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Biallelic Genetic Variant in the *DSG2* Gene Associated with Arrhythmogenic Right Ventricular Cardiomyopathy

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Abstract

Background: Arrhythmogenic right ventricular cardiomyopathy (ARVC) is a hereditary disorder characterized by fibrous or fibro-fatty myocardial tissue replacement leading to the life-threatening ventricular arrhythmias and progressive heart failure (HF). Most of the known genetic subtypes of ARVC, including the *DSG2*-associated form, are characterized by an autosomal dominant pattern of inheritance. Cases of compound heterozygous and digenic forms of ARVC are relatively rare and typically manifest in childhood or adolescence. The aim of this study is to characterize a rare biallelic form of ARVC caused by a homozygous mutation c.581C>T (p.Ser194Leu) in the *DSG2* gene based on our own and published data. **Methods**: The study cohort included 80 probands who underwent clinical examination and genetic testing using NGS of 16 genes. Haplotype analysis was performed for the probands homozygous for the *DSG2* c.581C>T (p.Ser194Leu) variant. **Results**: The rare form of ARVC was identified in three probands originating from the Republic of Dagestan and representing Dargin ethnic group. We assessed the frequency of the minor T allele in groups of 150 healthy volunteers, including 20 from the Republic of Dagestan, and 300 cardiomyopathy patients who are presumably natives of the North Caucasian Highland Region. The c.581C>T (p.Ser194Leu) variant was not detected in any of the DNA samples analyzed, indicating the rarity of this form of ARVC. The fact that three probands with the rare form of ARVC originate from the same minor ethnicity (Dargins) with a high rate of consanguineous marriages suggests a common ancestor for these families. All three probands shared a haplotype of approximately 4.01 Mb surrounding *DSG2* c.581C>T. **Conclusions**: Our study contributes to expanding the knowledge of the molecular causes of cardiomyopathies in poorly studied populations, underrepresented in international databases, and to establishing genotype–phenotypic correlations.

Keywords: arrhythmogenic right ventricular cardiomyopathy; ARVC; *DSG2*; founder effect; homozygosity; next-generation sequencing; DNA diagnostics; consanguineous marriage

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1. Introduction

Arrhythmogenic right ventricular cardiomyopathy (ARVC) is a hereditary multilocus disorder present in all ethnic groups. It is characterized by progressive fibrous or fibro-fatty replacement of the myocardium, leading to the life-threatening ventricular arrhythmias and progressive heart failure. The prevalence of ARVC in the European population is estimated to be approximately 1 per 1000–5000 individuals [1]) but can be much higher in selected ethnic groups [2].

In most familial cases, ARVC is inherited in an autosomal dominant manner, implying a high expected frequency of affected individuals among first-degree relatives [3]. The mutational spectrum of ARVC is highly heterogeneous and largely composed of numerous unique ("private") variants.

Recurrent pathogenic variants are relatively uncommon; however, several dominant mutations with a documented founder effect have been described. A well-known example is the p.S358L variant in the *TMEM43* gene identified in ARVC patients from Newfoundland (Canada), initially detected in members of 15 apparently unrelated families of English and Irish descent [4,5]. This mutation demonstrates complete penetrance and is associated with a severe clinical phenotype, especially in males [4,6]. Another example is the p.C796R variant in the *PKP2* gene, identified in nine unrelated patients, all of Dutch ancestry [5]. A further founder mutation, p.Arg14del in the *PLN* gene, was detected in the Netherlands in a cohort of 459 probands presenting with features of both dilated cardiomyopathy (DCM) (13% of probands) and ARVC (12% of probands) [7,8]. Haplotype analysis estimates the age of this mutation to be approximately 575–825 years [7]. In a study investigating the mutational spectrum of ARVC in Russian patients, no recurrent pathogenic variants or previously described founder mutations were identified [9].

Carriers of more than one causative variant generally exhibit a younger manifestation and a higher risk of life-threatening events compared with patients harboring a single pathogenic variant [10–12]. In reported cases of autosomal recessive forms, heterozygous carriers have minimal or no clinical symptoms [10,12].

With the advent of large-scale whole-exome (WES) and whole-genome (WGS) sequencing projects, it has become evident that the distribution of clinically significant variants in population samples is much higher than expected [13]. The authors suggested that the pathogenicity of a significant proportion of such variants may have been overestimated [13].

International databases, such as The Genome Aggregation Database (gnomAD), contains thousands of variants identified in presumably healthy volunteers from different populations. However, the clinical relevance of some variants might be occasionally downgraded as the volunteer enrolled to the study may have been asymptomatic at the time sampling. For example, in a study by Ramensky et al., the known pathogenic p.S358L variant in the *TMEM43* gene was identified in one individual with unknown clinical status from a population cohort of 1658 subjects from the Ivanovo region of Russia [14].

The prevalence of compound and digenic forms of ARVC in Russian multi-ethnic populations remains unexplored. Underrepresentation of the allele frequencies for selected ethnic groups in databases complicates the pathogenicity interpretation and genetic counseling.

The aim of the study was to characterize a rare biallelic form of ARVC associated with a homozygous mutation in the *DSG2* gene, based on our own and published data.

2. Materials and Methods

2.1. Clinical Evaluation and Genetic Counseling

The study cohort included 80 probands (36 males) diagnosed with ARVC in expert cardiology centers from 2018 to 2025 according to the timely available diagnostic criteria [15,16], and 16 first-degree relatives. The mean age of the probands at the time of referral for molecular genetic testing was 38.7 ± 14.4 years.

Clinical assessment included general examination, family history taking, resting and 24 h ECG recordings, transthoracic echocardiography, endomyocardial biopsy (when indicated), and cardiac MRI with gadolinium enhancement. All participants underwent genetic counseling prior to molecular testing.

2.2. Genetic Analysis

The genetic testing was approved by the Local Ethics Committee of the Petrovsky National Research Center of Surgery (Protocol No. 135) and was performed in accordance with the 1964 Helsinki declaration and later amendments.

Genomic DNA was extracted from venous blood using Quick-DNA Miniprep Plus Kit (Zymo Research Corp., Irvine, CA, USA) according to the manufacturer's instructions. Mutation screening of 5 desmosomal genes (*PKP2*, *DSG2*, *DSP*, *DSC2*, *JUP*) and 11 non-desmosomal genes (*TMEM43*, *LMNA*, *DES*, *TGFB3*, *PLN*, *SCN5A*, *CTNNA3*, *EMD*, *CRYAB*, *LDB3*, *FLNC*) was performed.

Sequencing was performed using high throughput semiconductor sequencing on an Ion PGM™ System (Thermo Fisher Scientific, Waltham, MA, USA) and bidirectional Sanger sequencing using an ABI 3730XL DNA Analyzer (Thermo Fisher Scientific), according to the manufacturer's instructions. The average amplicon length in the panel was 240 bp, with a mean coverage with at least 20 reads of 94.2% and a mean coverage with at least 100 reads of 79.25%.

Cascade family screening was performed by bidirectional Sanger sequencing or by PCR-RFLP analysis.

High-throughput sequencing data were processed using Torrent Suite Software (version 5.0.5), the Ion Reporter variant annotation platform (Thermo Fisher Scientific), and were visualized using the Integrative Genomic Viewer (IGV) tool [17] using hg38 as a reference genome. Variant interpretation was conducted in accordance with the criteria of the American College of Medical Genetics and Genomics (ACMG/AMP) [18] and ACGS Best Practice Guidelines for Variant Classification in Rare Disease 2024 [19]. The allele frequency (AF) was estimated using gnomAD v4.1.0 [20] and RuSeq [21].

2.3. Minor Allele Frequency Analysis of the c.581C>T Variant in the Cardiomyopathy Cohort and Healthy Volunteers

The minor allele frequency (MAF) of the *DSG2* c.581C>T variant using PCR-RFLP was analyzed in two groups:

One hundred and fifty unrelated adult heathy volunteers, including 20 individuals with established origin from the Republic of Dagestan. These participants reported no cardiac symptoms and had no family history of sudden cardiac death.

Three hundred probands who are presumably natives of the North Caucasian Highland Region diagnosed with primary cardiac disease.

2.4. SNP and STR Genotyping and Haplotype Analysis

Haplotype analysis was performed in three probands homozygous for the *DSG2* NM_001943.5:c.581C>T (p.Ser194Leu) variant. A panel of 17 markers spanning approximately 6.88 Mb of chromosome 18 flanking the *DSG2* gene region was analyzed, including

11 SNPs, five STR markers, and the target c.581C>T variant. All SNPs were genotyped by Sanger sequencing, while STRs were analyzed by multiplex PCR with FAM- and R6G-labelled primers followed by fragment analysis on an Applied Biosystems 3500 Genetic Analyzer (Thermo Fisher Scientific).

Genotypes were compared between the three probands to identify a conserved haplotype surrounding the disease-associated variant. We have estimated the genetic distance between the flanking markers of the shared haplotype using MareyMap Online [22]. The conserved core haplotype was defined by the most distant flanking markers showing identical alleles in all probands.

3. Results

We performed NGS-based genetic screening of 16 genes in 80 index cases diagnosed with ARVC. A total of 22 pathogenic (class V) or likely pathogenic (class IV) variants were identified in 21 probands. Most of the rare genetic variants were unique and heterozygous. The genetic variant NM_001943.5:c.581C>T (p.Ser194Leu) in exon 6 of the *DSG2* gene was detected in the homozygous state in three probands (NRF13-II-1, ARVD62-IV-1, ARVD120-V-1) from the Republic of Dagestan, and it was the only rare homozygous variant detected in whole ARVC cohort (Supplementary Figure S1).

All probands carrying c.581C>T (p.Ser194Leu) variant were Dargins, representatives of the indigenous ethnic group of the Northeast Caucasian population in the Republic of Dagestan. All were diagnosed with definite ARVC according to Task Force criteria [15,16] at a young age and shared similar clinical features (Table 1, Supplementary Figures S2 and S3).

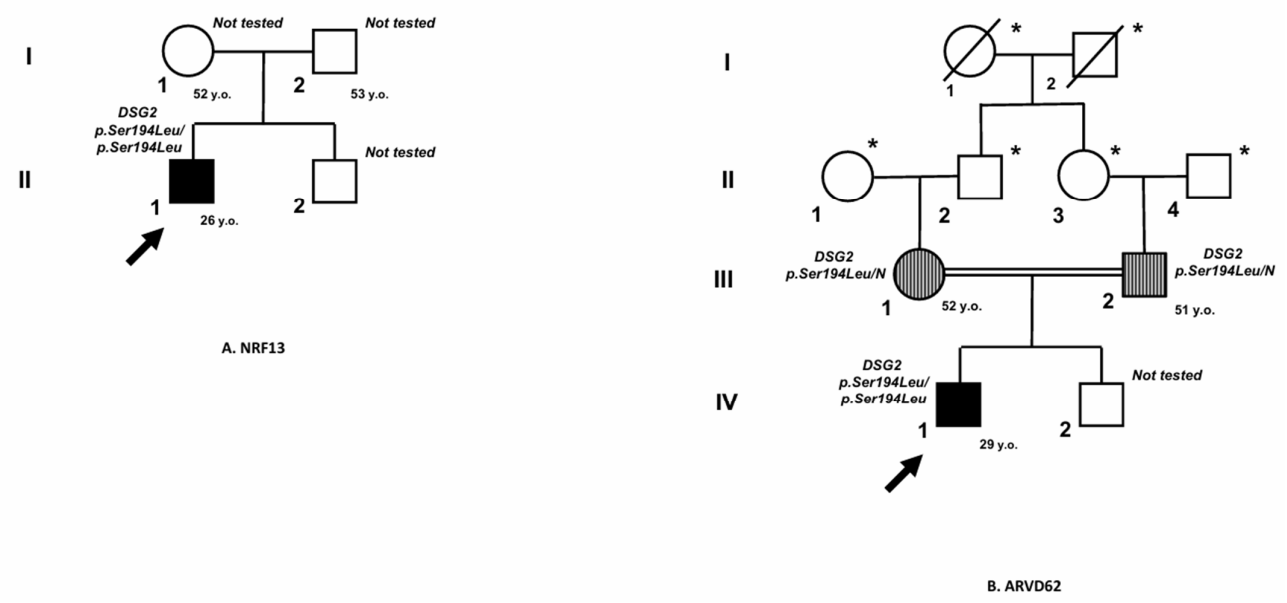
Table 1. Baseline characteristics of the probands who were homozygous carriers of the c.581C>T (p.Ser194Leu) variant in the *DSG2* gene.

	Family 1	**Family 2**	**Family 3**
Proband ID	NRF13-II-1	ARVD62-IV-1	ARVD120-V-1
Sex	Male	Male	Male
Age of genetic examination	26 y.o.	29 y.o.	17 y.o.
Age of symptoms onset	21 y.o.	13 y.o.	17 y.o.
ARVC diagnostic criteria	3 major, 2 minor (definite diagnosis)	3 major, 1 minor (definite diagnosis)	3 major, 1 minor (definite diagnosis)
Congenital heart defect	ASD (surgically repaired)	None	None
Sudden cardiac death in family	None	None	None
Parental consanguinity	No data	Confirmed	Confirmed
Cardiovascular disease in parents	Not reported	Not reported	Not reported
Arrhythmias and conduction abnormalities (resting ECG and 24 h Holter monitoring)	Low QRS voltage. Atrial flutter, PVC, incomplete RBBB, LV repolarization abnormalities, moderate ST depression, negative T-wave in V1–V3 leads	Low QRS voltage. Supraventricular ectopic beats, polymorphic PVC transient AVB (I). Negative T-wave in V1–V3 leads	Low QRS voltage with QRS fragmentation. Complete RBBB. Moderate ST depression, negative T waves in leads V1–V5. PVCs (5404 polymorphic; 18 non-sustained runs/day).

Structural and functional myocardial abnormalities (imaging studies)	Right heart dilation. Reduced RV systolic function. Severe (grade 3) tricuspid regurgitation. Signs of severe pulmonary hypertension. Inferior vena cava dilated to 3.4 cm	Right heart dilation; thrombus in the upper RV. Akinesia of the anterior RV wall (mid and apical segments); fibrosis of the IVS interventricular septum from the RV side throughout its length, as well as in the apex and anterior RV wall. Moderate (grade 2) tricuspid regurgitation	Thinning of the RV myocardium with fibrosis areas. Significant systolic and diastolic dysfunction of the RV (RVEF 18%) and LV (LVEF 23%); prominent apical LV trabeculation; dyskinetic motion of IVS. Moderate (grade 2) tricuspid regurgitation
Implantable cardioverter-defibrillator (ICD)	Yes (Implanted at age 26; appropriate shocks at ages of 30 and 31. RFA of atrial flutter at age of 34)	Yes (Implanted at age of 31; no data on shocks)	No (ICD recommended)

ASD—atrial septal defect, AVB—atrioventricular block; IVS—interventricular septum; LV—left ventricle, PVC—premature ventricular contraction, RBBB—right bundle branch block, RFA—radiofrequency ablation, RV—right ventricle.

In two families, the proband's parents were cousins; family data from the third proband were limited (Figure 1). All heterozygous carriers of the c.581C>T (p.Ser194Leu) variant in three families were asymptomatic. We classified the c.581C>T (p.Ser194Leu) variant as likely pathogenic (class IV) according to the ACMG (2015) criteria (PM2, PS3, PP1, PM3) (Supplementary Table S1).



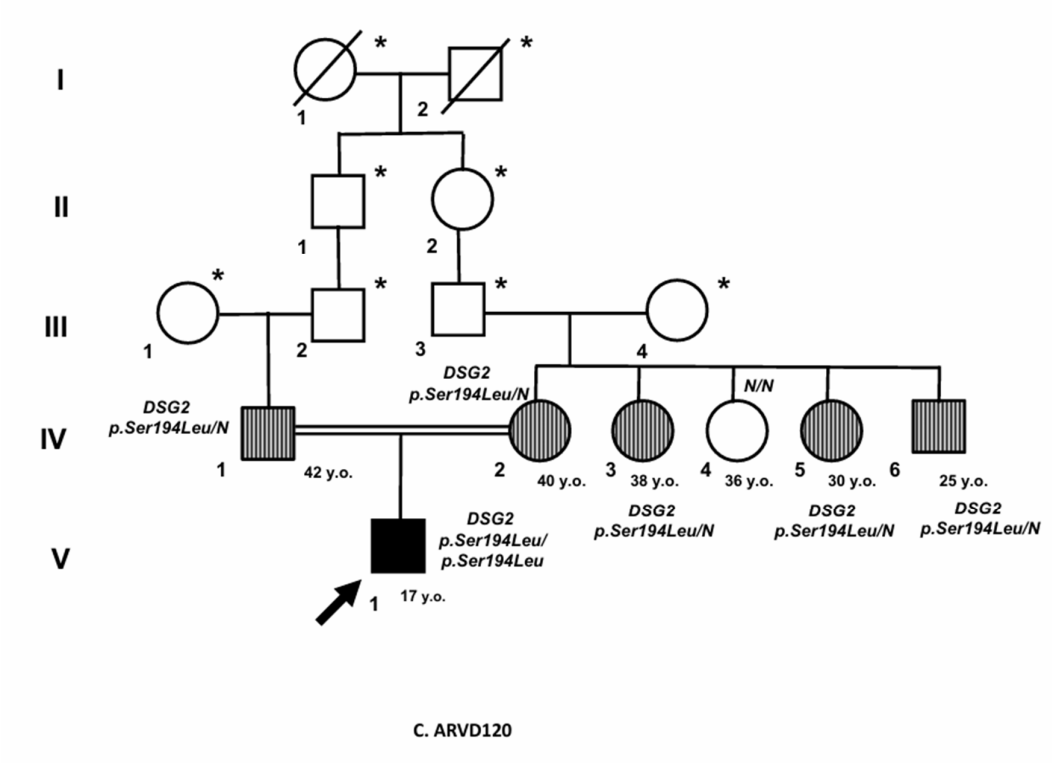


Figure 1. Pedigrees of three families. Probands are shown by closed symbols and marked by an arrow. Asymptomatic variant carriers are marked with a hatch. Symbol * means «not available».

We also analyzed the MAF, the c.581C>T (p.Ser194Leu) variant, in two groups of individuals. The c.581C>T (p.Ser194Leu) variant was not detected in a group of 150 healthy volunteers and in a group of 300 patients with primary cardiac diseases. This indicated the rarity of the c.581C>T allele in the Russian population.

We evaluated the c.581C>T variant representation in the clinical testing studies (Table 2) and in available databases. This variant was registered in the RUSeq database and detected in three individuals in a heterozygous state, two of whom were from the North Caucasian Highland Region, with one from the European part of Russia [21]. According to additional information from RUSeq curators, two of the three individuals carrying heterozygous variant had no cardiac complaints at the time of testing.

Table 2. Representation of the genetic variant c.581C>T (p.Ser194Leu) in the *DSG2* gene identified in clinical testing (present study and published data).

Source	Number of Identified Individuals	Genotype (Zygosity)	Phenotype	Origin/Ethnicity
Present study	3	Homozygous	ARVC	Republic of Dagestan/Dargins
	7	Heterozygous	Asymptomatic	Republic of Dagestan/Dargins
[23]	1	Compound heterozygous p.S194L/p.R292C (in trans)	ARVC	Japan
	2	Heterozygous	Asymptomatic mother and sibling	Japan

Laboratory for Molecular Medicine, Mass General Brigham Personalized Medicine (ClinVar Accession: SCV000060969.4)	1 (child)	n/a	ARVC, DCM	n/a
[24]	1	n/a	LVNC	n/a
[25]	2	Heterozygous	ARVC, left-dominant ACM	Israel (?)
	2	Heterozygous	Asymptomatic, mild phenotype?	Israel (?)

SNP and STR Haplotype Analysis

All three probands shared the same alleles at 12 consecutive markers from rs610906 to rs627346, including rs610906, rs529795, rs4800328, D18S847, D18S463, D18S56, rs3855547, rs930963, *DSG2* c.581C>T, D18S36, rs8096767, and rs627346. This conserved haplotype extended from approximately 3.46 Mb upstream to 0.56 Mb downstream of the variant of interest, corresponding to a physical length of the core haplotype approximately 4.01 Mb.

Outside this interval, discordant genotypes were observed at rs17224271, rs9966141, D18S457, rs12458025, and rs12232690. Thus, the complete 17-marker haplotype was not identical among the three probands, whereas a distinct conserved core surrounding *DSG2* was shared by all three (Table 3, Supplementary Figure S4). The 4.01 Mb haplotype corresponds to an estimated genetic distance of 4.23 cM.

Table 3. Haplotype analysis of the *DSG2* c.581C>T variant in three unrelated probands (P1–P3).

	Marker	Type	Position	Distance, Mb	P1	P2	P3
1	Rs17224271	SNP	18:25899906	−5.622	A/A	G/A	G/G
2	Rs9966141	SNP	18:27169016	−4.353	C/C	T/T	T/T
3	Rs610906	SNP	18:28067354	−3.455	A/A	A/A	A/A
4	Rs529795	SNP	18:28914047	−2.608	T/T	T/T	T/T
5	Rs4800328	SNP	18:29556712	−1.965	A/A	A/A	A/A
6	D18S847	STR	18:30123349	−1.399	221/221	221/221	221/221
7	D18S463	STR	18:30562228	−0.960	280/280	280/280	280/280
8	D18S56	STR	18:30715497	−0.807	505/505	505/505	505/505
9	Rs3855547	SNP	18:30960946	−0.561	T/T	T/T	T/T
10	Rs930963	SNP	18:31335073	−0.187	A/A	A/A	A/A
11	DSG2 c.581C>T	Variant	18:31522140	0	T/T	T/T	T/T
12	D18S36	STR	18:31584233	0.062	347/347	347/347	347/347
13	Rs8096767	SNP	18:31798660	0.277	T/T	T/T	T/T
14	Rs627346	SNP	18:32076993	0.555	T/T	T/T	T/T
15	D18S457	STR	18:32286279	0.764	219/223	223/223	223/223
16	Rs12458025	SNP	18:32505888	0.984	A/A	A/A	A/A
17	Rs12232690	SNP	18:32775910	1.254	A/C	A/A	C/C

Marker positions are given according to the GRCh38 reference genome, and distances are calculated relative to *DSG2* c.581C>T (0 Mb). Shaded cells indicate the conserved core haplotype shared by all three probands, extending from rs610906 to rs627346. The 4.01 Mb haplotype corresponds to an estimated genetic distance of 4.23 cM. SNP, single-nucleotide polymorphism; STR, short tandem repeat.

4. Discussion

The desmoglein protein encoded by the *DSG2* gene belongs to the desmosomal cadherins family. In a complex with the desmocollin protein, desmoglein constitute the two main transmembrane components of desmosomes, which are involved in the interaction between intermediate filaments and proteins mediating intercellular adhesion. Each member of the DSG protein family (types 1–4) consists of four extracellular cadherin domains and one transmembrane domain [26]. Desmoglein-2 is the only desmoglein isoform expressed in cardiomyocytes.

Mutations in the *DSG2* gene account for 3–20% of cases of ARVC in the disease structure [27,28]. Most of the known *DSG2* mutations cause autosomal dominant ARVC with or without decreased contractility, in some cases with the combined phenotype of ARVC/DCM or DCM [1]. Penetrance of *DSG2* mutations is variable and increases with age, and most (but not all) mutation carriers are symptomatic at the age of 65 [29–31].

The average age of clinical manifestation is approximately 30 years old, with patients with limited disease expression becoming clinically evident before age 65 [29–31]. Despite the marked clinical heterogeneity of ARVC, important genotype-specific patterns are observed even within the classical desmosomal form of the disease [32]. Currently, an oligogenic model of inheritance is also being considered, which may better explain the incomplete penetrance and variable expressivity of ARVC [23,27,33].

Several clinical reports have described an autosomal recessive form of ARVC in which homozygous carriers exhibited a classical disease phenotype, whereas heterozygous carriers were either asymptomatic or had a mild phenotype [10,12]. The homozygous c.1592T>G (p.Phe531Cys) variant in the *DSG2* gene was identified only in patients from East Asia (China, Taiwan, Japan). Genetic analysis of Chinese ARVC patients revealed that eight homozygous *DSG2* p.Phe531Cys mutation probands shared a haplotype spanning >450 kb [10]. Since all unrelated patients shared the same haplotype [10], the "founder effect" was confirmed [10,12].

We revealed the homozygous genetic variant NM_001943.5:c.581C>T (p.Ser194Leu) in the *DSG2* gene in three probands with ARVC. This alteration affects the second extracellular cadherin domain, responsible for Ca2+-dependent interactions with both desmosomal and classical cadherins. At the time of genetic counseling, the probands were young adults and similar to the typical age of onset of the classical autosomal dominant form of ARVC. The primary symptoms and severity of the disease in these patients were also similar (Table 1).

According to the ClinVar database (https://www.ncbi.nlm.nih.gov/clinvar, accessed 17 August 2026), the interpretation of the c.581C>T (p.Ser194Leu) variant was ambiguous. The combined classification based on six interpretations entered in the database assigned the status of "variant of unknown significance". In silico analysis provided conflicting evidence for the effect of the variant on the gene or gene product. The minor allele frequency (MAF) in the total population according to the GnomAD database (v4.1.0) was 9.296×10^{-6} (15/1,613,684), and all 15 carriers (12 males) were heterozygous. In the non-Finnish European population, the variant frequency was 4.238×10^{-6} (5/1,179,810). The highest prevalence was observed in the South Asian population, with a frequency of 8.785×10^{-5}.

The c.581C>T (p.Ser194Leu) variant is located within a "hotspot" region (codons 24–388) described by Kapplinger et al. for missense mutations in the N-terminal domain of the DSG2 gene [5]. It represents a highly conserved region of desmoglein-2 (Supplementary Figure S5). In the study by Blich et al. (2023) [25], there was structural distortion of the intercalated disc in myocardium harboring the p.Ser194Leu variant compared with controls, and reduced expression of desmoglein-2 and connexin 43 was observed both in the cytoplasm and at intercalated discs [25].

Bi-allelic inheritance associated with variant c.581C>T (p.Ser194Leu) in trans with another missense variant p.R292C, has been described in a Japanese patient, who was 38 y.o., with a definite ARVC diagnosis [23]. In that case, the patient's parents and sister were asymptomatic and obligate carriers of only one of the *DSG2* variants.

It was also described in a family with arrhythmogenic cardiomyopathy predominantly affecting the left ventricle in the proband and the right ventricle in his brother [25]. Histological examination of the myocardial proband's tissue demonstrated a reduced number of cardiomyocytes with fibrofatty replacement in the right ventricular endocardium, disorganization of intercalated discs, disruption of desmosomal junctions with widening of intercellular spaces, a marked decrease in DSG2 and GJA1 expression both in the cytoplasm and at cell junctions, as well as perinuclear accumulation of DSG2 and GJA1 [25].

Taking into account the variant's location, population frequency, and its cosegregation with disease in the family, the homozygous c.581C>T (p.Ser194Leu) variant in *DSG2* is classified as likely pathogenic (class IV) according to the ACMG criteria. Given the same origin and ethnicity of our probands (Dargins, the Republic of Dagestan), we assumed a common ancestral origin of the alleles.

Haplotype analysis revealed a shared allelic background surrounding DSG2:c.581C>T (p.Ser194Leu) in all three unrelated probands. The shared haplotype extended across approximately 4.01 Mb and included multiple consecutive SNP and STR markers located on both sides of the variant. In contrast, the flanking markers at the outer boundaries of the analyzed region showed more allelic variation between probands, supporting the presence of a conserved central haplotypic segment. The concordance of SNP and STR profiles across this interval supports the inheritance of DSG2:c.581C>T on a common ancestral chromosomal background and is consistent with a possible founder effect.

A founder-associated haplotype has previously been reported for another DSG2 variant, c.1592T>G (p.Phe531Cys), in an East Asian population. However, the haplotype reported for p.Phe531Cys was approximately 450 kb in length and differed from the approximately 4 Mb haplotype identified in the present study. The markedly different extent and allelic composition of these haplotypes suggest that DSG2:c.581C>T and c.1592T>G arose from distinct ancestral chromosomal backgrounds, consistent with independent founder events. The substantially longer shared haplotype observed for DSG2:c.581C>T may indicate a more recent common ancestral origin of its carriers; however, direct comparison of haplotype lengths should be interpreted cautiously, because haplotype boundaries are influenced by marker density, genetic distance, recombination rates, and the criteria used for defining the shared segment.

The shared haplotype observed in the three unrelated probands may be consistent with a common ancestral origin of DSG2:c.581C>T and raises the possibility of a founder effect. This hypothesis should be further evaluated in a larger cohort of unrelated carriers and population-matched controls, which may help determine the extent and population frequency of the shared haplotype and provide a more reliable estimate of the time to the most recent common ancestor.

In several studies, consanguinity between parents carrying heterozygous *DSG2* variants was either presumed based solely on the proband's report (due to limited communication with the parents) [12] or confirmed by single-nucleotide polymorphism (SNP) analysis [11].

For founder mutations in *DSG2*, differences in clinical manifestations between homozygous and heterozygous carriers have been demonstrated. In particular, the penetrance of ARVC among homozygous carriers of the p.Phe531Cys mutation was 100%, whereas only 25% of heterozygous carriers developed mild symptoms at an older age. These findings were reproduced in Chinese and Japanese cohorts, in which a higher carrier frequency of the mutant allele was observed [12].

Interestingly, the age of the first symptoms associated with the biallelic form of ARVC ranged from 14 to 34 years old (mean age 24.5 ± 5.9) [10], overlapping with the mean age of the manifestation of monoallelic forms of ARVC. This overlap may complicate accurate determination of the mode of inheritance.

In the study by Nakajima et al. regarding the p.Ser194Leu variant, as well as in reports describing other *DSG2* variants, it was shown that disease development may result from compound heterozygous biallelic variants in *DSG2* or from digenic inheritance. In such cases, carriers of only one of the two variants are typically asymptomatic, and the disease manifests only in individuals harboring two rare variants. These rare variants are often classified as variants of uncertain significance (class III), which limits their immediate clinical utility and does not allow for definitive clinical decision-making.

The North Caucasian Highland Region contains approximately 110 ethnic groups belonging to Caucasians, who are largely underrepresented in global genetic databases. Representatives of over 30 indigenous ethnicities live in the Republic of Dagestan over an area of 50,270 square kilometers and characterized by a high inbreeding rate. Dargins is the second largest ethnic group in this area, and its population is more than 600,000 in Russia.

Highly inbred populations remain insufficiently studied; however, the prevalence of recessively inherited disorders may be associated with inbreeding in geographically isolated regions [34]. In a study by Magomedova investigating progressive muscular dystrophies in the populations of the Republic of Dagestan, autosomal recessive forms were shown to predominate among residents of two mountainous isolates [35]. The high frequency of rare cardiomyopathy-associated gene variants among patients from the Republic of Dagestan indicates the need for detailed epidemiological study in order to optimize genetic counseling, DNA diagnostics, and health care taking into account ethnic and cultural diversity of populations.

5. Conclusions

We identified a biallelic mutation c.581C>T (p.Ser194Leu) in the *DSG2* gene in three young Darginian probands with arrhythmogenic right ventricular cardiomyopathy from the Republic of Dagestan. The identification of three probands from the same ethnic group with a rare recessive form of cardiomyopathy in certain regions may be explained by a possible founder effect and a high frequency of consanguinity.

Our findings expand the spectrum of molecular causes of cardiomyopathies in understudied populations and suggest that further research is needed to identify and characterize the genetic causes of cardiomyopathy in different regions of Russia.

Limitations: Genetic testing was limited to a 16-gene panel with lack of possible CNVs, structural variants or variants outside the regions covered by the panel.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/genes17111392/s1. Figure S1: A. Homozygous c.581C>T (p.Ser194Leu) substitution in exon 6 of the *DSG2* gene in proband ARVD120 V-1 B. Heterozygous c.581C>T (p.Ser194Leu) substitution in proband's mother ARVD120 IV-2. Figure S2: Fragment of a single-lead ECG of ARVD62-IV-1. Figure S3: Contrast-enhanced cardiac MRI (Omniscan) of proband ARVD62-IV-1. Figure S4: Electropherograms of STR markers analyzed in the three probands (P1, P2, and P3). Figure S5: Comparison of the Desmoglein-2 amino acid sequences across different species. Table S1. Variant interpretation according to the ACMG (2015) and ACGS (2024) criteria.

Author Contributions: A.G.S.: conceptualization, data curation, genetic investigation, methodology, validation, writing—original draft. O.V.B., Y.A.L., A.P.M., A.A.N., M.A.A., E.A.T., M.V.T. and S.L.D.: clinical investigation, writing—review and editing. V.P.P.: genetic investigation, methodology, validation, writing—original draft. E.V.Z.: data curation, investigation (genetic counseling),

funding acquisition, project administration, writing—original draft, review and editing. All authors have read and agreed to the published version of the manuscript.

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Abbreviations

The following abbreviations are used in this manuscript:

ARVC	Arrhythmogenic right ventricular cardiomyopathy
WES	Whole-exome sequencing
WGS	Whole-genome sequencing
AF	Allele frequency
MAF	Minor allele frequency
DCM	Dilated cardiomyopathy

References

1. Corrado, D.; Link, M.S.; Calkins, H. Arrhythmogenic right ventricular cardiomyopathy. *N. Engl. J. Med.* **2017**, *376*, 61–72.
2. Protonotarios, A.; Asimaki, A.; Basso, C.; Xylouri, Z.; Monda, E.; Protonotarios, I.; Crisci, G.; Abrahms, D.J.; Anastasakis, A.; Antoniades, L.; et al. Naxos Disease and Related Cardio-Cutaneous Syndromes. *JACC Adv.* **2025**, *4*, 101547. https://doi.org/10.1016/j.jacadv.2024.101547. PMID: 39877668; PMCID: PMC11773020.
3. Oomen, A.W.G.J.; Semsarian, C.; Puranik, R.; Sy, R.W. Diagnosis of arrhythmogenic right ventricular cardiomyopathy: Progress and pitfalls. *Heart Lung Circ.* **2018**, *27*, 1310–1317.
4. Merner, N.D.; Hodgkinson, K.A.; Haywood, A.F.; Connors, S.; French, V.M.; Drenckhahn, J.D.; Kupprion, C.; Ramadanova, K.; Thierfelder, L.; McKenna, W.; et al. Arrhythmogenic right ventricular cardiomyopathy type 5 is a fully penetrant, lethal arrhythmic disorder caused by a missense mutation in the TMEM43 gene. *Am. J. Hum. Genet.* **2008**, *82*, 809–821. https://doi.org/10.1016/j.ajhg.2008.01.010. PMID: 18313022; PMCID: PMC2427209.
5. Kapplinger, J.D.; Landstrom, A.P.; Salisbury, B.A.; Callis, T.E.; Pollevick, G.D.; Tester, D.J.; Cox, M.G.; Bhuiyan, Z.; Bikker, H.; Wiesfeld, A.C.; et al. Distinguishing arrhythmogenic right ventricular cardiomyopathy/dysplasia-associated mutations from background genetic noise. *J. Am. Coll. Cardiol.* **2011**, *57*, 2317–2327. https://doi.org/10.1016/j.jacc.2010.12.036.
6. Hodgkinson, K.A.; Connors, S.P.; Merner, N.; Haywood, A.; Young, T.L.; McKenna, W.J.; Gallagher, B.; Curtis, F.; Bassett, A.S.; Parfrey, P.S. The natural history of a genetic subtype of arrhythmogenic right ventricular cardiomyopathy caused by a p.S358L mutation in TMEM43. *Clin. Genet.* **2013**, *83*, 321–331. https://doi.org/10.1111/j.1399-0004.2012.01919.x.
7. van der Zwaag, P.A.; van Rijsingen, I.A.W.; Asimaki, A.; Jongbloed, J.D.H.; van Veldhuisen, D.J.; Wiesfeld, A.C.P.; Cox, M.G.P.J.; van Lochem, L.T.; de Boer, R.A.; Hofstra, R.M.W.; et al. Phospholamban R14del mutation in patients diagnosed with dilated

cardiomyopathy or arrhythmogenic right ventricular cardiomyopathy: Evidence supporting the concept of arrhythmogenic cardiomyopathy. *Eur. J. Heart Fail.* **2012**, *14*, 1199–1207.

8. van der Heijden, J.F.; Hassink, R.J. The phospholamban p.Arg14del founder mutation in Dutch patients with arrhythmogenic cardiomyopathy. *Neth. Heart J.* **2013**, *21*, 284–285.
9. Shestak, A.G. Molecular Genetic Polymorphism of Arrhythmogenic Right Ventricular Cardiomyopathy in Russian Patients. Ph.D. Thesis, Moscow, Russia, 2022; 149p. EDN QCIOUS. (In Russian)
10. Chen, L.; Rao, M.; Chen, X.; Chen, K.; Ren, J.; Zhang, N.; Zhao, Q.; Yu, W.; Yuan, B.; Song, J. A founder homozygous DSG2 variant in East Asia results in ARVC with full penetrance and heart failure phenotype. *Int. J. Cardiol.* **2019**, *274*, 263–270.
11. Brodehl, A.; Meshkov, A.; Myasnikov, R.; Kiseleva, A.; Kulikova, O.; Klauke, B.; Sotnikova, E.; Stanasiuk, C.; Divashuk, M.; Pohl, G.M.; et al. Hemi- and Homozygous Loss-of-Function Mutations in DSG2 (Desmoglein-2) Cause Recessive Arrhythmogenic Cardiomyopathy with an Early Onset. *Int. J. Mol. Sci.* **2021**, *22*, 3786. https://doi.org/10.3390/ijms22073786.
12. Murakami, H.; Tanimoto, Y.; Tanimoto, K.; Inoue, S.; Ishikawa, T.; Makita, N.; Yamazawa, K. Arrhythmogenic right ventricular cardiomyopathy in a Japanese patient with a homozygous founder variant of DSG2 in the East Asian population. *Hum. Genome Var.* **2022**, *9*, 28. https://doi.org/10.1038/s41439-022-00206-9.
13. Andreasen, C.; Nielsen, J.B.; Refsgaard, L.; Holst, A.G.; Christensen, A.H.; Andreasen, L.; Sajadieh, A.; Haunsø, S.; Svendsen, J.H.; Olesen, M.S. New population-based exome data are questioning the pathogenicity of previously cardiomyopathy-associated genetic variants. *Eur. J. Hum. Genet.* **2013**, *21*, 918–928. https://doi.org/10.1038/ejhg.2012.283.
14. Ramensky, V.E.; Ershova, A.I.; Zaicenoka, M.; Kiseleva, A.V.; Zharikova, A.A.; Vyatkin, Y.V.; Sotnikova, E.A.; Efimova, I.A.; Divashuk, M.G.; Kurilova, O.V.; et al. Targeted sequencing of 242 clinically important genes in the Russian population from the Ivanovo region. *Front. Genet.* **2021**, *12*, 709419. https://doi.org/10.3389/fgene.2021.709419.
15. Corrado, D.; Perazzolo Marra, M.; Zorzi, A.; Beffagna, G.; Cipriani, A.; Lazzari, M.; Migliore, F.; Pilichou, K.; Rampazzo, A.; Rigato, I.; et al. Diagnosis of arrhythmogenic cardiomyopathy: The Padua criteria. *Int. J. Cardiol.* **2020**, *319*, 106–114. https://doi.org/10.1016/j.ijcard.2020.06.005. PMID: 32561223.
16. Corrado, D.; Anastasakis, A.; Basso, C.; Bauce, B.; Blomström-Lundqvist, C.; Bucciarelli-Ducci, C.; Cipriani, A.; De Asmundis, C.; Gandjbakhch, E.; Jiménez-Jáimez, J.; et al. Proposed diagnostic criteria for arrhythmogenic cardiomyopathy: European Task Force consensus report. *Int. J. Cardiol.* **2024**, *395*, 131447. https://doi.org/10.1016/j.ijcard.2023.131447. PMID: 37844667.
17. Robinson, J.T.; Thorvaldsdóttir, H.; Winckler, W.; Guttman, M.; Lander, E.S.; Getz, G.; Mesirov, J.P. Integrative genomics viewer. *Nat. Biotechnol.* **2011**, *29*, 24–26. https://doi.org/10.1038/nbt.1754. PMID: 21221095; PMCID: PMC3346182.
18. Richards, S.; Aziz, N.; Bale, S.; Bick, D.; Das, S.; Gastier-Foster, J.; Grody, W.W.; Hegde, M.; Lyon, E.; Spector, E.; et al. Standards and guidelines for the interpretation of sequence variants: A joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet. Med.* **2015**, *17*, 405–424. https://doi.org/10.1038/gim.2015.30.
19. Durkie, M.; Cassidy, E.-J.; Berry, I.; Owens, M.; Turnbull, C.; Scott, R.H.; Taylor, R.W.; Deans, Z.C.; Ellard, S.; Baple, E.L.; et al. *ACGS Best Practice Guidelines for Variant Classification in Rare Disease 2024*; Association for Clinical Genomic Science: London, UK, 2024.
20. Chen, S.; Francioli, L.C.; Goodrich, J.K.; Collins, R.L.; Kanai, M.; Wang, Q.; Alföldi, J.; Watts, N.A.; Vittal, C.; Gauthier, L.D.; et al. A genomic mutational constraint map using variation in 76,156 human genomes. *Nature* **2024**, *625*, 92–100. Correction in *Nature* **2024**, *626*, E1. https://doi.org/10.1038/s41586-023-06045-0.
21. Barbitoff, Y.A.; Khmelkova, D.N.; Pomerantseva, E.A.; Slepchenkov, A.V.; Zubashenko, N.A.; Mironova, I.V.; Kaimonov, V.S.; Polev, D.E.; Tsay, V.V.; Glotov, A.S.; et al. Expanding the Russian allele frequency reference via cross-laboratory data integration: Insights from 7452 exome samples. *Natl. Sci. Rev.* **2024**, *11*, nwae326. https://doi.org/10.1093/nsr/nwae326.
22. Siberchicot, A.; Bessy, A.; Guéguen, L.; Marais, G. MareyMap Online: A User-Friendly Web Application and Database Service for Estimating Recombination Rates Using Physical and Genetic Maps. *Genome Biol. Evol.* **2017**, *9*, 2506–2509. https://doi.org/10.1093/gbe/evx178.
23. Nakajima, T.; Kaneko, Y.; Irie, T.; Takahashi, R.; Kato, T.; Iijima, T.; Iso, T.; Kurabayashi, M. Compound and digenic heterozygosity in desmosome genes as a cause of arrhythmogenic right ventricular cardiomyopathy in Japanese patients. *Circ. J.* **2012**, *76*, 737–743. https://doi.org/10.1253/circj.cj-11-0927.
24. Mazzarotto, F.; Hawley, M.H.; Beltrami, M.; Beekman, L.; de Marvao, A.; McGurk, K.A.; Statton, B.; Boschi, B.; Girolami, F.; Roberts, A.M.; et al. Systematic large-scale assessment of the genetic architecture of left ventricular noncompaction reveals diverse etiologies. *Genet. Med.* **2021**, *23*, 856–864. https://doi.org/10.1038/s41436-020-01049-x.

25. Blich, M.; Zohar, Y.; Cohen-Kaplan, V.; Minkov, I.; Asleh, R.; Horowitz-Cederboim, S.; Weiss, K.; Paperna, T.; Lessick, J.; Abadi, S.; et al. Ser194Leu DSG2 mutation, associated with arrhythmogenic left ventricular cardiomyopathy and ventricular tachycardia. *Pacing Clin. Electrophysiol.* **2024**, *47*, 503–510. https://doi.org/10.1111/pace.14950.
26. Saito, M.; Tucker, D.K.; Kohlhorst, D.; Niessen, C.M.; Kowalczyk, A.P. Classical and desmosomal cadherins at a glance. *J. Cell Sci.* **2012**, *125*, 2547–2552.
27. Pourebrahim, K.; Marian, J.G.; Tan, Y.; Chang, J.T.; Marian, A.J. A combinatorial oligogenic basis for the phenotypic plasticity between late-onset dilated and arrhythmogenic cardiomyopathy in a single family. *J. Cardiovasc. Aging* **2021**, *1*, 12. https://doi.org/10.20517/jca.2021.15.
28. Garcia-Montero, M.; Fanous, Y.; Krahn, A.D.; Davies, B.; Cadrin-Tourigny, J.; Roberts, J.D. New insights into genetic right ventricular cardiomyopathies. *Can. J. Cardiol.* **2025**, *41*, 1023–1037. https://doi.org/10.1016/j.cjca.2025.02.020.
29. Cho, Y. Arrhythmogenic right ventricular cardiomyopathy. *J. Arrhythm.* **2018**, *34*, 356–368.
30. DeWitt, E.S.; Chandler, S.F.; Hylind, R.J.; Beausejour Ladouceur, V.; Blume, E.D.; VanderPluym, C.; Powell, A.J.; Fynn-Thompson, F.; Roberts, A.E.; Sanders, S.P.; et al. Phenotypic manifestations of arrhythmogenic cardiomyopathy in children and adolescents. *J. Am. Coll. Cardiol.* **2019**, *74*, 346–358. https://doi.org/10.1016/j.jacc.2019.05.022.
31. Stevens, T.L.; Wallace, M.J.; Refaey, M.E.; Roberts, J.D.; Koenig, S.N.; Mohler, P.J. Arrhythmogenic cardiomyopathy: Molecular insights for improved therapeutic design. *J. Cardiovasc. Dev. Dis.* **2020**, *7*, 21. https://doi.org/10.3390/jcdd7020021.
32. Chen, L.; Hu, Y.; Saguner, A.M.; Bauce, B.; Liu, Y.; Shi, A.; Guan, F.; Chen, Z.; Bueno Marinas, M.; Wu, L.; et al. Natural History and Clinical Outcomes of Patients With *DSG2*/*DSC2* Variant-Related Arrhythmogenic Right Ventricular Cardiomyopathy. *Circulation* **2025**, *151*, 1213–1230. https://doi.org/10.1161/CIRCULATIONAHA.124.072226.
33. James, C.A.; Calkins, H. Arrhythmogenic right ventricular cardiomyopathy: Evidence for progression increases. *Eur. Heart J.* **2020**, *41*, 1411–1413.
34. Bulayeva, K.B.; Jorde, L.; Ostler, C.; Bulayev, O.A.; Pavlova, T.A.; Harpending, H. STR Polymorphism in Indigenous Daghestan Populations. *Russ. J. Genet.* **2004**, *40*, 558–569. https://doi.org/10.1023/B:RUGE.0000029160.56297.eb.
35. Magomedova, R.M. Clinical and Genetic Study of Progressive Muscular Dystrophies in the Population of the Republic of Dagestan. Ph.D. Thesis, Moscow, Russia, 2015; 128 p. EDN OEFUXI (In Russian)

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