Assessment of genetic polymorphism associated with ATP-binding cassette transporter A1 (ABCA1) gene and fluctuations in serum lipid profile levels in patients with coronary artery disease

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A B S T R A C T

Background: Coronary artery disease (CAD) is one of the common genetic and clinical risk factors associated with cardiovascular and multifactorial disorder. ATP-binding cassette transporter A1 (ABCA1) gene plays an important role in lipid metabolism and in multiple studies associated with CAD. However, more studies are needed to identify the exact role of single nucleotide polymorphisms which may cause CAD.

Objectives: The aim of this study is to investigate the genetic association of polymorphism g.1051G > A in the ABCA1 gene with CAD patients in the Saudi population.

Methods: We included 315 confirmed CAD cases, and 205 non-CAD or control subjects in this case-control study. DNA isolation was carried out for all registered participants and the polymorphism g.1051G > A was genotyped with Polymerase Chain Reaction followed by Restriction Fragment Length Polymorphism analysis with EcoNI restriction enzyme.

Results: Modifiable risk factors such as Body Mass Index, smoking and diabetes were strongly associated and non-modifiable risk factors such as hypertension (Systolic Blood Pressure and Diastolic Blood Pressure) and serum analysis such as Fasting Blood Glucose, Total cholesterol (TC), Triglyceride (TG) and LDL-c were significantly associated in CAD cases (p < 0.05). Allele (OR=1.73;95% CI:1.33–2.26; p = 0.0004), GA vs GG (OR=2.26; 95% CI: 1.53–3.35; p = 0.0003 and dominant inheritance pattern (OR=2.23; 95% CI:1.56–3.20; p = 0.00009 was strongly associated with CAD cases and control subjects. The

Abbreviations: CAD, Coronary artery disease; ABCA1, ATP-binding cassette transporter A1; PCR, Polymerase Chain Reaction.

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1. Introduction

Coronary artery disease (CAD) is commonly known to cause premature mortality worldwide (Alshammary et al., 2021). The CAD prevalence rate is extremely high despite all of the precautionary methods that have been provided to the public. In 2015, CAD was responsible for 17.9 million of all death occurrences globally (Ruan et al., 2018). It is well known that the outcomes of congestive heart failure caused by CAD are worse than those caused by Acquired Immune Deficiency Syndrome (AIDS) or cancer (McMurray and Stewart, 2000). There are several factors that could cause CAD, including family history, lifestyle and the environment. Other risk factors include but are not limited to hypertension, diabetes, obesity and high cholesterol (McMurray and Stewart, 2000).

Previous genetic studies showed that most genetic variants that are associated with CAD were located close to specific genes. These genes have various roles in regulating the metabolism of triglyceride-rich lipoproteins (TRLs) and low-density lipoproteins (LDLs), thus underlining their roles in causing CAD (Toth, 2016). In addition, a whole exome sequencing study investigated mutations in 9 genes that were previously linked to CAD occurrence. The results from this study revealed destructive mutations in the LDLR gene, which were associated with an increased incidence of CAD (Do et al., 2015). Recently, several genome-wide association studies (GWAS) have linked numerous SNPs in non-coding regions of various genes with CAD occurrence. Although it was proposed that these SNPs could cause CAD through the stimulation of atherosclerosis, the exact mechanisms require further investigation (Christiansen et al., 2017). To date, GWAS have identified 161 genetic loci that are related to the causation of CAD. The genetic locus chr9p21.3 has the strongest effect as a risk factor of CAD development. This locus has multifarious SNPs that can be found in the CDKN2B-AS1 locus of the non-coding RNA. SNPs in this locus were also linked to other disorders, such as atherosclerosis and ischaemic stroke (Giral et al., 2018). A study that was performed on European and Non-European subjects documented a number of genetic loci that could lead to CAD. These loci have SNPs in an abundant number of CAD genes, including but not limited to BRAP, ATP2B1, PPAP2B, APOB and ABCG8 (McPherson, 2014). Nevertheless, a network analytical study has prioritized CAD candidate genes through bioinformatics tools. This study revealed the likelihood of 481 genes causing CAD based on their top scores in GWAS significance and included HLA-DQA1, SAYS1D, EXOC6B, LRP1, NDUFA4L2, SHMT2 and CRABP1.

Recent studies have focused on the effects of various mutations in the ABCA1 gene, which is also known as ATP-binding-cassette-transporter-A1, facilitates the transfer of lipid-derived molecules across the cell membrane. This mechanism is essential in maintaining preventive measures against atherosclerosis and ensuring cellular cholesterol homeostasis (Kyriakou et al., 2007). Previous studies have suggested a link between elevated levels of triglycerides and low levels of HDL cholesterol, which could lead to CAD. Although single nucleotide polymorphisms (SNPs) that target the coding sequence in ABCA1 could affect the severity of CAD, SNPs in the non-coding sequence could disrupt ABCA1 expression.

Conclusion: The polymorphism g.1051G > A in the gene ABCA1 is closely associated with the existence of the CAD subjects. This polymorphism could also affect the serum levels of the lipid profile, suggesting a possible occurrence of CAD in the Saudi population.

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Scientific (Dreieich, Germany) and stored at – 20 °C (Khan et al., 2019). The extracted DNA was subjected to molecular genotyping using the restriction fragment length polymorphism technique (RFLP) for the ABCA1 gene c.g.1051G > A variant. The primer sequence was as follows: forward: 5’-GTA TTT TTG CAA GGC TAC CAG TTA CAT TTG ACA A-3’, and reverse: 5’-GAT TGG CTT CAG GAT GTC CAT GTT GGA A-3’. The primers were adopted from previously published article (Clee et al., 2001) and from local manufacturer, primers were synthesized (lg Biosystems, Saudi Arabia). The amplification was carried out using a Veriti thermal cycler from Life Technologies (Darmstadt, Germany) with Promega GoTaq Master Mix (Walldorf, Germany) with the following programming conditions: 95 °C for 5 min, followed by 37 cycles of 95 °C for 30 s, 47 °C for 30 s, and 72 °C for 1 min, and a final extension step at 72 °C for 10 min. The selected primers produced a DNA fragment of 177 base pairs. Subsequently, 3 U of EcoNI (CCTACCAA/GGAG) restriction endonuclease from New England BioLabs (Frankfurt am Main, Germany) was added to 20 µl of amplified products to a total volume of 25 µl and incubated at 37 °C for overnight. Electrophoretic separation was carried out for the digestion products on a 3% agarose gel with ethidium bromide, and the bands were then visualized using an ultraviolet light cabinet (Rao et al., 2015). DNA fragments of 177 bp, 107 bp and 70 bp were released. There was a single 170-bp product for GG homozygotes; 3 bands for GA heterozygotes of 170 bp, 107 bp and 70 bp; and 2 bands for AA homozygotes of 107 bp and 70 bp (Fig. 1). Validation was performed with Sanger sequencing analysis (Fig. 2) based on our recent publication (Alharbi et al., 2021).

2.5. Statistical considerations and analysis

Data were analyzed using statistical software SPSS 21.0 (IBM Inc., Chicago, United States). The interpretation of the categorical variables was using descriptive statistics (frequencies and percentages). The Chi-square ($\chi^2$) test of Pearson was used to compare CAD cases and controls as well as to distribute categorical study variables across the three genotypes of the g.1051G > A (GG, GA & AA) ABCA1 gene. The $\chi^2$ test is commonly applied for testing the relationships between the categorical variables and the null hypothesis of $\chi^2$ test is that no relationship exists on the categorical variables in the independent population. All the frequencies were counted with the method of gene counting and the $\chi^2$ test was applied to validate the departures from the equilibrium Hardy-Weinberg (HWE). The genotype distribution of g.1051G > A polymorphism has been contrasted by the $\chi^2$ test using openepi software between CAD cases and subject controls (Khan et al., 2014). Two-sided experiments aimed at statistical significance. The bonferroni correction has been applied to the polymorphism g.1051G > A for different inheritance modes and different genotypes (Alharbi et al., 2014). A non-parametric Kruskal-Wallis test was used to compare the mean ranges of skewed quantitative variables with the three ABCA1 gene products g.1051G > A (GG, GA, and AA). A p-value of < 0.05 has been used to describe the results of significant correlations.

3. Results

3.1. Clinical data

Table 1 shows the basic demographic characteristics of CAD cases and subjects of control. This study was conceived as a balanced sample of age and gender which indicates that both of them are not significantly associated (p > 0.05). CAD appears to be associated more often with BMI, smoking, diabetes, HTN, SBP, and DBP compared to non-CAD cases/healthy controls. Serum samples include fasting blood glucose, positive association of lipid profile parameters of TC, TG and LDL-c (p < 0.05). In CAD cases, HDL-c is not related and showed non-significant association when compared with controls (p > 0.05).

3.2. Genetic analysis of g.1051G > A genotyping

In the investigated CAD cases the genotypical distribution was consistent with HWE (p = 0.5461). The genotypical distribution of the g.1051G > A polymorphism between CAD cases and control subjects was shown in Table 2. The frequencies of the genotypes
GG, GA and AA were 35.9%, 46.3% and 17.8% in CAD cases and 55.6%, 31.7% and 12.7% in control subjects. Allele (OR-1.73; 95% CI: 1.33–2.26; p = 0.0004) and various genotype frequencies (AA vs GG: OR-2.17; 95% CI: 1.27–3.70; p = 0.003, GA + AA vs GG: OR-2.23; 95% CI: 1.56–3.20; p = 0.00009) were significantly associated within the cases and controls. After adjusting the age, BMI and sex between the CAD cases and controls, bonferroni correction was calculated and found a significant association between genotypes (GA vs GG: OR-2.5; 95% CI: 1.8–3.5; p = 0.00006 & GA vs AA + GG: OR-2.0; 95% CI: 1.5–2.9; p = 0.0003). The measurement of sample and power size in g.1051G > A polymorphism was found to be 56%. The interaction was picked into the R219 K polymorphism after modification of the potential confounding factors.

3.3. Comparison of the use of medicines

The use of different medicines among these CAD subjects was observed, and its frequency of use was compared across the three products of the ABCA1 gene c.g.1051G > A. Out of 9 different medicines, the frequency of use of two medicines (atorvastatin and nitroglycerin) was significantly different among the subjects who had GG, GA, and AA. That is, 65 out of 91 (71.4%) with GG and 94 out of 117 (80.3%) with GA were using atorvastatin when compared with subjects with AA in which 28 out of 47 (59.5%) were using it, which was statistically significant (p = 0.022). Additionally, 9 out of 117 (7.7%) subjects with GA were using nitroglycerin when compared with subjects with GG (1 out of 91) and subjects with AA (1 out of 47), which was statistically significant (p = 0.048) (Table 3).

3.4. Comparison of the distribution and mean rank values

The plasma levels of LDL, cholesterol, HDL and triglycerides were compared across the three genotypes of the ABCA1 gene g.1051G > A in which a statistically significant difference was observed for the categories of cholesterol. That is, the proportions of borderline high cholesterol (52.4%) and high cholesterol (81.8%) were higher in persons with GA and GG than those with AA (p = 0.023). No statistical significance was observed for the other 3 variables across the three genotypes of the ABCA1 gene g.1051G > A (Table 4). The mean ranks of all clinical variables

| Table 1 | Clinical characteristics between control subjects and CAD cases. |
|---------|---------------------------------------------------------------|
| Variables | CAD cases (n = 315) | Controls (n = 205) | P-values |
| Age (Mean ± SD) | 59.7 (10.87) | 58.4 (9.28) | p = 0.157 |
| BMI (Mean ± SD) | 29.26 (6.49) | 26.26 (6.49) | p = 0.002 |
| Gender (Male%) | 206 (65.4%) | 119 (58%) | p = 0.055 |
| Smoking (%) | 150 (47.6%) | 58 (28.3%) | p = 0.001 |
| Exercise (%) | 72 (22.9%) | 67 (32.7%) | p = 0.009 |
| Diabetes (%) | 187 (59.4%) | 35 (17.1%) | p = 0.001 |
| Fasting glucose (mg/dl) | 140.64 | 83.56 | p = 0.001 |
| Hypertension (%) | 221 (70.2%) | 41 (20%) | p = 0.001 |
| Systolic BP (mmHg) | 146.47 | 122.03 | p = 0.002 |
| DBP (mmHg) | 91.84 | 74.25 | p = 0.003 |
| Event Stroke (%) | 72 (22.9%) | 7 (3.4%) | p < 0.001 |
| Total Cholesterol (mg/dl) | 154.17 (23.6) | 140.88 (18.09) | p = 0.003 |
| HDL-c (mg/dl) | 39.95 (8.05) | 41.14 (8.65) | p = 0.111 |
| LDL-c (mg/dl) | 115.80 (38.28) | 88.6 (30.18) | p = 0.001 |
| Triglycerides (mg/dl) | 145.97 (60.27) | 120.12 (22.71) | p = 0.001 |

Table 2 | g.1051G > A polymorphic genotype and allele frequencies in the ABCA1 gene in CAD cases and control subjects and Bonferroni correction. |
|-----------------|---------------------------|---------------------|-------------------|-----------------|-----------------|-------------------|
| Genotypes | CAD Cases (n = 315) | Controls (n = 205) | OR (95% CI)<sup>a</sup> | P-Value | OR (95% CI)<sup>b</sup> | P-Value |
| GG | 113 (35.9%) | 114 (55.6%) | Reference | Reference | 1.0 | 1.0 |
| GA | 146 (46.3%) | 65 (31.7%) | OR-2.26 (95% CI: 1.53–3.35) | 0.0003 | OR-2.2 (95% CI: 1.4–3.3) | 0.0001 |
| AA | 56 (17.8%) | 26 (12.7%) | OR-2.17 (95% CI: 1.27–3.70) | 0.003 | OR-2.4 (95% CI: 1.5–4.0) | 0.001 |
| GA + AA vs GG | 202 (64.1%) | 91 (44.4%) | OR-2.23 (95% CI: 1.56–3.20) | 0.00009 | OR-2.5 (95% CI: 1.8–3.5) | 0.00006 |
| GA vs GG + AA | 146 (46.3%) | 65 (31.7%) | OR-1.86 (95% CI: 1.28–2.68) | 0.0008 | OR-2.0 (95% CI: 1.5–2.9) | 0.0003 |
| AA vs GG + GA | 56 (17.8%) | 26 (12.7%) | OR-1.48 (95% CI: 0.90–2.46) | 0.11 | OR-1.6 (95% CI: 1.1–2.7) | 0.08 |
| G allele | 372 (0.59) | 293 (0.71) | Reference | Reference | – | – |
| A allele | 258 (0.41) | 117 (0.29) | OR-1.73 (95% CI: 1.33–2.26) | 0.0004 | – | – |

<sup>a</sup>Indicates 95% CI with crude odds ratio and <sup>b</sup>indicates Bonferroni correction adjusted with age, BMI and gender.
ABCA1 gene

Comparison of the mean rank values of clinical variables across three genotypes of the ABCA1 gene

Comparison of the distribution of clinical variables across three genotypes of the ABCA1 gene

Comparison of the use of medicines across three products of the ABCA1 gene

(p = 0.047) (Table 5).

GA had higher TG values when compared with those with AA observed for TG, which indicates that the persons with GG and g.1051G > A, where a statistically significant difference was

ABCA1 gene compared among the three genotypes of the

Clinical variables were correlated with genotype levels in CAD patients. There were no significant differences in sex and age between CAD cases and controls as this study was chosen for similar age and genders

Table 3
Comparison of the use of medicines across three products of the ABCA1 gene c.g.1051G > A in CAD study subjects.

| Name of medicines | ABCA1 gene c.g.1051G > A | X²-value | p-value |
|-------------------|--------------------------|----------|---------|
|                   | GG | GA | AA |          |          |
| Amiodipine (yes/no)n = 110;143;54 | 23/87 | 26/1117 | 9/45 | 0.514 | 0.774 |
| Enoxaparinclexame(Yes/no)n = 111;143;54 | 14/96 | 16/127 | 7/47 | 0.190 | 0.909 |
| Aspirin (yes/no)N = 91;117;47 | 69/22 | 92/25 | 31/16 | 2.917 | 0.233 |
| Atorvastatin (yes/no)n = 91;117;47 | 65/26 | 94/23 | 28/19 | 7.657 | 0.022* |
| Rosuvastatin (yes/no)n = 91;117;47 | 7/84 | 2/115 | 3/44 | 4.447 | 0.108 |
| Clopidogrel (yes/no)n = 91;117;47 | 43/48 | 60/57 | 23/24 | 0.338 | 0.845 |
| Pantoprazole (yes/no)n = 91;117;47 | 48/43 | 67/50 | 24/23 | 0.697 | 0.706 |
| Nitroglycerin (yes/no)n = 91;117;47 | 1/90 | 9/108 | 1/46 | 6.058 | 0.048* |
| Perindopril arginine (yes/no)n = 91;117;47 | 33/58 | 47/70 | 21/26 | 0.947 | 0.623 |

*Statistically significant.

Table 4
Comparison of the distribution of clinical variables across three genotypes of the ABCA1 gene c.g.1051G > A in CAD study persons

| Classification based on biochemical variables | ABCA1 gene c.g.1051G > A | X²-value | p-value |
|-----------------------------------------------|--------------------------|----------|---------|
|                   | GG | GA | AA |          |          |
| LDL-C (mg/dl) | 70(36.3) | 91(47.2) | 32(16.6) |          |          |
| Optimum (>100) | 19(29.7) | 34(51.1) | 11(17.2) |          |          |
| Near optimum (100–129) | 13(32.5) | 17(42.5) | 10(25.0) | 9.130 | 0.166 |
| Borderline high (130–159) | 11(64.7) | 4(23.5) | 2(11.8) |          |          |
| High (>160) |          |          |          |          |          |
| T. Chol (mg/dl) | 90(34.5) | 122(46.7) | 49(18.8) |          |          |
| Optimum (>200) | 14(33.3) | 22(52.4) | 6(14.3) | 11.292 | 0.023* |
| Near optimum (200–239) | 9(21.8) | 2(18.2) | 0(0) |          |          |
| Borderline high (240–440) |          |          |          |          |          |
| HDL-C (mg/dl) | 70(33.3) | 93(45.6) | 41(20.1) |          |          |
| Major risk factor low (<40) | 38(40.0) | 46(48.4) | 11(11.6) | 3.466 | 0.483 |
| Moderate (40–60) | 9(33.3) | 7(46.7) | 3(20.0) |          |          |
| Protective (<60) | 76(34.2) | 100(45.0) | 46(20.7) |          |          |
| Triglycerides (mg/dl) | 16(41.0) | 18(46.2) | 5(12.8) | 6.018 | 0.198 |
| Normal (>150) | 21(39.6) | 28(52.8) | 4(7.3) |          |          |
| Borderline high (150–200) |          |          |          |          |          |
| High (>200) |          |          |          |          |          |

*Statistically significant.

(LDL, CH, HDL, TG, PPBS, WBC, HB, RBC, BUN and albumin) were compared among the three genotypes of the ABCA1 gene g.1051G > A, where a statistically significant difference was observed for TG, which indicates that the persons with GG and GA had higher TG values when compared with those with AA (p = 0.047) (Table 5).

Table 5
Comparison of the mean rank values of clinical variables across three genotypes of the ABCA1 gene c.g.1051G > A in CAD study persons.

| Clinical variables | ABCA1 gene c.g.1051G > A | p-value |
|-------------------|--------------------------|---------|
|                   | GG | GA | AA |          |
| LDL | 154.51 | 156.92 | 159.49 | 0.943 |
| CH | 158.10 | 154.64 | 143.69 | 0.619 |
| HDL | 166.69 | 152.76 | 133.69 | 0.081 |
| TG | 153.78 | 160.77 | 126.54 | 0.047* |
| PPBS | 152.74 | 161.68 | 156.16 | 0.580 |
| WBC | 158.69 | 158.73 | 151.79 | 0.875 |
| HB | 160.45 | 150.82 | 169.17 | 0.498 |
| RBC | 163.72 | 150.14 | 164.25 | 0.403 |
| BUN | 167.42 | 154.10 | 143.24 | 0.209 |
| Albumin | 157.30 | 156.66 | 160.15 | 0.588 |

*Statistically significant.

3.5. Relation of risk factors associated with disease and genotyping of g.1051G > A in CAD-patients

In Table 6, when performing the \( \chi^2 \)-analysis, we tested the relationship between smoking vs. DM and HTN in the CAD cases and found to be insignificant association in DM (p = 0.35) and HTN (p = 0.58). At the same time, in Table 7, we cross-checked the g.1051G > A polymorphism with good and bad compliances of HTN in the CAD cases and found a non-significant association (p = 0.68) when the \( \chi^2 \) analysis was applied.

4. Discussion

Studies have been carried out in this current case - control study on the genetic association between g.1051G > A polymorphism in the gene ABCA1 and CAD in the Saudi population. The genetic interactions demonstrated the strong significant association between Allele, genotypes, inheritance pattern dominant and recessive. The parameters of the lipid profile (TC, TG and LDL-c levels were correlated with genotype levels in CAD patients. There were no significant differences in sex and age between CAD cases and controls as this study was chosen for similar age and genders involved in the study with respect to the presence or absence of the
mutated allele in the \( \text{ABCA}1 \) gene. This result was similar to results from another study where no association between the mutated gene \( \text{ABCA}1 \) and hypertension (Yao et al., 2016) was found. However, there was a significant correlation between Diabetes Mellitus and the g.1051G > A variant in the \( \text{ABCA}1 \) gene. A large number of patients whose genome contains polymorphisms have shown T2DM. This result suggested that patients carrying the variant g.1051G > A were more likely to develop DM. By comparison, the association between the possibility of developing T2DM and the \( \text{ABCA}1 \) variant rs2230806 was confirmed by a study conducted on an Asian population. This research conducted a meta-analysis and concluded that having the minor A allele could affect the prevalence of T2DM in senior patients in Asia (Jung et al., 2018a). Although the minor allele rs2230806 in \( \text{ABCA}1 \) has an impact on type II DM prevalence, according to the findings of a recent study (Cai et al., 2017), it has shown no influence on mild cognitive impairment (MCI) associated with DM.

The gene \( \text{ABCA}1 \) (OMIM: 600046) is known to be a membrane protein involved in membrane transport. The gene \( \text{ABCA}1 \) plays one of the key roles in an efflux of cholesterol from peripheral tissues transferred to the liver. The gene \( \text{ABCA}1 \) occurs on human chromosome region9q31.1. Due to the lack of HDL-c in plasma, the molecular function of genetic mutations involved in the \( \text{ABCA}1 \) gene was shown to be involved in the pathogenesis of Tangier disease characteristics and the propensity to develop premature CVD. There have been numerous variants in the \( \text{ABCA}1 \) gene and one of the well-known variants is rs2230806 (g.1051G > A or p. R219 K) SNP which appears on exon-7 and includes amino acid alterations from lysine to arginine and these alterations occur as a result of the substitution of G-A in the \( \text{ABCA}1 \) gene at position 1051 (Ghaznavi et al., 2018;Karimian et al., 2020).

The results from this study also presented a significant difference in cholesterol levels among patients carrying the g.1051G > A variants and the controls. The carriers of the wild-type allele and the carriers of the heterozygous allele had higher cholesterol levels than the homozygous carriers of the g.1051G > A variant. Furthermore, the levels of TG were significantly higher in CAD patients who were heterozygous for the g.1051G > A variant and patients who carried the wild-type allele when compared to the levels in the AA allele carriers. There was no significant difference in the levels of HDL and LDL between g.1051G > A carriers and wild-type carriers. However, HDL levels were higher in patients carrying the AA allele. This finding is consistent with another study that showed an association between the g.1051G > A variant in the \( \text{ABCA}1 \) gene and reduced HDL levels. The study concluded that the presence of the A allele could reduce the risk of developing CAD (Mokuno et al., 2015;Srinivasan et al., 2003).

Several case-control studies conducted around the globe in polymorphism g.1051G > A and found both positive and negative associations in the CAD. A recent meta-analysis study was conducted with g.1051G > A polymorphism in the CAD disease involving 43 different CAD case-control studies, and confirmed that K allele was significantly associated with reduced CAD risk in Asian and Caucasian populations (Fan et al., 2020). Our finding, however, was not in line with this review of meta-analysis and one of the explanations might be difference in an ethnicity. In addition to CAD, the g.1051G > A polymorphism was also used as meta-analysis studies in T2DM (Jung et al., 2018b) and Atherosclerosis (Yin et al., 2014), and confirmed negative association in T2DM and positive association with atherosclerosis. Our study was conducted in a relatively small subpopulation in the Saudi population’s western region, and the generalizability of current findings to other Saudi populations may require a further complementary study with a larger sample size from other Saudi Arabian regions. Nonetheless, with the associated polymorphism (g.1051G > A), a few studies with small and large sample sizes have been published. Nonetheless, research by Zargar et al (2013) showed an insignificant correlation with large sample size between the CAD cases

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**Table 6**

| Relation between Smoking vs DM, vs HTN | Smoking | Total | Chi-Square Tests | P value |
|----------------------------------------|---------|-------|-----------------|---------|
| DM                                     |         |       |                 |         |
| No                                     | n 63    | 65    | 128             | 0.352*  |
| %                                      | 49.2%   | 50.8% | 100%            |         |
| Yes                                    | n 102   | 85    | 187             |         |
| %                                      | 54.5%   | 45.5% | 100%            |         |
| HTN                                    |         |       |                 |         |
| No                                     | n 47    | 47    | 94              | 0.581*  |
| %                                      | 50.0%   | 50.0% | 100%            |         |
| Yes                                    | n 118   | 103   | 221             |         |
| %                                      | 53.4%   | 46.6% | 100%            |         |
| Total                                  | n 165   | 150   | 315             |         |
| %                                      | 52.4%   | 47.6% | 100%            |         |

*Insignificant statistically at \( \alpha \) 0.05%.

**Table 7**

| Relation between Genotyping and HTN | HTN | Total | Chi-Square Tests | P value |
|-------------------------------------|-----|-------|-----------------|---------|
| Genotyping                          |     |       |                 |         |
| GG                                  | n 31| 82    | 113             | 0.681   |
| %                                   | 27.4% | 72.6% | 100%            |         |
| GA                                  | n 44| 102   | 146             |         |
| %                                   | 30.1% | 69.9% | 100%            |         |
| AA                                  | n 19| 37    | 56              |         |
| %                                   | 33.9% | 66.1% | 100%            |         |
| Total                               | n 94| 221   | 315             |         |
| %                                   | 29.8% | 70.2% | 100%            |         |

*Insignificant statistically at \( \alpha \) 0.05%.
and Cyrus et al (2016). Whereas in our study we have chosen a modest sample size of 315 CAD cases and found to be associated with them, and one of the main reasons for significant association is to recruit Saudi subjects for CAD cases and healthy controls from western Kingdom area. In the Saudi population with C69T polymorphism, Alharbi et al (2013) conducted the case-control study in T2DM cases and found to be associated in the ABCA1 gene.

The study's strength was to include 315 CAD cases and 205 healthy Saudi-born controls. Another big strength of this study was drug definition in the patients with CAD. There are several limitations to this study, such as (i) lacking family history (ii) lack of evidence and (iii) performing only one SNP. The other limitations could be identified in the current study: (iv) specific coding SNPs similar to p.(V771 M), p.(I883 M) and p.(R1587 K) were not investigated in the ABCA1 gene and their relationship to the production of CADs, and (v) the functional effects of the g.1051G > A variant in the cholesterol efflux assay need to be examined to establish the cell impact.

5. Conclusion

CAD is one of the most prevalent diseases that endangers human life. The focus of updated studies was on revising the genetic causes of CAD and establishing a correlation between common genetic variants and CADs. In conclusion, the current study was established as the initial studies to show genetic association in the CAD patients with g.1051G > A polymorphism in the ABCA1 gene conducted in the Saudi population. By its lipid profile results, this study defines a correlation between the genetic variant g.1051G > A in the ABCA1 gene and the CAD. The A allele carriers have higher HDL than the wild type carriers, thereby reducing the risk for these patients to develop CAD later in life. Future studies in the entire ethnicities should be conducted to rule out the role of genetic polymorphism in the CAD and may be useful as a genetic marker.

Declaration of Competing Interest

The authors declare that they have no competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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