Research Article
Volume 7:3, 2023

Clinical and Medical Case Reports

ISSN: 2684-4915 Open Access

SLC8A1 Gene Polymorphism Rs13017968 and Hematological Parameters in Kawasaki Disease

Evdoxia Sapountzi[1]*, Andreas Giannopoulos[1], Styliani Fidani[1], Maria Trachana[2], Fragiskos Bersimis[3], Spiros Gerou[4] and Assimina Galli-Tsinopoulou[1]

[1]2nd Department of Paediatrics, School of Medicine, Aristotle University of Thessaloniki, AHEPA University General Hospital, Thessaloniki, Greece

[2]Department of Paediatrics, Paediatric Immunology and Rheumatology Referral Center, Hippokration General Hospital, Aristotle University of Thessaloniki, Thessaloniki, Greece

[3]Department of Statistics and Insurance Science, University of Piraeus, Piraeus, Greece

[4]Analysis Iatriki S.A., Biopathological Diagnostic Research Laboratories, 54623 Thessaloniki, Greece

Abstract

Kawasaki disease, an acute inflammatory vasculitis, affects children <5 years old. Coronary artery aneurysms constitute a serious complication of the disease. Both genetic factors and hematological parameters (e.g., mean platelet volume-to-lymphocyte ratio) have been implicated in the development of Kawasaki disease and coronary artery aneurysms. We explored the role of rs13017968, a single nucleotide polymorphism in *SLC8A1*, and of the mean platelet volume-to-lymphocyte ratio in Kawasaki disease and coronary artery aneurysms.

This single-center, case-control study included children with Kawasaki disease treated in our clinic and healthy children registered in the emergency unit. All patients received intravenous immunoglobulin upon diagnosis. Primary outcomes included rs13017968 frequency and the mean platelet volume-to-lymphocyte ratio in patients (with/without coronary artery aneurysms) and healthy children. Secondary outcomes included the mean platelet volume-to-lymphocyte ratio before and after intravenous immunoglobulin in patients (with/without coronary aneurysms) and correlation of the mean platelet volume-to-lymphocyte ratio with inflammation markers, age, and sex.

Overall, 107 healthy children and 59 patients (mean age: 139.38 months vs 37.36 months) were analyzed. Coronary aneurysms developed in 33.9% of the patients. No statistically significant association was detected between rs13017968 and occurrence of Kawasaki disease or coronary aneurysms. The mean platelet volume-to-lymphocyte ratio was significantly lower in patients than in healthy children and significantly decreased after therapy. No significant interaction was found between the mean platelet volume-to-lymphocyte ratio and coronary aneurysms. The only significant associations were between occurrence of coronary aneurysms and sex and between age and the mean platelet volume-to-lymphocyte ratio (before and after therapy) in patients.

Although our results do not support an association of rs13017968 with Kawasaki disease or coronary aneurysms, the relatively small sample size should be considered. The mean platelet volume-to-lymphocyte ratio, age, and sex appear as significant factors in Kawasaki disease and coronary artery aneurysms. Therefore, larger scale studies are warranted.

Keywords: Mean platelet volume • Coronary aneurysm • Kawasaki • Single nucleotide polymorphism

Abbreviations: ANOVA: Analysis of Variance; CAA: Coronary Artery Aneurysms; CRP: C-Reactive Protein; ECG: Electrocardiogram; ESR: Erythrocyte Sedimentation Rate; IVIG: Intravenous Immunoglobulin; KD: Kawasaki Disease; LYM: Lymphocytes; MPV: Mean Platelet Volume; MPVLR: Mean Platelet Volume-to-Lymphocyte Ratio; RT-PCR: Real-Time Polymerase Chain Reaction; SD: Standard Deviation; SNP: Single-Nucleotide Polymorphism

Introduction

Kawasaki Disease (KD) is an acute febrile and systemic vasculitis disease mainly affecting children <5 years old [1]. Since the first case of KD, reported in 1967 [1], extensive research has been performed on KD. Nevertheless, the cause of the disease is yet to be deciphered. Coronary artery lesions constitute the most common complications of KD and can significantly increase the risk of coronary heart disease. Coronary Artery Aneurysms (CAA) occur in up to 30% of untreated patients and can lead to ischemic heart disease, myocardial infarction, and even death [2]. The standard treatment for KD is high-dose Intravenous Immunoglobulin (IVIG) plus aspirin within 10 days from symptoms' appearance, which decreases the incidence of CAA to 5–7% [3-5]. Regardless, around 25% of the patients treated with IVIG become resistant or unresponsive, which increases the risk for CAA development [6].

Both non-genetic and genetic factors have been associated with KD susceptibility and IVIG unresponsiveness. Among the non-genetic factors, certain clinical features and laboratory factors such as a high erythrocyte sedimentation rate (ESR), low hemoglobin and platelet counts, oral mucosa alterations, cervical lymphadenopathy, extremities' swelling, and polymorphous rash have been linked to IVIG resistance [7]. Moreover, the Mean Platelet Volume-To-Lymphocyte Ratio (MPVLR) was found to be lower in patients with KD [8], whereas a high MPVLR (≥ 2.78) was identified as an independent risk predictor for IVIG resistance in infants with KD under 1 year [9].

***Address for Correspondence:** Evdoxia Sapountzi, 2nd Department of Paediatrics, School of Medicine, Aristotle University of Thessaloniki, AHEPA University General Hospital, Thessaloniki, Greece, Tel: 00306946465101, E-mail: esapountzi@gmail.com*

Received: 17 May, 2023, Manuscript No. cmcr-23-100794; **Editor assigned:** 18 May, 2023, Pre QC No. P-100794; **Reviewed:** 27 May, 2023, QC No. Q-100794; **Revised:** 31 May, 2023, Manuscript No. R-100794; **Published:** 09 June, 2023, DOI: 10.37421/2684-4915.2023.7.265

Among the genetic factors, polymorphisms in various genes such as *ITPKC* and *CASP3*, encoding for inositol-trisphosphate 3-kinase C and caspase 3, respectively [10-15]; cytokine-encoding and cytokine-related genes [6,16-19], Fcyreceptors [20-22]; genes involved in the TGF-ypathway [12,18,23]; and *PLA2G7* [18, 24,25], to name a few, have been associated with KD susceptibility and IVIG resistance (for a comprehensive review of polymorphisms affecting IVIG resistance, see [26]).

In this study, we evaluated polymorphism rs13017968 in *SLC8A1*, encoding for solute carrier family 8, member 1, and various hematological parameters, including MPVLR, as predictors of KD. A previous study by Shimizu C, et al. [27] identified 3 Single-Nucleotide Polymorphisms (SNPs) in *SLC8A1* that were associated with KD susceptibility in a Japanese cohort, among which rs13017968 was associated with a higher probability of CAA development and coronary vasodilation. These findings suggest that *SLC8A1* rs13017968 may influence both KD susceptibility and the risk of aneurysm formation and dilatation. Hence, we hypothesized that rs13017968 may play a role in the development of KD.

Materials and Methods

Study design

This case-control study was conducted in the Department of Pediatrics of AHEPA University Hospital (Thessaloniki, Greece) and was approved by the corresponding Ethical Review Board (Bioethics Committee, Protocol No. 2.623, 2/27.2.2019). The patients' parents or legal guardians provided written informed consent prior to study data collection.

Selection criteria

The study included 59 children aged up to 16 years who were diagnosed with KD based on the criteria of the American Heart Association for classic or complete, incomplete, and atypical Kawasaki [4] and treated in our department. Patients with the following characteristics were excluded from the study: history of hepatic disease, history of hematologic disease, malignancy, comorbidity of chronic inflammatory disease, history of chronic itching or eczema, receipt of medication that influence the hematologic parameters. Upon diagnosis, all patients received IVIG infusion at 2 g/kg for 12 hours.

One hundred and seven healthy children aged 6 to 16 years who were registered in the emergency unit were enrolled as control subjects. The selection of this age group was based on the fact that KD rarely occurs in children older than 5 years. Children with a recent history of fever or a medical history of KD at a younger age were excluded.

Outcome measures

Primary outcomes included the frequency of the genetic polymorphism rs13017968 in *SLC8A1* and comparison of the MPVLR in patients and control subjects, as well as in patients with and without CAA.

Secondary outcomes included the comparison of the MPVLR before and after the initial IVIG therapy in patients with and without CAA and correlation of the MPVLR with markers of inflammation, age, and sex.

Data collection

For every patient enrolled in the study, the following data were collected: patient demographics (sex, race, birth date, age in months), presence of the genetic polymorphism rs13017968 in *SLC8A1*. Additionally, the following were recorded before and after IVIG therapy: hematologic parameters (white blood cell count and type, platelet count, hemoglobulin levels, hematocrit, Mean Platelet Volume (MPV), platelet distribution width, and red blood cell distribution width), the levels of acute phase proteins (C Reactive Protein [CRP], ESR), and the MPVLR. All patients were subjected to classic Electrocardiogram (ECG) with Doppler imaging for the assessment of coronary vessels.

The following were recorded for all control subjects: demographics (sex, race, birth date, age in months), presence of the genetic polymorphism rs13017968 in *SLC8A1*, hematologic parameters (white blood cell count and type, platelet count, hemoglobulin levels, hematocrit, mean platelet volume, platelet distribution width, and red blood cell distribution width), and the MPVLR.

Examinations

After the parents' or legal guardians' consent, whole blood was collected from both healthy subjects and patients before IVIG treatment. The levels of urea, creatinine, transaminases, and albumin were determined in the serum using standard methods. CRP and ESR were also determined in all patients.

The number and type of white blood cells, platelet count, hemoglobulin, hematocrit, MPV, platelet distribution width, red blood cell distribution width, and MPVLR were determined. The MPVLR was calculated by dividing the MPV with the absolute number of lymphocytes ([LYM], 103/mm^3). For patients, the MPVLR was determined before and after the IVIG therapy.

In both patients and control subjects, 2 mL of heparinized venous blood was collected in EDTA tubes and kept at -80°C until use. DNA isolation from the blood samples was performed according to standard methods according to the manufacturer's instructions by using commercially available kits.

The identification of rs13017968 in *SLC8A1* was performed by real-time polymerase chain reaction (RT-PCR) using specific primers targeting the SNP and TaqMan SNP Genotyping Assay (Thermo Fisher Scientific; Waltham, MA). The RT-PCR was performed using the Real-Time PCR QuantStudio 5 (Thermo Fisher Scientific) and the following PCR cycling conditions: 95℃ for 10 minutes, and 40 cycles of 95℃ for 15 seconds and 60℃ for 60 seconds.

Coronary vessels evaluation

Classic 2D ultrasound ECG combined with Doppler imaging was performed in all patients to evaluate the coronary vessels and specifically the left coronary artery, anterior descending artery, and circumflex artery, as well as the right coronary artery. The ultrasound ECG remains the method of choice for early identification of aneurysms in coronary vessels in the first 6 weeks of disease onset [28].

The diagnosis criteria for coronary artery aneurysms related to Kawasaki disease are based on the absolute size of coronary vessels according to age [29].

The z-score was used to evaluate the imaging results of coronary arteries. The z-score is widely used and provides an association of the diameter of the coronary vessels with the total body surface, as well as the Standard Deviation (SD) relative to the mean in z units according to specific normograms. The presence of coronary artery aneurysms or ectasia in combination with a z-score >2.5 is considered a pathological finding. Z-score values <2.5 are considered normal. This classification constitutes a rough patient discrimination system and is recommended when KD is still suspected, aiming to reduce the number of patients at high risk of developing coronary aneurisms. The absolute aneurysm size is an indicator of damages that have already been detected at early stages [4,30,31].

The severity of Kawasaki disease is classified into 5 stages according to the ECG findings [30].

Statistical analysis

Sample size calculation

According to the literature [8], MPVLR is expected to be 3.32 with an SD of 2.66 in patients and 5.32 with an SD of 4.83 in control subjects. To determine the difference between the two groups and considering a statistical power of 80% and significance of 5%, we calculated that a total 120 participants were needed, i.e., 60 per group. The sample size was calculated using OpenEpi Version 3.01.

Statistical methods

Quantitative variables are described by using mean and standard deviation, and categorical variables are presented by using frequency and the corresponding percentage [32]. Kolmogorov-Smirnov and Shapiro-Wilk tests were applied to test the normality of quantitative variables' distributions [33].

The correlation of qualitative variables was assessed using the chi-square test or Fisher's exact test when the expected frequency was less than 5 [34-36]. For groups' comparisons concerning the means of quantitative variables, the parametric Student's t test for independent samples and paired samples was used [37]. In the case of paired samples (pre *vs.* post therapy), the effect of the independent variables "IVIG dosage" and "CAA" on the differentiation of the average MPVLR values between the two time points was investigated by applying analysis of variance (ANOVA) with a between subjects factor (mixed ANOVA) [38]. For applying mixed ANOVA, the corresponding assumptions were checked, and no particular violation was found (Box's test, Levene's test, etc.). Spearman correlation coefficient was used for assessing the correlation between quantitative variables that were not distributed normally and was validated by Pearson correlation coefficient [39,40]. All statistical analyses were performed using SPSS, and the level of statistical significance was set at 0.05.

Results

Study population characteristics

The research sample consisted of 60 patients with KD (cases) and 107 healthy participants (control). One case was removed as an outlier during data preprocessing; hence, the analyzed sample was 59 patients. The genetic and phenotypic characteristics of the study population are listed in Table 1. The allele of rs13017968 with the highest frequency was GT and that with the lowest frequency was TT, both in patients with KD and in healthy participants. Sex distribution differed slightly between cases and control, while the mean age of participants in the control group (139.38 months) was much higher than that of patients (37.36 months) (Table 1).

The KD-related features and hematological measurements according to the survey design (pre vs post therapy) are listed in table 2. Most patients had typical symptoms of the disease (79.7%) and quite a few had non-typical ones (18.6%). Only one patient had incomplete KD symptoms. CAA appeared in 33.9% of the patients and pericardial fluid in 22.0%. The complications of ectasia and mitral insufficiency were identified in 45.8% and 15.3% of the patients, respectively. Most patients received one IVIG dose (78.0%). The mean MPVLR decreased after IVIG treatment (Table 2).

Correlation between Kawasaki disease and rs13017968

No statistically significant relationship was detected between the allele of rs13017968 and the occurrence of KD (χ2=0.162, p=0.922, Chi Square test) (Table 3).

Correlation between CAA occurrence and rs13017968

In patients with KD, no statistically significant relationship was detected between the rs13017968 allele and the occurrence of CAA (p=0.096, Fisher's exact test (Figure 1). Nevertheless, there was a trend for a greater probability of CAA in patients carrying the TT allele than in those carrying the GG allele.

Difference in mean MPVLR values between patients and healthy participants

Kolmogorov-Smirnov and Shapiro-Wilk tests showed that the MPVLR was not normally distributed neither between patients and healthy participants

Table 1. Sample's genetic and phenotypic characteristics.

Variable	Category	Cases	Control
Polymorphism rs13017968	GG	22 (37.3%)	42 (39.3%)
	GT	30 (50.8%)	51 (47.7%)
	TT	7 (11.9%)	14 (13.1%)
Sex	Female	21 (35.6%)	61 (57.0%)
	Male	38 (64.4%)	46 (43.0%)
Age (Months)		37.36 (35.07)	139.38 (31.88)

Note: Categorical variables are presented as frequency (percent) and quantitative variables as mean (standard deviation).

Table 2. Kawasaki disease characteristics.

Variable	Category	Cases
Kawasaki symptoms	Typical	47 (79.7%)
	Non-typical	11 (18.6%)
	Incomplete	1 (1.7%)
CAA	Yes	20 (33.9%)
	No	39 (66.1%)
Pericardial fluid	Yes	13 (22.0%)
	No	46 (78.0%)
Ectasia	Yes	27 (45.8%)
	No	32 (54.2%)
Mitral insufficiency	Yes	9 (15.3%)
	No	50 (84.7%)
IVIG doses	1	46 (78.0%)
	2	13 (22.0%)
MPVLR	Pre	3.56 (2.24)
	Post	2.40 (1.03)

CAA: Coronary Artery Aneurysm; IVIG: Intravenous Immunoglobulin; MPVLR: Mean Platelet Volume-To-Lymphocyte Ratio

Note: Categorical variables are presented as frequency (percent) and quantitative variables as mean (standard deviation).

Table 3. Genotype frequency distribution of rs13017968 in patients with KD and healthy participants.

			Polymorphism RS13017968			Total
			GG	GT	TT	
Group	Control	Count	42	51	14	107
		% within Polymorphism rs13017968	65.6%	63.0%	66.7%	64.5%
	Case	Count	22	30	7	59
		% within Polymorphism rs13017968	34.4%	37.0%	33.3%	35.5%
All subjects		Count	64	81	21	166
		% within Polymorphism rs13017968	100.0%	100.0%	100.0%	100.0%

KD: Kawasaki Disease

Note: Case refers to patients with KD; Control refers to healthy participants.

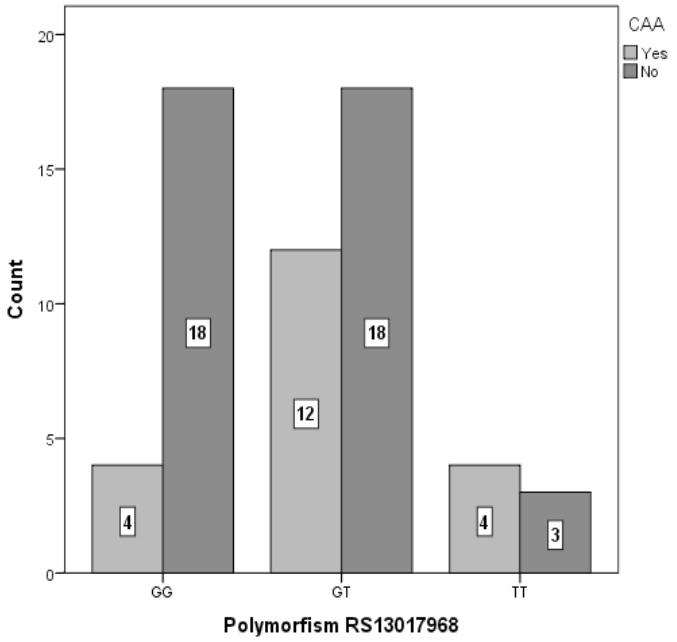

Figure 1. Comparative cluster bar chart between rs13017968 and CAA occurrence. CAA: Coronary Artery Aneurysm.

nor between patients before and after IVIG therapy; however, due to the sample sizes (n>30), parametric tests could be applied [41]. For investigating differences in the mean MPVLR between patients with KD and healthy participants, an independent samples t-test was applied. The mean MPVLR was significantly lower in patients with KD than in healthy participants (3.56 ± 2.24 *vs.* 4.58 ± 1.60, t=3.087, p=0.003). To analyze this result further, a corresponding hypothesis test was conducted for the hematological parameters "LYM" and "MPV". The mean LYM value was statistically significantly higher in patients than in healthy participants (4059.73 ± 2907.29 *vs.* 2518.93 ± 771.65, t=-3.994, p<0.05). In contrast, the mean MPV value was statistically significantly lower in patients than in healthy children (9.44 ± 1.03 *vs.* 10.39 ± 0.92, t=-6.056, p<0.05).

Difference in mean MPVLR values before and after IVIG therapy

A paired t-test was used for investigating differences in the mean MPVLR before and after IVIG treatment in patients with KD. IVIG therapy significantly decreased the mean MPVLR in patients (before: 3.56 ± 2.24 *vs.* after: 2.40 ± 1.03, t=3.963, p<0.01). In further analysis, no statistically significant difference was detected in hematological measurements "LYM" (t=-1.390, p=0.170) and "MPV" (t=0.502, p=0.618) between the two time points. Mixed ANOVA showed no significant interaction (F(1,57)=2.638, p=0.110) between "MPVLR" and "IVIG" (Figure 2, top) and no significant main effect of the number of IVIG doses (F(1,57)=0.043, p=0.837). There was also no significant interaction (F(1,57)=1.090 p=0.301) between "MPVLR" and "CAA" (Figure 2, bottom), and no significant main effect of MPVLR in the CAA group (F(1,57)=0.337, p=0.564). The assumptions for applying mixed ANOVA were satisfied (e.g., Box's Test p>0.05, Levene's Test p>0.05).

Correlation between CAA occurrence and sex in patients with KD

A statistically significant relationship was detected between CAA occurrence and sex ($\chi 2$=5.597, p=0.023). In particular, the probability of CAA occurrence was greater among boys than among girls with KD (Figure 3).

Correlation among Age, Pre MPVLR, and Post MPVLR for KD patients

The variables Age, Pre MPVLR, and Post MPVLR did not follow a normal distribution, and no clear linear pattern was visible in a scatter diagram (Figure 4). Therefore, the non-parametric Spearman's correlation coefficient was used for investigating the potential correlation among them, which was verified by the corresponding parametric Pearson's correlation coefficient. Age was positively and moderately correlated with the Pre MPVLR value (Spearman's rho=0.543, p<0.05; Table 4) and with the Post MPVLR value (Spearman's rho=0.534, p<0.05; Table 4). In addition, Pre MPVLR and Post MPVLR were also positively and weakly to moderately correlated (Spearman's rho=0.319, p<0.05; Table 4).

Discussion

In the present study, we investigated the relationship between a SNP in *SLC8A1*, rs13017968, and KD, as well as the impact of hematologic factors in KD expression.

According to a previous study, rs13017968 was identified as one of three SNPs affecting KD susceptibility in a Japanese cohort [27]. The three validated *SLC8A1* SNPs (rs10490051, rs13017968, and rs12989852), found in the same linkage disequilibrium block as a cluster of associated SNPs identified by GWAS in a European cohort, are located 80 kb downstream from the splice donor site of the first exon that is shared by all 15 transcript variants of *SLC8A1*. Patients who were homozygous for the risk allele had a higher risk of developing aneurysms/dilation [27].

The same group [42] utilized expression quantitative trait loci analysis and showed that patients homozygous for rs13017968 or any of the other two risk alleles in *SLC8A1* had elevated mRNA and plasma protein levels of urotensin

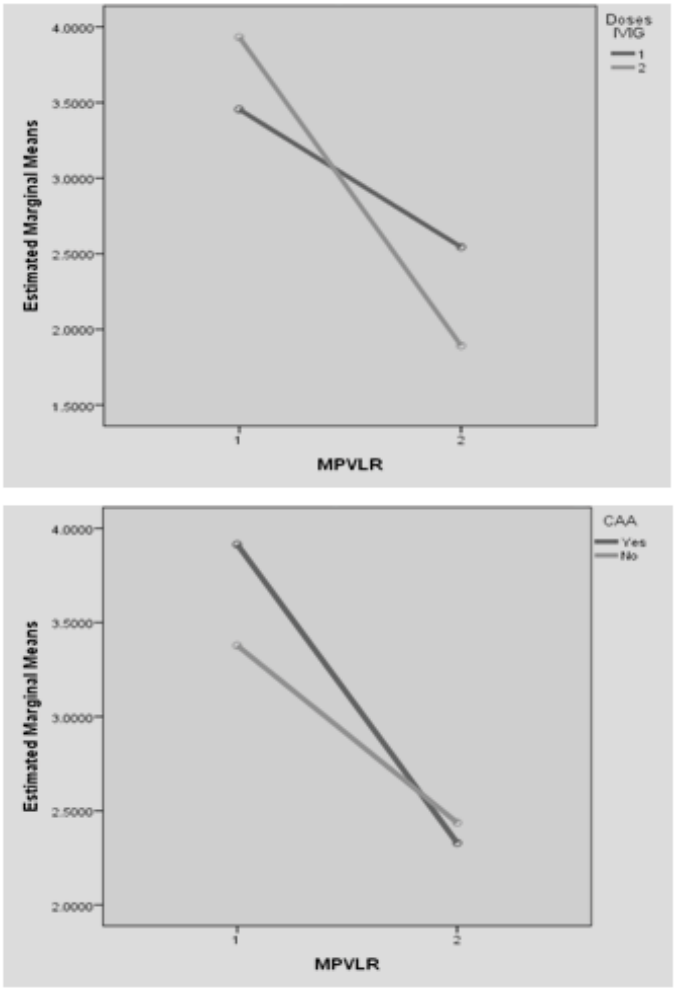

Figure 2. Estimated marginal means of MPVLR for each IVIG dose group (top graph) and each CAA group (bottom graph). CAA: Coronary Artery Aneurysm; IVIG: Intravenous Immunoglobulin; KD: Kawasaki Disease; MPVLR: Mean Platelet Volume-To-Lymphocyte Ratio.

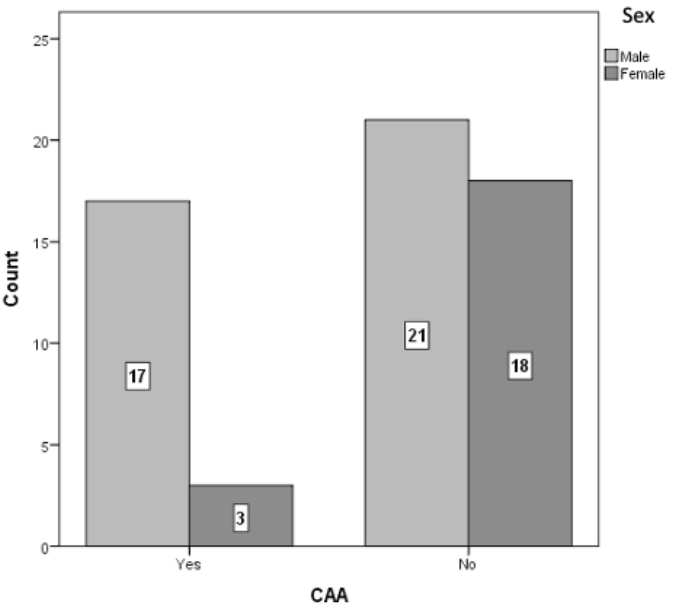

Figure 3. Comparative cluster bar chart between sex and CAA occurrence. CAA: Coronary Artery Aneurysm.

2, a molecule involved in vasoconstriction (*via* vascular smooth muscle cells) [43]. Based on the above findings, we postulated a role for rs13017968 in KD. However, when comparing a cohort of children with KD and healthy children,

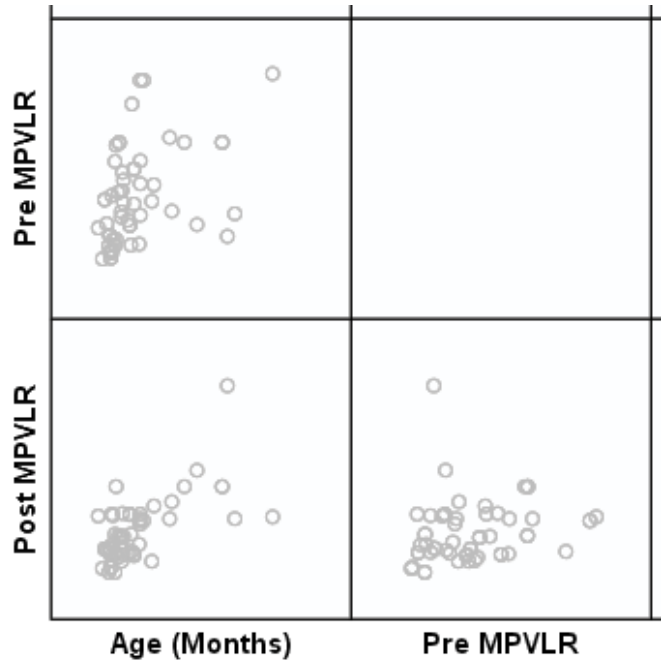

Figure 4. Matrix scatter plot of Age, Pre MPVLR, and Post MPVLR parameters in patients with KD. KD: Kawasaki Disease; MPVLR: Mean Platelet Volume-To-Lymphocyte Ratio.

Table 4. Correlation among Age, Pre MPVLR, and Post MPVLR in patients with KD.

Correlation Coefficient	Variable Pairs		
	Age - Pre MPVLR	Age - Post MPVLR	Pre MPVLR - Post MPVLR
Spearman's rho (p value)	0.543 (p<0.05)	0.534 (p<0.05)	0.319 (p=0.014)
Pearson (p value)	0.421 (p<0.05)	0.640 (p<0.05)	0.226 (p=0.055)

KD: Kawasaki Disease; MPVLR: Mean Platelet Volume-To-Lymphocyte Ratio

we did not confirm a significant difference in the frequency of rs13017968. Moreover, we could not confirm a significant difference in the frequency of rs13017968 between patients with and those without CAA despite a trend for a higher probability for developing CAA among patients carrying the TT than the GG allele. Possible population differences might explain the discrepancy between our and previous findings.

The MPVLR was found to be significantly lower in patients with KD than in healthy participants, whereas there was no significant interaction between MPVLR and CAA. Previous finding regarding the MPVLR in KD are contradictory, as both lower [8] and higher values have been reported in patients. The lower ratio in our study is justified by the significantly lower MPV values in patients than in controls and the respective significantly higher LYM values.

In agreement, two previous studies by Yu-wei HU, et al. [44] and Liu R, et al. [45] also reported significantly lower MPV in patients with KD than in control subjects. In contrast, Kim SH, et al. [46] showed no differences in the MPV between patients and controls at the time of KD diagnosis. In our study, we did not found an interaction between MPVLR and CAA development, which contradicts previous findings indicating lower MPVLR in patients with CAA [8]. However, consistently with previous studies [46], IVIG treatment decreased the mean MPVLR in our patient cohort. The children with KD in our study were significantly younger than the included healthy children, and we found that age significantly correlated with the MPVLR both before and after IVIG therapy. Therefore, potential differences might be explained by different population characteristics.

CAA is an important and the most potentially debilitating complication of KD. In our study, 33.9% of the patients had developed CAA. A significant association was identified between sex and the occurrence of CAA Specifically; boys were at higher risk of developing CAA, in agreement with the findings of previous studies [47-50].

An important limitation of the present study is the relatively small sample size of patients, which did not allow the use of parametric hypothesis tests to investigate all of the research questions. Moreover, the patients were enrolled from a single center. Nonetheless, the rarity of the disease justifies the small sample, particularly in a small country like Greece. Future similar investigations on a larger scale may be necessary to address the involvement of rs13017968 in KD and CAA formation, as well as the significance of the MPVLR in KD and/or CAA risk prediction. The identification of such genetic or hematological predictors is crucial in determining appropriate treatment plans for patients.

Conclusion

Overall, our results provide insight on KD characteristics and association with rs13017968 and hematological parameters in a small sample in Greece. Further, larger sample studies are needed to verify our results.

Competing Interests

The authors have no competing interests to declare that are relevant to the content of this article.

Funding

No funding was received to assist with the preparation of this manuscript.

References

1. Kawasaki, T. "Acute febrile mucocutaneous syndrome with lymphoid involvement with specific desquamation of the fingers and toes in children." *Arerugi* 16 (1967): 178-222.
2. Kato, Hirohisa, Tetsu Sugimura, Teiji Akagi and Noboru Sato, et al. "Long-term consequences of Kawasaki disease: a 10-to 21-year follow-up study of 594 patients." *Circulation* 94 (1996): 1379-1385.
3. Burns, Jane C., Edmund V. Capparelli, Jennifer A. Brown and Jane W. Newburger, et al. "Intravenous gamma-globulin treatment and retreatment in Kawasaki disease." *Pediatr Infect Dis* 17 (1998): 1144-1148.
4. McCrindle, Brian W., Anne H. Rowley, Jane W. Newburger and Jane C. Burns, et al. "Diagnosis, treatment and long-term management of Kawasaki disease: A scientific statement for health professionals from the American Heart Association." *Circulation* 135 (2017): e927-e999.
5. Tremoulet, Adriana H., Brookie M. Best, Sungchan Song and Susan Wang, et al. "Resistance to intravenous immunoglobulin in children with Kawasaki disease." *J Pediatr* 153 (2008): 117-121.
6. Amano, Yuji, Yohei Akazawa, Jun Yasuda and Kazuhisa Yoshino, et al. "A low-frequency IL4R locus variant in Japanese patients with intravenous immunoglobulin therapy-unresponsive Kawasaki disease." *Pediatr Rheumatol Online J* 17 (2019): 1-11.
7. Li, Xuan, Ye Chen, Yunjia Tang and Yueyue Ding, et al. "Predictors of intravenous immunoglobulin-resistant Kawasaki disease in children: A meta-analysis of 4442 cases." *Eur J Pediatr* 177 (2018): 1279-1292.
8. Bozlu, Gulcin, Derya Karpuz, Olgu Hallioglu and Selma Unal, et al. "Relationship between mean platelet volume-to-lymphocyte ratio and coronary artery abnormalities in Kawasaki disease." *Cardiol Young* 28 (2018): 832-836.
9. Wu, Shu, Yuan Long, Selena Chen and Yaqian Huang, et al. "A new scoring system for prediction of intravenous immunoglobulin resistance of Kawasaki disease in infants under 1-year old." *Front Pediatr* 7 (2019): 514.
10. Onouchi, Yoshihiro, Tomohiko Gunji, Jane C. Burns and Chisato Shimizu, et al. "ITPKC functional polymorphism associated with Kawasaki disease susceptibility and formation of coronary artery aneurysms." *Nat Genet* 40 (2008): 35-42.
11. Kim, Kyu Yeun, Yoon-Sun Bae, Woohyuk Ji and Dongjik Shin, et al. "ITPKC and

SLC11A1 gene polymorphisms and gene-gene interactions in Korean patients with Kawasaki disease." *Yonsei Med J* 59 (2018): 119-127.

12. Kuo, Ho-Chang, Kuender D. Yang, Suh-Hang Hank Juo and Chi-Di Liang, et al. "ITPKC single nucleotide polymorphism associated with the Kawasaki disease in a Taiwanese population." *PloS one* 6 (2011): e17370.
13. Peng, Qian, Changhui Chen, Yu Zhang and Hailan He, et al. "Single-nucleotide polymorphism rs2290692 in the 3′ UTR of ITPKC associated with susceptibility to Kawasaki disease in a Han Chinese population." *Pediatr Cardiol* 33 (2012): 1046-1053.
14. Kuo, Ho-Chang, Yu-Wen Hsu, Chung-Min Wu and Shawn Hsiang-Yin Chen, et al. "A replication study for association of ITPKC and CASP3 two-locus analysis in IVIG unresponsiveness and coronary artery lesion in Kawasaki disease." *PLoS One* 8 (2013): e69685.
15. Onouchi, Y., Y. Suzuki, H. Suzuki and M. Terai, et al. "ITPKC and CASP3 polymorphisms and risks for IVIG unresponsiveness and coronary artery lesion formation in Kawasaki disease." *Pharmacogenomics J* 13 (2013): 52-59.
16. Kim, Hea-Ji, Jae-Jung Kim, Sin Weon Yun and Jeong Jin Yu, et al. "Association of the IL16 Asn1147Lys polymorphism with intravenous immunoglobulin resistance in Kawasaki disease." *J Hum Genet* 65 (2020): 421-426.
17. Weng, Ken-Pen, Kai-Sheng Hsieh, Tsyr-Yuh Ho and Shih-Hui Huang, et al. "IL-1B polymorphism in association with initial intravenous immunoglobulin treatment failure in Taiwanese children with Kawasaki disease." *Circ J* 74 (2010): 544-551.
18. Meng, Li, Zhen Zhen, Qian Jiang and Xiao-hui Li, et al. "Predictive model based on gene and laboratory data for intravenous immunoglobulin resistance in Kawasaki disease in a Chinese population." *Pediatr Rheumatol Online J* 19 (2021): 1-10.
19. Mamtani, Manju, Tomoyo Matsubara, Chisato Shimizu and Susumu Furukawa, et al. "Association of CCR2-CCR5 haplotypes and CCL3L1 copy number with Kawasaki disease, coronary artery lesions and IVIG responses in Japanese children." *PLoS One* 5 (2010): e11458.
20. Khor, Chiea Chuen, Sonia Davila, Willemijn B. Breunis and Yi-Ching Lee, et al. "Genome-wide association study identifies FCGR2A as a susceptibility locus for Kawasaki disease." *Nat Genet* 43 (2011): 1241-1246.
21. Shrestha, Sadeep, Howard Wiener, Aditi Shendre and Richard A. Kaslow, et al. "Role of activating Fc γR gene polymorphisms in Kawasaki disease susceptibility and intravenous immunoglobulin response." *Circ Cardiovasc Genet* 5 (2012): 309-316.
22. Shrestha, Sadeep, Howard W. Wiener, Aaron K. Olson and Jeffrey C. Edberg, et al. "Functional FCGR2B gene variants influence intravenous immunoglobulin response in patients with Kawasaki disease." *J Allergy Clin Immunol* 128 (2011): 677-680.
23. Shimizu, Chisato, Sonia Jain, Sonia Davila and Martin L. Hibberd, et al. "Transforming growth factor-β signaling pathway in patients with Kawasaki disease." *Circ Cardiovasc Genet* 4 (2011): 16-25.
24. Minami, Takaomi, Hiroyuki Suzuki, Takashi Takeuchi and Shigeru Uemura, et al. "A polymorphism in plasma platelet-activating factor acetylhydrolase is involved in resistance to immunoglobulin treatment in Kawasaki disease." *J Pediatr* 147 (2005): 78-83.
25. Gu, Xueping, Wenchun Lin, Yufen Xu and Di Che, et al. "The rs1051931 G> A polymorphism in the PLA2G7 gene confers resistance to immunoglobulin therapy in Kawasaki disease in a Southern Chinese population." *Front Pediatr* 8 (2020): 338.
26. Sapountzi, E., L. Fidani, A. Giannopoulos and A. Galli-Tsinopoulou. "Association of genetic polymorphisms in Kawasaki disease with the response to intravenous immunoglobulin therapy." *Pediatr Cardiol* 44 (2023): 1-12.
27. Shimizu, Chisato, Hariklia Eleftherohorinou, Victoria J. Wright and Jihoon Kim, et al. "Genetic variation in the SLC8A1 calcium signaling pathway is associated with susceptibility to Kawasaki disease and coronary artery abnormalities." *Circ Cardiovasc Genet* 9 (2016): 559-568.
28. McCrindle, Brian W. and Barbara Cifra. "The role of echocardiography in Kawasaki disease." *Int J Rheum Dis* 21 (2018): 50-55.
29. Japan Kawasaki Disease Research Committee. "Report of subcommittee on standardization of diagnostic criteria and reporting of coronary artery lesions in Kawasaki disease." Tokyo: Ministry of Health and Welfare (1984).
30. McCrindle, Brian W., Jennifer S. Li, L. LuAnn Minich and Steven D. Colan, et al. "Coronary artery involvement in children with Kawasaki disease: Risk factors from analysis of serial normalized measurements." *Circulation* 116 (2007): 174-179.
31. JCS Joint Working Group. "Guidelines for diagnosis and management of cardiovascular sequelae in Kawasaki disease (JCS 2013)–digest version–." *Circ J* 78 (2014): 2521-2562.
32. Bersimis, S, Bersimis F, Sahlas A. "Introduction to Statistics and Probability, 2nd Edition, Tziola & Sons SA Publications (Greek Language)." (2022).
33. Hanusz, Zofia and Joanna Tarasińska. "Normalization of the Kolmogorov–Smirnov and Shapiro–Wilk tests of normality." *Biometrical Lett* 52 (2015): 85-93.
34. Agresti, Alan. "A survey of exact inference for contingency tables." *Stat Sci* 7 (1992): 131-153.
35. Fisher, Ronald A. "On the interpretation of χ^2 from contingency tables and the calculation of P." *J R Stat Soc* 85 (1922): 87-94.
36. Mehta, Cyrus R. and Nitin R. Patel. "A network algorithm for performing Fisher's exact test in r × c contingency tables." *J Am Stat Assoc* 78 (1983): 427-434.
37. Derrick, Ben. "How to compare the means of two samples that include paired observations and independent observations: A companion to Derrick, Russ, Toher and White (2017)." *Quant Meth Psych* 13 (2017): 120-126.
38. Huynh, Huynh and Leonard S. Feldt. "Conditions under which mean square ratios in repeated measurements designs have exact F-distributions." *J Am Stat Assoc* 65 (1970): 1582-1589.
39. Lehman, A. "JMP for basic univariate and multivariate statistics: A step-by-step guide. 481p." SAS Institute. (2005).
40. Wright, Sewall. "Correlation and causation." *J Agric Res* 20(1921): 557-585.
41. Curran-Everett, Douglas. "Explorations in statistics: The assumption of normality." *Adv Physiol Educ* 41 (2017): 449-453.
42. Huang, Cassidy Y., Jane C. Burns and Chisato Shimizu. "Urotensin 2 in Kawasaki disease pathogenesis." *Pediatr Res* 82 (2017): 1048-1055.
43. Ross, Bryan, Katherine McKendy and Adel Giaid. "Role of urotensin II in health and disease." *Am J Physiol Regul Integr Comp Physio* 298 (2010): R1156-R1172.
44. Yu-wei, H. U., Zhou Chuan-xin and Chen Li-hua. "Significance of platelet parameters in children with Kawasaki disease in diagnosis and prognosis." *J Appl Clin Pediatr* 13 (2006).
45. Liu, Ruixi, Fang Gao, Junming Huo and Qijian Yi. "Study on the relationship between mean platelet volume and platelet distribution width with coronary artery lesion in children with Kawasaki disease." *Platelets* 23 (2012): 11-16.
46. Kim, Sung Hoon, In Ji Hwang and Young Kuk Cho. "Platelet indices as diagnostic marker for Kawasaki disease." *Chonnam Med J* 58 (2022): 110-118.
47. Yalcinkaya, Rumeysa, Fatma Nur Öz, Sevgi Yaşar Durmuş and Ali Fettah, et al. "Is there a role for laboratory parameters in predicting coronary artery involvement in Kawasaki disease?." *Klin Padiatr* 234(2022): 382-387.
48. Chiang, Chun-Yen, Chung-Han Ho, Chin-Chen Chu and Zhih-Cherng Chen, et al. "Coronary artery complications in pediatric patients with Kawasaki disease: A 12-year national survey." *Acta Cardiol Sin* 29 (2013): 357.
49. Tang, Yunjia, Xiang Gao, Jie Shen and Ling Sun, et al. "Epidemiological and clinical characteristics of Kawasaki disease and factors associated with coronary artery abnormalities in East China: Nine years experience." *J Trop Pediatr* 62 (2016): 86-93.
50. Van Stijn, Diana, Justin M. Korbee, Stejara A. Netea and Vera C. de Winter, et al. "Treatment and coronary artery aneurysm formation in Kawasaki disease: A per-day risk analysis." *J Pediatr* 243 (2022): 167-172.

How to cite this article: Sapountzi, Evdoxia, Andreas Giannopoulos, Styliani Fidani and Maria Trachana, et al. "*SLC8A1* Gene Polymorphism Rs13017968 and Hematological Parameters in Kawasaki Disease." *Clin Med Case Rep* 7 (2023): 265.