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Distribution of the Factor XIII-A Val34Leu (rs5985) Polymorphism in a Libyan Coronary Artery Disease Study Cohort: A Pilot Genotyping Study

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ABSTRACT

Background: The factor XIII-A (FXIII-A) Val34Leu (rs5985) variant may influence fibrin structure, but its distribution in Libyan clinical datasets is poorly described. This study aimed to describe recorded FXIII-A Val34Leu genotypes in a Libyan dataset containing case- and control-labelled records; group comparisons were secondary and exploratory.

Methods: The study dataset from Al Fouad Cardiac Center, Tripoli, contained 82 records, 65 labelled case and 17 control. Fifty-four records had valid genotype calls (42 cases and 12 controls), determined with a commercial strip assay. Genotype counts, allele proportions and recorded D-dimer and lipid measurements were statistically analyzed using two-sided Fisher exact, exact Hardy–Weinberg and descriptive biochemical comparisons.

Results: Among valid genotype records, cases included 26 Val/Val and 16 Val/Leu; controls included 7 Val/Val and 5 Val/Leu. No Leu/Leu call was present. The Leu34 allele frequency was 19.0% (16/84; exact 95% confidence interval (CI) 11.3%–29.1%) in cases and 20.8% (5/24; 7.1%–42.2%) in controls. The case–control genotype comparison gave Fisher $p = 1.000$ and an odds ratio (OR) of 0.86 (Woolf 95% CI 0.23–3.18). Mean age was 63.8 ± 11.9 versus 35.4 ± 7.8 years. D-dimer was recorded for 38 cases and 7 controls and was higher in case-labelled records (median 0.75 vs 0.14 µg/mL, unadjusted Mann–Whitney $p < 0.001$).

Conclusions: These results provide a small descriptive distribution of recorded FXIII genotypes, not a reliable coronary artery disease (CAD) risk association estimate. Unequal ages, high genotype failure, incomplete clinical phenotyping and assay documentation, and limited control numbers require careful source-record verification and independent replication.

توزيع تعدد الأشكال Val34Leu (rs5985) في الوحدة A للعامل الثالث عشر لدى مجموعة دراسية ليبية من مرضى الشريان التاجي: دراسة تنميط جيني تجريبية

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الكلمات المفتاحية:
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الملخص

الخلفية: قد يؤثر المتغير Val34Leu (rs5985) في الوحدة A للعامل الثالث عشر (FXIII-A) في بنية الفيبرين، غير أن توزيعه في البيانات السريرية الليبية ما يزال غير موصوف جيدًا. هدفت هذه الدراسة إلى وصف الأنماط الجينية المسجلة لهذا المتغير في بيانات ليبية تضم سجلات حالات وضوابط، وكانت المقارنات بين المجموعتين ثانوية واستكشافية.

الطرائق: تضمنت بيانات الدراسة المأخوذة من مركز الفؤاد للقلب بطرابلس 82 سجلًا، منها 65 حالة و17 ضابطًا. وتوافرت نتائج تنميط صالحة لـ54 سجلًا (42 حالة و12 ضابطًا)، حُددت باستخدام اختبار شرائط تجاري. وحُللت أعداد الأنماط الجينية ونسب الأليلات وقياسات D-dimer والدهون المسجلة إحصائيًا باستخدام اختبار فيشر الدقيق ثنائي الاتجاه، واختبار هاردي–واينبرغ الدقيق، ومقارنات كيميائية حيوية وصفية.

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النتائج: بين السجلات الصالحة، شملت الحالات 26 بالنمط Val/Val و16 بالنمط Val/Leu، وشملت الضوابط 7 بالنمط Val/Val و5 بالنمط Val/Leu، ولم يُسجل النمط Leu/Leu. وبلغ تكرار الأليل Leu34 نسبة 19.0% (16/84؛ فاصل الثقة الدقيق 95%: 11.3%–29.1%) لدى الحالات و20.8% (5/24؛ 7.1%–42.2%) لدى الضوابط. وأعطت مقارنة الأنماط الجينية قيمة فيشر $p = 1.000$ ونسبة أرجحية 0.86 (فاصل الثقة 95% بطريقة وولف: 0.23–3.18). وبلغ متوسط العمر 63.8 ± 11.9 مقابل 35.4 ± 7.8 سنة. وسُجلت قيمة D-dimer لدى 38 حالة و7 ضوابط، وكانت أعلى لدى الحالات (الوسيط 0.75 مقابل 0.14 ميكروغرام/مل، قيمة مان–ويتني غير المعدلة $p < 0.001$).

الاستنتاجات: تقدم هذه النتائج توزيعًا وصفيًا محدودًا للأنماط الجينية المسجلة للعامل الثالث عشر، ولا تمثل تقديرًا موثوقًا لارتباطه بخطر الإصابة بمرض الشريان التاجي. ويستلزم تفاوت الأعمار، وارتفاع نسبة فشل التنميط الجيني، ونقص التوصيف السريري وتوثيق الاختبار، ومحدودية عدد الضوابط، التحقق من السجلات الأصلية وتكرار الدراسة بصورة مستقلة.

1. Introduction

Coronary artery disease (CAD) is influenced by atherosclerotic and thrombotic processes, and genetic variation in hemostatic proteins has been studied as a possible contributor to disease susceptibility [1,2]. Factor XIII (FXIII) helps stabilize fibrin clots after activation, and variation in the FXIII-A subunit may modify clot formation [3,4].

The *F13A1* Val34Leu polymorphism (rs5985) is located close to the thrombin activation site of FXIII-A. Previous experimental and observational studies have explored its effects on activation and clot characteristics, and cardiovascular associations have not been uniform across study settings [5–7]. Findings in premature coronary disease should not automatically be extrapolated to an older hospital-based cohort.

Given the small Libyan dataset available, this analysis is designed primarily to report observed genotype and allele frequencies in case-labelled records. A control comparison is included for descriptive context, not as a definitive test of an association between genotype and angiographically confirmed CAD.

2. Materials and Methods

2.1. Data source and eligibility for analysis

The source for all participant-level results was the investigators' study database, containing 82 records with study-group, demographic, diagnosis, treatment, laboratory and FXIII genotype fields. Group entries were coded as case or control. Records with FXIII genotype fields marked “Wild Type” or “Heterozygous” formed the analytic cohort; entries marked “Test failed” were excluded from genotype-dependent analyses but included in the screening and failure summary. No missing laboratory values were imputed.

Case–control status was taken from the recorded group label and was not independently adjudicated against angiography or hospital source records. The linked diagnosis field includes heterogeneous descriptions: 28 of 42 genotyped case-labelled records explicitly contain “CAD” or “IHD” (ischaemic heart disease), while 14 do not. Therefore, all case-labelled records cannot be represented as independently verified CAD diagnoses from the study records alone.

2.2. Study setting, recruitment and ethics

The study was conducted at Al Fouad Cardiac Center, Tripoli, Libya, between February and October 2025. Patients were recruited consecutively from those referred for diagnostic coronary angiography during the study period. Cases were defined as patients with angiographically confirmed coronary artery disease, defined as the presence of ≥50% stenosis in at least one major epicardial coronary artery (or ≥70% in a major epicardial vessel, depending on the diagnostic threshold applied) on coronary angiography. Controls were selected from patients who underwent coronary angiography during the same period and were found to have normal coronary arteries or non-obstructive disease (<50% stenosis), with no history of prior myocardial infarction or revascularization. No age or sex matching was applied in the available analytic sample. The study was approved by the Libyan Medical Research Center (approval no. NBC:018.H.24.64, dated 03 January 2025) and was conducted in accordance with the Declaration of Helsinki. Informed consent was obtained from all participants.

2.3. Genotyping and clinical measurements

Genotyping was performed using the CVD StripAssay (ViennaLab Diagnostics GmbH, Vienna, Austria). Genomic DNA was extracted with the silica-membrane reagents supplied with the CVD Strip Assay kit (Vienna Lab Diagnostics GmbH, Vienna, Austria; catalogue no. 6-120-A-100), and DNA quality and yield were assessed on a NanoDrop 2000 spectrophotometer (A260/A280 and A260/A230). The FXIII field was recorded as “Wild Type,” “Heterozygous” or “Test failed”; for this descriptive analysis, these are interpreted as Val/Val, Val/Leu and missing genotype, respectively. No Leu/Leu label was present.

The numerical fields comprise D-dimer, total cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol, and triglycerides, measured using analyzers and reagents (Integra, Roche Diagnostics, Rotkreuz, Switzerland). Lipid summaries use the recorded values; the LDL values do not consistently agree with Friedewald calculations from the other lipids,

so the actual LDL measurement or calculation method remains unverified. D-dimer was recorded in µg/mL, but the records do not specify fibrinogen-equivalent units (FEU) versus D-dimer units (DDU), analyzer, clinical threshold or lower limit of quantification. No information establishing the blood-draw interval after coronary events is present in the records.

2.4. Statistical analysis

Counts and percentages summarize categorical fields. Age and lipid results are shown as mean ± sample standard deviation (SD); D-dimer, which is strongly right-skewed, is shown as median and interquartile range (IQR, 25th to 75th percentile using linear interpolation). The two-sided Fisher exact test compares Val/Leu versus Val/Val between case and control labels (a dominant-model comparison in the absence of Leu/Leu). The odds ratio (OR) is defined as the odds of Val/Leu among cases divided by the corresponding odds among controls; its 95% confidence interval (CI) uses the log/Woolf approximation. Allele frequencies are described with exact Clopper–Pearson 95% intervals; the allelic Fisher test is exploratory because paired alleles within individuals are not statistically independent. Exact Hardy–Weinberg equilibrium (HWE) tests condition on the observed allele totals [8].

Available-case Welch t-tests were computed for lipids; two-sided Mann–Whitney U tests were used for D-dimer. These are unadjusted exploratory comparisons, and p-values are not evidence of an effect of CAD or genotype. There was no correction for multiple testing. Analyses used Python 3.13.5 (Python Software Foundation, Wilmington, DE, USA), SciPy 1.17.0 and NumPy 2.3.5. Missing measurements were excluded only from analyses requiring those measurements.

3. Results

3.1. Participant flow and basic characteristics

Of 82 records in the study database, 65 were recorded as cases and 17 as controls. Valid genotype calls were available for 42/65 cases (64.6%) and 12/17 controls (70.6%); 23 case and 5 control records had failed genotype results (overall, 28/82, 34.1%). The genotyped group comprised 42 cases and 12 controls. The observed age ranges were 35–79 years in cases and 24–50 years in controls (Table 1).

Table 1. Record flow and characteristics of participants with valid genotype calls

Characteristic	Case label	Control label
All records, n	65	17
Valid genotype, n	42 (64.6%)	12 (70.6%)
Failed genotype, n	23 (35.4%)	5 (29.4%)
Women, n (%)	24 (57.1%)	8 (66.7%)
Men, n (%)	18 (42.9%)	4 (33.3%)
Age, years, mean ± SD	63.8 ± 11.9	35.4 ± 7.8
Age range, years	35–79	24–50
Diagnosis field contains “CAD” or “IHD”, n	28/42	Not applicable (12 labelled healthy)

Group and diagnoses are transcribed from the study records. Presence of “CAD” or “IHD” is an explicit text search, not verification of the clinical diagnosis. Valid genotype percentages use screened records within the corresponding group. CAD, coronary artery disease; IHD, ischaemic heart disease; SD, standard deviation.

Case-labelled records were older than controls by approximately 28.4 years in the valid-genotype subset. Clinical diagnoses in the case group were heterogeneous; therefore, a confirmed CAD-only cohort cannot be identified solely from the study records. Reported medications appear as free text, but statin use, antiplatelet use and anticoagulant use cannot be reliably enumerated as verified binary variables without clinical validation. Smoking and body mass index (BMI) were not present as structured fields.

3.2. Genotype distribution, alleles and HWE

Val/Val was recorded in 33/54 (61.1%) genotyped participants, and Val/Leu in 21/54 (38.9%); no Leu/Leu genotype was recorded. The Val/Leu proportion was 16/42 (38.1%) among cases and 5/12 (41.7%) among controls. The two-sided genotype Fisher test gave $p = 1.000$ (Table 2). The OR for Val/Leu versus Val/Val was 0.86 (95% CI 0.23–3.18), a wide interval consistent with considerable uncertainty rather than a demonstration of equivalence.

Table 2. FXIII-A Val34Leu genotype frequencies by study-group label

Genotype	Cases (n = 42)	Controls (n = 12)	Total (n = 54)
Val/Val	26 (61.9%)	7 (58.3%)	33 (61.1%)
Val/Leu	16 (38.1%)	5 (41.7%)	21 (38.9%)
Leu/Leu	0 (0.0%)	0 (0.0%)	0 (0.0%)
Total	42 (100%)	12 (100%)	54 (100%)

Genotype Fisher exact test, two-sided $p = 1.000$; dominant-model OR = 0.86 (Woolf 95% CI 0.23–3.18). No Leu/Leu calls were present. CI, confidence interval; OR, odds ratio.

The Leu34 allele was observed 16 times among 84 case alleles (19.0%; exact 95% CI 11.3%–29.1%) and 5 times among 24 control alleles (20.8%; exact 95% CI 7.1%–42.2%). An exploratory allelic Fisher comparison gave $p = 1.000$. Exact HWE tests gave $p = 0.312$ in cases, $p = 1.000$ in controls, and $p = 0.181$ in the pooled genotyped sample. Given the small control group, these HWE tests have limited ability to detect genotyping errors.

3.3. Recorded biochemical measurements

D-dimer was available for 38/42 cases and 7/12 controls. The recorded case median was 0.746 (0.464–1.476) µg/mL compared with 0.140 (0.135–0.155) µg/mL in controls. The two-sided Mann–Whitney p-value was < 0.001. The interpretation depends on missingness and unverified assay and blood-collection information. All four lipid measurements were complete for each of the 54 valid-genotype records (Table 3).

Table 3. Biochemical data in valid-genotype case and control records

Measurement	Cases	Controls	Exploratory *p*
D-dimer (µg/mL), median (IQR)	0.746 (0.464–1.476) (n = 38)	0.140 (0.135–0.155) (n = 7)	< 0.001
Total cholesterol (mg/dL)	144.6 ± 48.3	169.1 ± 24.1	0.021
HDL cholesterol (mg/dL)	41.5 ± 13.1	53.4 ± 17.8	0.048
LDL cholesterol (mg/dL)	86.5 ± 35.5	104.8 ± 28.6	0.078
Triglycerides (mg/dL)	137.8 ± 92.6	103.5 ± 47.8	0.093

D-dimer is median (25th–75th percentile), Mann–Whitney U test; lipids are mean ± SD, Welch *t*-test. All *p* values are two-sided, unadjusted and descriptive. Group imbalance, treatment exposure and unverified diagnosis limit clinical interpretation. D-dimer assay unit basis (FEU/DDU) and LDL methodology are unknown. HDL, high-density lipoprotein; IQR, interquartile range; LDL, low-density lipoprotein; SD, standard deviation.

Mean recorded total cholesterol and HDL were numerically lower in the case group; however, these unadjusted differences should not be attributed to CAD. Treatment notes in the records suggest differences in medication exposure but verified structured medication histories are not available. LDL values are taken directly from the recorded data without assuming that they were calculated by the Friedewald formula.

3.4. Biochemistry by FXIII genotype within case-labelled records

Biochemical measures by genotype among case-labelled records are presented in Table 4.

Table 4. Biochemical measures by genotype among case-labelled records

Measurement	Val/Val (n = 26)	Val/Leu (n = 16)	Exploratory *p*
D-dimer (µg/mL), median (IQR)	0.640 (0.450–1.363) (n = 22)	0.862 (0.622–2.076) (n = 16)	0.214
Total cholesterol (mg/dL)	134.0 ± 48.0	161.8 ± 45.0	0.067
HDL cholesterol (mg/dL)	39.5 ± 12.1	44.6 ± 14.6	0.246
LDL cholesterol (mg/dL)	80.0 ± 38.2	97.1 ± 28.5	0.105
Triglycerides (mg/dL)	132.1 ± 91.1	147.1 ± 97.3	0.623

D-dimer is median (IQR), available in 22 Val/Val and all 16 Val/Leu case records; Mann–Whitney U test. Lipids are mean ± SD; Welch *t*-tests. *p* values are unadjusted, exploratory comparisons. Abbreviations as in Table 3.

No genotype-stratified biochemical comparison within case-labelled records met the nominal $p < 0.05$ threshold. In particular, total cholesterol differed numerically between Val/Leu and Val/Val but had Welch $p = 0.067$. The lack of observed differences should not be interpreted as evidence of no biological effect.

4. Discussion

This descriptive analysis identifies 33 Val/Val and 21 Val/Leu calls among 54 genotyped individuals, including 16 Val/Leu calls among 42 case-labelled participants and 5 Val/Leu calls among 12 controls. The case–control genotype comparison gave Fisher $p = 1.000$ with OR 0.86 (95% CI 0.23–3.18). A nonsignificant result in this small, clinically heterogeneous sample cannot be taken as evidence that FXIII-A Val34Leu has no relationship with CAD.

The case Leu34 allele frequency of 19.0% (exact 95% CI 11.3%–29.1%) is the most defensible primary descriptive result. The pooled allele frequency is not a population estimate because the sample combines clinical cases with a small, age-disparate control group. Although earlier observational studies and meta-analyses explore this variant in coronary outcomes [1,7], meaningful comparison of effect estimates requires comparable participant selection, disease definitions, genotype verification and sufficient sample size.

The observed absence of a Leu/Leu genotype prevents an analysis of variant homozygotes. The HWE results do not independently validate strip-assay accuracy. The 34.1% overall genotype-failure rate further undermines precision: failure occurred in 23 of 65 case-labelled records and 5 of 17 controls. In the absence of documented assay quality-control records, it is not possible to establish whether missingness is related to collection conditions, testing batches or genotype itself.

Biochemical values are particularly vulnerable to interpretation error. Case-labelled participants are nearly three decades older than controls, and treatment histories are encoded as free text rather than verified medication categories. D-dimer was missing disproportionately in controls (5/12 compared with 4/42 cases), and its analytical reporting system is not identified. The divergent recorded LDL and Friedewald-derived means do not prove either direct measurement or a specific transcription error; the laboratory method should be checked against the original reports.

A further limitation is diagnostic verification: 14/42 case-labelled genotyped records lack explicit “CAD” or “IHD” in their diagnosis text, and some list cerebrovascular or other cardiovascular conditions. These participants may still have coronary disease, but this cannot be demonstrated from the available records alone. For that reason, the findings do not show that all 42 cases have angiographically established CAD, nor do they distinguish acute coronary syndrome from stable CAD.

This is a single-center descriptive analysis rather than a fully validated individual-level clinical case–control study. The sample has only 12 genotyped controls, substantial age imbalance, 28 failed genotype calls, no Leu/Leu calls, no documented independent validation of genotyping, and heterogeneous case diagnostic descriptions. Recorded covariates are insufficient for meaningful multivariable adjustment. Standardized smoking and BMI variables and complete medication classification are lacking, while D-dimer is missing in 9/54 valid-genotype records. Neither the LDL assay methodology nor the D-dimer unit convention, analytical cut-off and

sampling time could be reconstructed from the records. The absence of funding also limited the ability to expand the cohort and perform additional confirmatory experiments, including Sanger sequencing.

5. Conclusion

In this cohort, FXIII-A Val34Leu genotypes were Val/Val in 26 and Val/Leu in 16 case-labelled records and Val/Val in 7 and Val/Leu in 5 controls. No Leu/Leu calls were present. The Leu34 allele frequency was 19.0% in cases and 20.8% in controls. The descriptive genotype comparison was imprecise (OR 0.86, 95% CI 0.23–3.18; Fisher $p = 1.000$) and cannot support conclusions about CAD risk. Source-record verification of phenotype, genotyping and assay procedures is essential, followed by replication in a larger, appropriately matched cohort.

Declarations

Author contributions: A.F.A.: principle author. S.M.H.: Researcher. M.B.A.: supervisor. All authors read and approved the final manuscript.

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Conflicts of interest: The authors declare no competing financial or personal interests.

Ethics approval and consent to participate: The study was approved by the Libyan Medical Research Center (approval no. NBC:018.H.24.64, dated 03 January 2025) and was conducted in accordance with the Declaration of Helsinki. Informed consent was obtained from all participants.

Data availability: The de-identified participant-level dataset is openly available in the Figshare repository at https://doi.org/10.6084/m9.figshare.35072461. Data sharing complies with the study's ethics approval and participant consent.

Use of generative AI: During the preparation of this work the authors used DeepSeek for assistance beyond spelling and grammar correction. The authors reviewed and edited the output and take full responsibility for the content of the publication.

Supplementary material: Not applicable.

References

[1] Jung JH, Song GG, Kim JH, Seo YH, Choi SJ. Association of factor XIII Val34Leu polymorphism and coronary artery disease: a meta-analysis. Cardiol J. 2017;24(1):74–84. doi:10.5603/CJ.a2016.0070

[2] Wang H, Liu Z, Shao J, Jiang M, Lu X, Lin L, et al. Pathogenesis of premature coronary artery disease: focus on risk factors and genetic variants. Genes Dis. 2022;9(2):370–80. doi:10.1016/j.gendis.2020.11.003

[3] Alshehri FSM, Whyte CS, Mutch NJ. Factor XIII-A: an indispensable “factor” in haemostasis and wound healing. Int J Mol Sci. 2021;22(6):3055. doi:10.3390/ijms22063055

[4] Bronić A, Ferenčak G, Bernat R, Leniček-Krleža J, Dumić J, Dabelić S. Association of fibrinogen and plasmin inhibitor, but not coagulation factor XIII gene polymorphisms with coronary artery disease. J Med Biochem. 2021;40(2):138–49. doi:10.5937/jomb0-26839

[5] Kattula S, Bagoly Z, Tóth NK, Muszbek L, Wolberg AS. The factor XIII-A Val34Leu polymorphism decreases whole blood clot mass at high fibrinogen concentrations. J Thromb Haemost. 2020;18(4):885–94. doi:10.1111/jth.14744

[6] Silvain J, Pena A, Vignalou JB, Hulot JS, Galier S, Cayla G, et al. FXIII-A Leu34 genetic variant in premature coronary artery disease: a genotype-phenotype case control study. Thromb Haemost. 2011;106(3):511–20. doi:10.1160/TH11-01-0027

[7] Sarecka-Hujar B, Łoboda D, Paradowska-Nowakowska E, Gołba KS. Coagulation factor XIII Val34Leu polymorphism in the prediction of premature cardiovascular events: the results of two meta-analyses. J Clin Med. 2022;11(12):3454. doi:10.3390/jcm11123454

[8] Wigginton JE, Cutler DJ, Abecasis GR. A note on exact tests of Hardy-Weinberg equilibrium. Am J Hum Genet. 2005;76(5):887–93. doi:10.1086/429864