} gene SNPs with respect to ACS in Kashmiri population. Thus with an increasing number of ACS cases being reported here with lack of any significant study, we designed our study with an aim to demonstrate the association between various genotypes of \textit{APOA1} -75 G/A, +83 C/T and ACS.

**Material and Methods**

The current cross-sectional case-control study was conducted at Advanced Center for Human Genetics, Sheri-I-Kashmir Institute of Medical Sciences (SKIMS), North India. A total of 240 subjects were included in the study comprising of 90 ACS patients and 150 healthy controls free from any disease who visited the hospital for general checkups. Consent information was duly sought from each ACS patient and healthy controls. Clinical parameters were recorded according to the given proforma. The inclusion criteria of ACS patients included: ACS with or without undergoing percutaneous coronary intervention, ST elevation myocardial infarction, non-ST elevation myocardial infarction, unstable angina. The exclusion criteria was history of bleeding diathesis, stroke less than 3 months platelet count<70 000/mm\textsuperscript{3}, hematocrit <30%

**Extraction of genomic DNA and polymerase chain reaction for amplification**

The blood samples of ACS patients and healthy controls were subjected to extraction of genomic DNA by common phenol-chloroform method as well as kit based DNA Extraction (Zymo Research Corporation, USA). The quality and quantity was determined by absorbance at 260nm and 280nm in a Spectrophotometer or by running on 1% agarose gel. The isolated DNA was stored at -20°C until for further analysis.

For amplification of \textit{APOA1}-75G/A (rs1799837) and +83C/T (rs5069) polymorphic regions, a 25ul reaction containing genomic DNA 250 ng/mL was used with other ingredients as 1x PCR buffer: 100 mM Tris–HCl, pH 8.3; 500 mM KCl; 15 mM MgCl\textsubscript{2}; deoxyribonucleotide triphosphate (Biotools, B & M Labs, Madrid, Spain): 10mM dATP; 10mM dCTP; 10mM dGTP; 10mM dTTP, primers (Sigma–Aldrich, USA): 5 pM in sterile deionized water and \textit{Taq DNA polymerase} 5 U/\textmu L (Biotools, Madrid, Spain). A single primer pair (Sigma–Aldrich, USA) was designed (using primer3 software) to amplify the required 433bp amplicon covering both SNPs within the \textit{APOA1} region with forward primer 5'-AGGGACAGAGCTGATCCTTGAACTCT TAAG-3' and reverse primer 5'-TTAGGGAACCTAGCCCTAGGAAGGCA-3' (reverse)\textsuperscript{16}. The thermal cycling conditions were included: as an initial denaturation at 95°C for 7 min, followed by 35 cycles at 95°C for 35 s, 63°C for 35 s and 74°C for 35 s. The final extension step was at 72°C for 10 minutes. The amplified PCR products were separated by electrophoresis on an agarose gel (1.5%) stained with ethidium bromide. The gel was visualized under ultraviolet light with a 100bp ladder.

**Polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP)**

PCR-RFLP was performed to genotype the \textit{APOA1}-75 G/ A and +83 C/T polymorphic sequence variants using 5-10 units of restriction endonuclease enzyme \textit{MspI} (NEB, New England Biolabs), to digest amplified product overnight at 37°C. The digested product was subjected to electrophoresis in a 3% agarose gel followed by ethidium bromide staining and ultraviolet illumination to identify the genotypes. \textit{MspI} restriction site at \textit{APOA1} -75 bp (G allele) and at +83 bp (C allele) digests into 209, 113, 45 and 66bp. The lack of the restriction site at-75 (A allele) and +83(T allele) displays 3 products of 209, 179, 45bp and one single product 254bp respectively (Figure 1).

For quality control and reproducibility of results, two independent researchers randomly selected 10% samples from both control and cases for genotyping.

**Statistical analysis**

IBM Statistics SPSS software (Version-23) was used for statistical evaluation. The cases and controls were compared using the Chi-square test for various categorical parameters, like gender and age of the demographic variables. A goodness-of-fit Chi-square test was used to analyze if the polymorphisms were in Hardy-Weinberg equilibrium between cases and controls. Odds ratios (OR) were used as estimates of the relative risk, and 95% confidence intervals (CI) were calculated to estimate the association between certain genotypes or other related

![Figure 1](image-url)
risk factors of ACS. P values for all analysis below the level of 0.05 are taken as statistically significant (P < 0.05).

Results

The clinicopathological characteristics of ACS patients and controls are listed in Table 1. The cases and controls were frequency matched in terms of their age, gender, and smoking status. In this study ratio of the cases, male: female was 3:1 (67 versus 23 respectively). The mean age was 60 ± 15 years for cases and 55 ± 15 years for the controls. Of the total number of cases and controls, 61(72.6%) and 115(77%) were smokers respectively. In the study groups, no significant sex-related or smoking status differences were observed between the cases and controls (P > 0.05).

In cohort group, 81.4% ACS cases had hypertension. ACS STEMI group numbered 71% as compared to 22.2% while as preMI included 19.1% versus 80.9% with no pre MI history. The demographic features of the rest of the parameters for ACS patients are recorded in Table 1.

APOA1 -75 G/A polymorphisms and ACS

In APOA1 -75 G/A, frequencies of GG, GA and AA genotypes among cases were 49 (55%), 39 (43.33%) and 02 (1.66%) while in controls as 79 (53%), 62 (41%), and 9 (6%), respectively. Three different genetic models were performed to see the relation of different genotypes among cases and controls which include additive, dominant and recessive model as shown in Table 2. In overall, the distribution of genotypes for APOA1-75G/A among cases and controls was seen almost proportionally the same in all the genetic models performed that showed no significant difference among two groups (P > 0.05; Table 2). When classified further into groups, no major difference in frequency of APOA1-75G/A genotype was found in age, gender or patients with different smoking status as against the control group (P > 0.05; Table 3).

When the patients were classified with respect to different pathologies, like hypertension, although variant genotype GA+AA was found more often in hypertensive (77.3%) than non-hypertensive (22.7%) group, yet the difference was insignificant, OR = 0.63 (C.I:0.15-2.5; P = 0.3). Despite the differences in the frequency of APOA1-75G/A genotypes among different parameters of ACS, none of them could achieve the significant association (P > 0.05) (Table 3).

APOA1 +83 C/T polymorphisms and ACS

In APOA1 +83 C/T, frequencies of CC, CT and TT genotypes observed among cases were 75 (83.33%), 13 (15%) and 02 (1.66%), while in healthy controls frequencies were 82 (55%), 50 (33%) and 18 (12%) respectively (Table 2). The distribution of APOA1 +83 C/T homozygous TT and heterozygous CT genotypes were significantly more among cases than controls as 1.6% vs. 12% for TT (P = 0.004) and 15% vs. 33% CT (P = 0.002). In case of dominant model, combined genotype CT+TT was found more often in controls 45% than cases 16.6%, with an OR=0.24 (C.I:0.11-0.53; P = 0.0001). The frequency of variant ‘T’ allele observed in cases and controls was found to be 9.4% versus 28.5% respectively with an OR=0.2 (C.I:0.12-0.50; P = 0.0001). In males, CT+TT genotype was found higher (76.6%) in controls than 81.5% in cases, yet the difference was insignificant, OR = 0.63 (C.I:0.15-2.5; P > 0.05) (Table 3).

When the patients were classified as cohort with respect to different pathologies as shown in Table 3, no significant differences were observed within different parameters with respect to variant genotypes of APOA1 +83 C/T. Haplotypic analysis was performed to demonstrate the pattern of linkage disequilibrium and few haplotypes were found with frequencies more than 5% among both cases and controls and those with <1% frequency was not considered. Table 4 shows the frequencies for the estimated 2-marker haplotypes among patients and controls. The haplotype observed in cases and controls with highest frequency as G/C haplotype that accounted for 40.0% in cases and 27.4% in controls of the SNPs studied haplotypes (APOA1-75G/A and APOA1 +83 C/T). Haplotype GT showed a marked difference when compared with wild type GC between cases and controls (P < 0.0001) as depicted in Table 4.

Discussion

Apolipoprotein A1, a protein in humans is encoded by the APOA1 gene that has a vital role in lipid metabolism.

Table 1. Characteristics of ACS patients and controls for polymorphic analysis of APOA-gene

| Demographic feature | Cases (n=90) | Controls (n=150) | P value |
|---------------------|-------------|-----------------|--------|
| Age                 |             |                 |        |
| ≤55                 | 23 (25%)    | 53 (35%)        | 0.1    |
| >55                 | 67 (75%)    | 97 (65%)        |        |
| Gender              |             |                 |        |
| Male                | 67 (75%)    | 123 (82%)       | 0.1    |
| Female              | 23 (25%)    | 27 (18%)        |        |
| Smoking status      |             |                 |        |
| Smoker              | 61 (72.6%)  | 115 (77%)       | 0.3    |
| Non-smoker          | 23 (27.3%)  | 35 (23%)        |        |
| *Hypertension       |             |                 |        |
| Hypertensive        | 66 (61.4%)  | 15 (15.2%)      |        |
| Non-hypertensive    | 33 (30.7%)  | 50 (60.2%)      |        |
| *Diabetes mellitus  |             |                 |        |
| Diabetic            | 56 (62.2%)  |                |        |
| Non-diabetic        | 34 (37.8%)  |                |        |
| *PCI                |             |                 |        |
| Yes                 | 62 (83.7%)  | 12 (16.2%)      |        |
| No                  | 27 (26.3%)  | 98 (84%)        |        |
| *ACS                |             |                 |        |
| STEMI               | 64 (71%)    |                |        |
| NSTEMI              | 20 (22.2%)  |                |        |
| USA                 | 5 (6.6%)    |                |        |
| *Pre-MI             |             |                 |        |
| Yes                 | 17 (19.1%)  |                |        |
| No                  | 53 (60.9%)  |                |        |

Abbreviations: PCI, percutaneous coronary intervention; ACS, acute coronary syndrome; STEMI, ST-elevation myocardial infarction; NSTEMI, non-ST elevation myocardial infarction; USA, unstable angina; Pre-MI, previous myocardial infarction

* Description of clinicopathological parameters in cases (patients)
### Table 2: Overall distribution of genotypes/allele APOA1-75 and +83 frequencies in cases and controls

| Overall Genotyping | APOA1 genotype | Cases (%) N=90 | Controls (%) N=150 | OR (95% CI) | P value |
|--------------------|----------------|---------------|--------------------|-------------|---------|
| **APOA1-75G/A (rs1799837)** | -75GG | 49(55.0) | 79(53.0) | 1.0(0.5-1.9) | 0.5 |
| | -75GA | 39(43.3) | 62(41.0) | | |
| | -75AA | 2(1.6) | 9(6.0) | 0.2(0.03-2.3) | 0.1 |
| **Dominant Model** | GG | 49(55.0) | 79(53.0) | Reference | |
| | GA+AA | 41(45.0) | 71(47.0) | 0.9(0.4-1.7) | 0.4 |
| **Recessive Model** | AA | 2(1.6) | 9(6.0) | Reference | |
| | GA+GG | 88(98.3) | 141(94.0) | 3.7(0.4-32.0) | 0.1 |
| **Additive Model** | GG | 49(55.0) | 79(53.0) | Reference | |
| | AA | 2(1.6) | 9(6.0) | 0.2(0.03-2.3) | 0.1 |
| **Allele frequency** | -75 G | 138(76.6) | 220(73.5) | Reference | |
| | -75 A | 42(23.3) | 80(26.5) | 0.8(0.4-1.4) | 0.3 |
| **APOA1+83C/T (rs5069)** | +83CC | 75(83.3) | 82(55.0) | Reference | |
| | +83CT | 13(15) | 50(33.0) | 0.3(0.1-0.6) | 0.002 |
| | +83TT | 2(1.6) | 18(12.0) | 0.09(0.01-0.7) | 0.004 |
| **Dominant Model** | CC | 75(83.3) | 82(55.0) | Reference | 0.0001 |
| | CT+TT | 15(16.6) | 65(45.0) | 0.24(0.1-0.5) | 0.01 |
| **Recessive Model** | TT | 2(1.6) | 18(12.0) | Reference | |
| | CT+CC | 88(98.3) | 132(88.0) | 8.04(1.0-63.5) | 0.004 |
| **Additive Model** | CC | 75(97.4) | 82(82.0) | Reference | 0.0001 |
| | TT | 2(2.6) | 18(18.0) | 0.09(0.01-0.7) | 0.3 |
| **Allele frequency** | +83 C | 163(90.5) | 214(71.5) | Reference | |
| | +83 T | 17(9.4) | 86(28.5) | 0.25(1.0-5.0) | 0.0001 |

Abbreviations: GG: wild; GA: heterozygous; AA: homozygous variant. CC: wild; CT: heterozygous; TT: homozygous variant.

* Dominant model: GG, ** Recessive model: AA, ***Dominant model: CC, **** Recessive model: TT

### Table 3: Clinical-epidemiological variables of CAD patients versus the polymorphic phenotypes of the APO-A1 gene

| Parameter | *Cases APOA1-75 G/A | Control | OR (95% CI) | P value |
|-----------|---------------------|---------|-------------|---------|
| Age ≤55 | 12(24.3) | 24(29.5) | 0.4(0.1-1.5) | 0.1 |
| >55 | 7.5(75.7) | 30(74) | 1.0(0.4-2.2) | 0.5 |
| Gender Male | 42(69.6) | 33(81.5) | 1.0(0.48-2.1) | 0.5 |
| Female | 15(30) | 33(81.5) | 1.0(0.48-2.1) | 0.5 |
| Smoking Status Non-Smoker | 16(30.3) | 21(82.7) | 0.69(0.2-2.8) | 0.4 |
| Smoker | 20(80) | 4(16.7) | 0.69(0.2-2.8) | 0.4 |
| HTN Yes | 40(84.4) | 25(77.3) | 0.6(0.15-2.5) | 0.3 |
| No | 11(15.6) | 8(22.7) | 0.6(0.15-2.5) | 0.3 |
| DM Yes | 33(66.7) | 16(50) | 2.07(6.04) | 0.1 |
| No | 17(33.3) | 16(50) | 2.07(6.04) | 0.1 |
| PCI Yes | 36(88.8) | 25(77.3) | 0.4(0.08-2.0) | 0.2 |
| No | 5(11) | 8(22.7) | 0.4(0.08-2.0) | 0.2 |
| NSTEMI Yes | 10(20.8) | 9(21.4) | Ref | |
| No | 33(66.7) | 31(73.8) | Ref | |
| ACS STEMI Yes | 33(66.7) | 31(73.8) | 1.1(0.3-3.8) | 0.5 |
| No | 5(10.41) | 2(4.7) | 2.8(0.2-29.7) | 0.3 |
| USA Yes | 5(10.41) | 2(4.7) | 2.8(0.2-29.7) | 0.3 |
| No | 33(66.7) | 31(73.8) | 1.1(0.3-3.8) | 0.5 |
| PREMI Yes | 12(16) | 7(17.07) | 0.44(0.15-1.2) | 0.1 |
| No | 63(84) | 34(82.9) | 0.44(0.15-1.2) | 0.1 |

Abbreviations: HTN, hypertension; DM, diabetes mellitus; PCI, percutaneous coronary intervention; STEMI, ST-elevation myocardial infarction; NSTEMI, non-ST elevation myocardial infarction; USA, unstable angina; pre-MI, previous myocardial infarction

*Association of APOA1-75 G/A wild type (GG) and variant genotype (GA+AA) in demographic/pathological features between cases and controls. **Association of APOA1 +83 C/T wild type (GG) and variant genotype (CT+TT) in demographic/pathological features between cases and controls
Association of APOA1 polymorphic variation with acute coronary syndrome

APOA1 is a major protein ingredient of HDL and a relatively abundant plasma protein. In the APOA1 gene -75 G/A and +83C/T, play an important role in lipid metabolism. Regarding ACS, the studies have been very limited to reach the conclusive remark. A cross sectional case-control study was conducted to observe the role of APOA1 gene -75 G/A and +83C/T variations with respect to ACS in our population (Kashmir province, North India). In our report, APOA1 -75 G/A, the frequency of three genotypes of APOA1-75 G/A SNP among ACS patients and the controls revealed no significant differences. Similar scenario has been published by Yan Ding et al., that reported lack of association for APOA1-75 G/A in ACS. Further, APOA1 -75 G/A and ACS risk was not proved in European population with 98.6%, 1.3% and 0.1% genotypic frequency in cases and 99.2%, 0.8% and 0% in controls. It is worthwhile to mention that the frequency of APOA1-75 G/A varies considerably among different ethnic regions as can be seen among three studies which show quite different genotypic differences in both cases and controls. In our study frequency of variant A allele APOA1 -75 G/A showed no association which is supported by similar scenario reported by Yan et al. On the contrary, a study in Australian population, found the presence of the variant A allele increased the severity of ACS, a finding that is in stark contrast to our study and one more report carried earlier.

In this study, combined genotype (GA+AA) was found comparatively higher in smokers of control group (controls: 82.9% versus cases 78.3%). This is in discordance with the study by Morgan et al. Also, the combined genotype (GA+AA) was found more often in hypertensive in accordance with the study performed in the Chinese population. Interestingly, in contrast to our study, Morgan et al. has shown an association between the APOA1-75G/A polymorphism and diabetes mellitus (DM). The possible reasons for this contradiction are unclear though these could be due to different geographical regions. It must be stressed that even among populations of similar ethnicity and gene pool, environmental factors such as diet, stress and physical inactivity compound to an individual’s composite risk for ACS in inexplicable proportions.

In our study, APOA1 +83C/T showed a protective role with respect to the development of ACS where the frequencies of different genotypes showed significant differences between cases and controls. The frequency of heterozygous variant genotype TT allele was observed significantly higher in controls than cases (12% versus 1.6% respectively). This clearly demonstrates a significant association of APOA1 +83 C/T variant genotype with ACS in our population. APOA1 +83 variant T allele was found to be higher in controls (0.34 versus 0.12) than cases and clearly demonstrates that T allele has uniquely a protective role for ACS in our population. In contrast to our findings, Morgan et al. found no association between ACS and APOA1 +83 C/T gene polymorphism. The proposed protective role of APOA1 +83 variant T allele as found in our study is substantiated by an investigation by Wang et al. who found T allele associated with increased levels of HDL and APOA1, an evidence that suggests protective role over ACS. One of the possible explanations for this transition is that APOA1 +83 reside in the 5’ region of the APOA1 gene. The MspI site at APOA1+83bp embedded with CpG island is known to be hypermethylated in non-expressing cells but demethylated in cells expressing APOA1. It is possible that T substitution at this site may lead to further demethylation, resulting in more cells expressing the gene.

Further, in our study APOA1 +83 C/T variations was associated with ACS in men. This is partially in accordance with an earlier study that found no association in men and women. Our study found no significant association of ACS was found with hypertension and diabetes. Our results are in accordance with another study conducted earlier.

On the basis of cardiovascular events pre-MI, STEMI, NSTEMI and USA, no association with APOA1 +83 C/T was found in contradiction with the study by Morgan et al. Discrepancies in the findings of these studies with our report could be attributed to differences in the genetic susceptibility between different ethnic groups. Also, the APOA1 gene locus resides in a cluster with CIII and AIV loci, which could attribute to linkage disequilibrium with different APOA1 alleles in some but not in other populations.

The putative role of APOA1 -75 G/A and +83 C/T has been studied in various diseases across the globe where prominent diseases that have been compiled in this study to highlight the implication and role of this sequence variation include hypertension, renal cancer, Alzheimer’s disease, CAD, and all the diseases have shown distinct but varied associations. This augments for the need to study the role of APOA1 in more diseases.

Further, the effect of a single polymorphic nucleotide variation is questionable to lead to an outcome in the study of multifaceted diseases. Therefore, the amalgamation of different genetic variants in the set of connections of same loci strengthens their efficiency to improve the analytical influence for complex diseases. To demonstrate whether APOA1 -75 G/A and +83 C/T polymorphic variants could translate a synergistic effect...
in the development or progression of ACS, we looked at their collective impact to examine the inclination to the disease. The most frequently implicated haplotype was GT variant haplotype that showed a significant protective impact on ACS as compared to wild type haplotype GC. Our study showed the haplotypes at two restriction sites as more informative and demonstrated a significant association together to confer a protective role in ACS. In consistency with our data, there a few reports that have shown more or less the same trend, implicating that combining both polymorphic sites of APOA1 −75 G/A and +83 C/T sites as more informative and significantly portray the combined effect with almost twice the amount of phenotypic variation in plasma APOA1 compared to single RFLP site. This study shows that haplotype analysis in the APOA1 gene is very important to display its functional significance in ACS.

Conclusion
The study concludes that APOA1 -75G/A has no association with ACS but its other related polymorphic variant T allele and TT genotype of +83 C/T was observed to confer a protective effect with respect to the same condition. Our report further implies that a particular haplotype (GT) of APOA1 gene is highly implicated and possibly can act as low penetrance genotypes in the predilection to ACS. These findings are subject to be validated in large sample cohort studies to decide the course of ACS management at molecular perspective.

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Competing interest
All authors declare no competing interest.

Ethical approval
The procedures done in the study involving human participants were in agreement with the ethical standards of the institutional and/ or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Ethical approval for this study was obtained from Institutional Ethical Review Committee (IEC SKIMS Study ref: 1904/2014)

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References
1. Mendis S, Puska P, Norrving B. World Health Organization (WHO). Global Atlas on Cardiovascular Disease Prevention and Control. Geneva: WHO; 2011.
2. Lippi G, Sanchis-Gomar F, Cervellin G. Chest pain, dyspnea and other symptoms in patients with type 1 and 2 myocardial infarction. A literature review. Int J Cardiol. 2016;215:20-22. doi:10.1016/j.ijcard.2016.04.045
3. World Health Organization (WHO). Cardiovascular Diseases (CVDs) Fact Sheet. WHO; 2017.
4. Ohira T, Iso H. Cardiovascular disease epidemiology in Asia: an overview. Circ J. 2013;77(7):1646-1652. doi:10.1253/circj.cj-13-0702
5. Hobbs HH, Russell DW, Brown MS, Goldstein JL. The LDL receptor locus in familial hypercholesterolemia: mutational analysis of a membrane protein. Annu Rev Genet. 1990;24:133-170. doi:10.1146/annurev.ge.24.120190.001025
6. Lund-Katz S, Laplaud PM, Phillips MC, Chapman MJ. Apolipoprotein B-100 conformation and particle surface charge in human LDL subspecies: implication for LDL receptor interaction. Biochemistry. 1998;37(37):12867-12874. doi:10.1021/bi980828m
7. Abifadel M, Varret M, Rabès JP, Allard D, Ouguerram K, Devillers M, et al. Mutations in PCSK9 cause autosomal dominant hypercholesterolemia. Nat Genet. 2003;34(2):154-156. doi:10.1038/ng1161
8. Ouguerram K, Chetiveaux M, Zair Y, Costet P, Abifadel M, Varret M, et al. Apolipoprotein B100 metabolism in autosomal-dominant hypercholesterolemia related to mutations in PCSK9. Arterioscler Thromb Vasc Biol. 2004;24(8):1448-1453. doi:10.1161/01.vas.0000133684.77013.88
9. Pullinger CR, Eng C, Salen G, Shefer S, Battä AK, Erickson SK, et al. Human cholesterol 7alpha-hydroxylase (CYP7A1) deficiency has a hypercholesterolemic phenotype. J Clin Invest. 2002;110(1):109-117. doi:10.1172/jci15387
10. García CK, Wilund K, Arca M, Zuliani G, Fellin R, Maioli M, et al. Autosomal recessive hypercholesterolemia caused by mutations in a putative LDL receptor adaptor protein. Science. 2001;292(5520):1394-1398. doi:10.1126/science.1060458
11. Bodzioch M, Orsó E, Klucken J, Langmann T, Böttcher A, Diederich W, et al. The gene encoding ATP-binding cassette transporter 1 is mutated in Tangier disease. Nat Genet. 1999;22(4):347-351. doi:10.1038/13914
12. Brooks-Wilson A, Marcił M, Clee SM, Zhang LH, Roomp K, van Dam M, et al. Mutations in ABC1 in familial hypercholesterolemia and familial high-density lipoprotein deficiency. Nat Genet. 1999;22(4):336-345. doi:10.1038/11905
13. Liao B, Cheng K, Liu H, Xu Z. Effect of apolipoprotein A1 genetic polymorphisms on lipid profiles and the risk of coronary artery disease. Diagn Pathol. 2015;10:102. doi:10.1186/s13000-015-0328-7
14. Dawar R, Gurtoo A, Singh R. Apolipoprotein A1 gene polymorphism (G-75A and C+83T) in patients with myocardial infarction: a pilot study in a north Indian population. Am J Clin Pathol. 2010;134(2):249-255. doi:10.1093/ajcp/kxp031
15. Kamili M, Dar I, Ali G, Wazir H, Hussain S. Prevalence of coronary heart disease in Kashmiris. Indian Heart J.
Association of APOA1 polymorphic variation with acute coronary syndrome

2007;59(1):44-49.

16. Liu Z, Xiao Y, Tang L, Jiang L, Wang Y, Zhang R, et al. Apolipoprotein A1 -75 G/A and +83 C/T polymorphisms and renal cancer risk. Lipids Health Dis. 2015;14:143. doi:10.1186/s12944-015-0132-0

17. Breslow JL. Apolipoprotein genetic variation and human disease. Physiol Rev. 1988;68(1):85-132. doi:10.1152/physrev.1988.68.1.85

18. Saha N, Tay JS, Low PS, Humphries SE. Guanidine to adenine (G/A) substitution in the promoter region of the apolipoprotein A1 gene is associated with elevated serum apolipoprotein A1 levels in Chinese non-smokers. Genet Epidemiol. 1994;11(3):255-264. doi:10.1002/gepi.1370110304

19. Wang XL, Badenhop R, Humphrey KE, Wilcken DE. NewMspI polymorphism at +83 bp of the human apolipoprotein A1 gene: association with increased circulating high density lipoprotein cholesterol levels. Genet Epidemiol. 1996;13(1):1-10. doi:10.1002/(sici)1098-2272(1996)13:1<1::aid-gepi1>3.0.co;2-d

20. Dawar R, Gurtoo A, Singh R. Apolipoprotein A1 gene polymorphism (G-75A and C+83T) in patients with myocardial infarction: a pilot study in a north Indian population. Am J Clin Pathol. 2010;134(2):249-255. doi:10.1309/ajcptsxq3q1fg

21. Ding Y, Zhu MA, Wang ZX, Zhu J, Feng JB, Li DS. Associations of polymorphisms in the apolipoprotein APOA1-C3-A5 gene cluster with acute coronary syndrome. J Biomed Biotechnol. 2012;2012:509420. doi:10.1155/2012/509420

22. Vollbach H, Heun R, Morris CM, Edwardsdon JA, McKeith IG, Jessen F, et al. APOA1 polymorphism influences risk for early-onset nonfamiliar AD. Ann Neurol. 2005;58(3):436-441. doi:10.1002/ana.20593

23. Morgan TM, Krumholz HM, Lifon RP, Spertus JA. Nonvalidation of reported genetic risk factors for acute coronary syndrome in a large-scale replication study. JAMA. 2007;297(14):1551-1561. doi:10.1001/jama.297.14.1551

24. Sinha R, Singh R. Role of apolipoprotein A1 gene polymorphism (G-75A and C+83T) in essential hypertension in Indian population. Ann Clin Lab Sci. 2014;44(3):298-303.

25. Kadane JB, Lazar NA. Methods and criteria for model selection. J Am Stat Assoc. 2004;99(465):279-290. doi:10.1198/016214504000000269

26. Beuten J, Gelfond JA, Franke JL, Weldon KS, Crandall AC, Johnson-Pais TL, et al. Single and multigenic analysis of the association between variants in 12 steroid hormone metabolism genes and risk of prostate cancer. Cancer Epidemiol Biomarkers Prev. 2009;18(6):1869-1880. doi:10.1158/1055-9965.epi-09-0076

27. Kamboh MI, Aston CE, Nestlerode CM, McAllister AE, Hamman RF. Haplotype analysis of two APOA1/MspI polymorphisms in relation to plasma levels of apo A-I and HDL-cholesterol. Atherosclerosis. 1996;127(2):255-262. doi:10.1016/s0021-9150(96)05966-7

28. Yin RX, Li YY, Lai CQ. Apolipoprotein A1/C3/A5 haplotypes and serum lipid levels. Lipids Health Dis. 2011;10(1):140. doi:10.1186/1476-511x-10-140