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Research Article

**FREQUENCY OF R1193Q MUTATION IN SCNSA GENE ON  
SUSCEPTIBILITY FOR ARVC AND LQTS IN IRAN**

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**Abstract:**

*Cardiomyopathy, a disease of the heart muscle, is associated with cardiac dysfunction and results to severe heart failure, arrhythmias, and death. Arrhythmogenic right ventricular cardiomyopathy (ARVC) is a familial cardiomyopathy that is characterized by localized movement disorder in the wall of the right ventricle. Long QT syndrome (LQTS) is typical cardiac arrhythmia that causes hereditary or acquired cardiac arrest in patients with long QT interval and can be identified by electrocardiogram. It has been elucidated that R1193Q Polymorphism (R1193Q GGG> CAG, rs41261344) is a functional substitution in sodium voltage-gated channel alpha subunit 5 (SCN5A) gene, and correlates with cardiac arrhythmia syndromes such as ARVC and LQTS. This case-control study was conducted to consider the correlation between SCN5A rs41261344 and cardiac arrhythmia syndromes in Iranian population. In the present study, 40 patients with LQTS, and 40 patients with ARVC, and 80 healthy subjects as controls were analyzed for SCN5A rs41261344 by polymerase chain reaction- restriction fragment length polymorphism (RFLP-PCR) and 10 samples from each group were sequenced. The SCN5A rs41261344 C allele with a frequency of 100% was present in all the patients and the healthy subjects. However, we did not observe SCN5A rs41261344 T allele in our enrolled subjects. In fact, the frequency of T allele was calculated zero. Regard to very low frequency of the R1193Q polymorphism minor allele, SCN5A rs41261344 T allele, in our cohort, it can be suggested that other mutations may be associated with cardiac arrhythmia syndromes such as ARVC and LQTS in Iranian population.*

**Key Words:** *Cardiomyopathy, RFLP PCR, R1193Q polymorphism, SCN5A, ARVC, Long QT.***Corresponding author:****Arezoo Sabbagh Hadi,**Yong researchers and Elite club,  
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**INTRODUCTION:**

Cardiovascular diseases, is the cause of 12 million fetal deaths annually in the United States of American and it's the disorders of heart and blood vessels, are the main cause of mortality in modern societies [1]. Cardiomyopathy is described as a myocardial disorder in heart muscles is structurally abnormal and results in severe heart failure, arrhythmias (heart failure or irregular heartbeats), and finally death. The different cardiomyopathy types can be classified into the following categories: restrictive cardiomyopathy, hypertrophic cardiomyopathy and unclassified cardiomyopathy [2]. ARVC is a familial right ventricular dysplasia that is characterized by dysfunctionality of the muscular wall of the right ventricle. Moreover, this disorder commonly is indicated in males [3-5]. Imaging modalities commonly used for ARVC assessment contain echocardiography, cardiovascular magnetic resonance (CMR), and RV angiography [6] structural unnatural and right ventricular dysfunction have been confirmed in most of the ARVC patients [2] SCN5A is the candidate gene for ARVC about 30 to 50% of patients with ARVC have a family history of the disease. In addition, some of the ARVC patients with autosomal recessive pattern of inheritance have been reported with Naxos disease and Carvajal syndrome. [7-12] The SCN5A gene is located on chromosome 3p21.24 and belongs to a gene family that are coding the components of sodium channels. It has been shown that SCN5A gene expression leads to diverse isoforms of sodium channels i.e. brain-type, neuronal channels, skeletal muscle channels, and the cardiac sodium channel Nav1.5. These channels in heart play main roles in signaling of the start of each heartbeat, maintaining a normal heart rhythm and coordinating the contractions of the upper and lower chambers of the heart. The SCN5A gene contains 28 exons of which the exons 2 to 28 constitute the protein-coding sequence. More than 10 different splice isoforms have been described for SCN5A gene [13, 14] SCN5A splice variants, Nav1.5, functions as an electric transmission and produces potential impulses in the heart myocardium [15-17]. Also, the SCN5A gene variants might impact on different channel mechanisms, including its activation, inactivation, and reactivation. Mutations that impaired the Nav1.5 channel  $\alpha$ -subunit are known to be considered as risk factors for arrhythmia. It is including 6% of patient with LQTS subtype 3 (LQTS3) and 10 to 20% of patient with Brugada Syndrome [18-26]. Moreover, some mutations of SCN5A gene either result in the biophysical deficiencies or are associated with an increased risk of sudden cardiac death. The R1193Q polymorphism (CGG>CAG, refSNP ID rs41261344), for instance, in SCN5A gene is the underpinning reason of approximately 13% of patients

with LQTS. It is hypothesized that R1193Q polymorphism might increase the susceptibility to LQTS as a result of disturbing the sodium channel function [27]. Actually, R1193Q polymorphism, a missense mutation, results in the replacement of arginine with glutamine at position 1193. Thus, this alteration results in a non-conservative status which consequently causes inactivation of sodium channel current [28-30].

**MATERIALS AND METHODS:****Sampling**

DNA samples randomly selected from subjects referred to Rajaie cardiovascular medical and research center. Written consent was obtained from all participants prior to blood sampling. 5 ml peripheral whole blood was taken from each patient (40 patients with ARVC, 40 patients with LQTS and 80 healthy controls). All the patients had a familial history of heart disease, heart failure or an unexpected sudden cardiac death. The investigation included a review of clinical history, echocardiography, complete physical examination, and standard 12-lead ECG measured at rest. In some cases, a 24-h ambulatory ECG was measured as well. The normal range of the ECG measurements was based on the age of each person.

**DNA extraction**

Total genomic DNA was extracted from 5 ml peripheral whole blood. A salt precipitation method (salting out) was used to extract of DNA from each sample [31]. The purity and concentration of DNA was measured by using of Nano-drop device.

**PCR amplification and analyses**

Coding region (R1193Q) and intron-exon boundary in SCN5A gene were amplified by using the following primers:

Forward primer (CAGGGAAGGTCTGGTGGC)

Reverse primer (TCCTACTCAGCAGTGGAGCG)

The PCR reactions were carried out in a 25  $\mu$ l total volume containing: 1  $\mu$ l of DNA genomic, 3  $\mu$ l of master mix (Taq polymerase, MgCl<sub>2</sub>, Buffer), 0.5  $\mu$ l of forward primer, 0.5  $\mu$ l of reverse primer, and 20  $\mu$ l of distilled water. The cycles were as: 1 cycle at 95°C for 5 min; 30 cycles (95°C for 30 secs, 65°C for 30 secs, 72°C for 35 secs, 72°C for 5 min). 5 ml of the PCR product was expanded at 100V on 1% agarose gel electrophoresis. Then RFLP technique was done using ACII restriction enzyme and fragments were separated. Data confirmation of 10% of samples was done by sequencing assay (Figure 1) then, the data were analyzed.

**FINDING:**

Ischemic heart disease and heart failure is the most common causes of sudden cardiac death. However, in 5 to 8 % of sudden cardiac death victims, no evidence of

structural heart diseases was found. Ion channel disorders constitute 35% of causes of sudden unexplained death (SUD) in young people and 9% of sudden infant death syndrome (SIDS) that which was the largest LQTS [26, 32]. The R1193Q minor allele, T allele, was not observed in the cases and controls. Overall, 160 people studied population was composed of 80 patients and 80 healthy controls. a R1193Q mutation was not found in any of the 80 normal subjects and 80 patients, Genotypic distribution of R1193q polymorphism in CC Genotype (healthy) in all of cases (ARVC cases, Long QT cases, Control cases) was %100, also Genotypic distribution of R1193q polymorphism in CT, TT Genotype(mutant) in all of cases (ARVC cases, Long QT cases, Control cases) was %0. Also, allelic frequency of R1193q polymorphism in C (Healthy) allelic for Arvc cases was 100%, for Long QT cases 100%, also for Control cases was 100%, also allelic frequency of R1193q polymorphism for T allelic(Mutant) in Arvc cases was 0, in Long QT cases 0 and in control cases was 0. In our study, the results showed that, contrary to previous observations in Asian countries, R1193Q polymorphism is not Iran's population common polymorphisms. According to this result, suggesting Contrary to the previous observations in Asian countries, the results of this study showed that R1193Q polymorphism is not common in Iranian population. However, the results of this study are in concordance with European population.

#### DISCUSSION:

R1193Q Polymorphism (R1193Q GGG> CAG, rs41261344) is located at the interface between domain 2 and 3 of the SCN5A gene. The arginine residue at position 1193 in different isoforms of sodium channel is a strongly conserved site amongst mammalian. It seems that R1193Q missense

substitution is a functional mutation that can result in sodium channel inactivation and increases susceptibility to LQTS [29-32]. Also, this substitution was detected in patients with Brugada syndrome R1193Q mutation (Refseq: NM – 000335) was reported in Japanese baby with Brugada syndrome [26, 30 and 32]. Interestingly, the R1193Q polymorphism acts as a gain of function mutation in patients with LQTS3, whilst this functional substitution results in a loss of function status in patients with Brugada syndrome. All together, these observations suggest that R1193Q polymorphism could be considered as a risk factor in development of these diseases. Although it was elucidated that SCN5A gene mutations are involved in sudden death and also R1193Q replacement is common polymorphism in different Asian populations, we did not observe association between R1193Q polymorphism and the cardiomyopathy syndromes, LQTS and ARVC, in Iranian population. The geographical distribution of R1193Q polymorphism was analyzed in more than 4,000 populations of Asia, Europe and Africa. In this context, overall, the R1193Q polymorphism might have several consequences, including arrhythmias, electrophysiological abnormality associated with heart disorders, and sudden death [26]. the presence of R1193Q polymorphism has been reported in 7 cases of LQTS, which is supportive of the idea that this mutation might increase the susceptibility to LQTS. If we access the variants of the genes that are involved in the risk of ARVC and LQTS, we would be able to use these genetic profiles to screen high risk families or even reduce the mortality of the children suffering from ARVC and LQTS at least in Iranian population. It also suggests that to approve accuracy of this study, further investigation with larger participant numbers is extremely recommended.

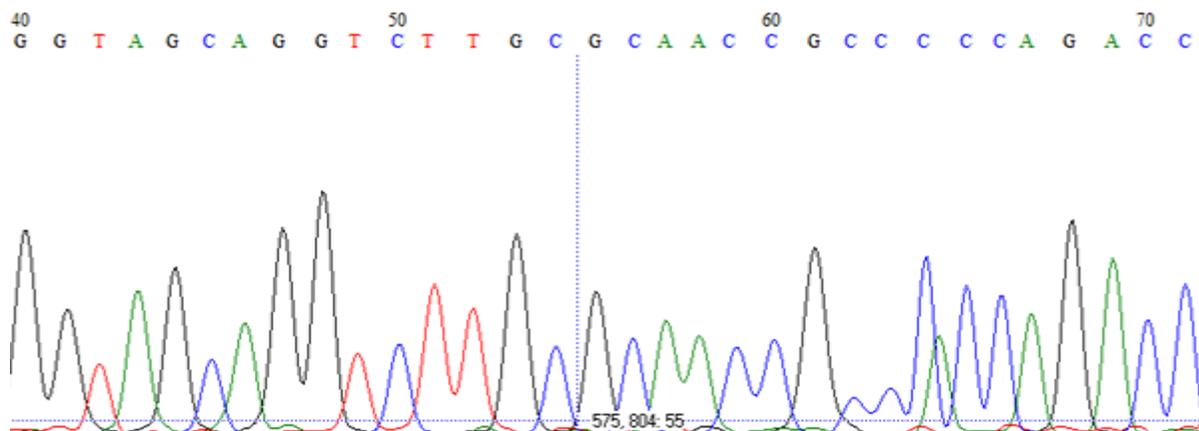


Fig 1: Results of sequence SCN5A gene, R1193Q polymorphism

**Ethical Considerations:**

This case-control association study has been approved by ethics committee of Islamic Azad University, Tehran, Iran.

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**Conflict of interest**

The authors have no conflict of interest

**REFERENCES:**

1. Abbate A, Biondi-Zoccai GG, Bussani R, Dobrina A, Camilot D, Feroce F, Rossiello R, Baldi F, Silvestri F, Biasucci LM, Baldi A. Increased myocardial apoptosis in patients with unfavorable left ventricular remodeling and early symptomatic post-infarction heart failure. *Journal of the American College of Cardiology*. 2003; 41(5): 753-60.
2. Van Tintelen JP, Pieper PG, Van Spaendonck-Zwarts KY, Van Den Berg MP. Pregnancy, cardiomyopathies, and genetics. *Cardiovascular research*. 2014; 101(4): 571-78.
3. Oliva A, Hu D, Viskin S, Carrier T, Cordeiro JM, Barajas-Martinez H, Wu Y, Burashnikov E, Brugada R, Rosso R, Guerschicoff A. SCN5A Mutation associated with acute myocardial infarction. *Legal Medicine*. 2009; 11(2): 206-9.
4. Gandjbakhch E, Vite A, Gary F, Fressart V, Donal E, Simon F, Hidden-Lucet F, Komajda M, Charron P, Villard E. Screening of genes encoding junctional candidates in arrhythmogenic right ventricular cardiomyopathy/dysplasia. *Europace*. 2013; 15(10): 1522-25.
5. Schulze-Bahr E, Eckardt L, Breithardt G, Seidl K, Wichter T, Wolpert C, Borggrefe M, Haverkamp W. Sodium channel gene (SCN5A) mutations in 44 index patients with Brugada syndrome: different incidences in familial and sporadic disease. *Human mutation*. 2003; 21(6): 651-2.
6. Te Riele AS, Tandri H, Bluemke DA. Arrhythmogenic right ventricular cardiomyopathy (ARVC): cardiovascular magnetic resonance update. *Journal of Cardiovascular Magnetic Resonance*. 2014; 16(1):50-50.
7. Fressart V, Duthoit G, Donal E, Probst V, Deharo JC, Chevalier P, Klug D, Dubourg O, Delacretaz E, Cosnay P, Scanu P. Desmosomal gene analysis in arrhythmogenic right ventricular dysplasia/cardiomyopathy: spectrum of mutations and clinical impact in practice. *Europace*. 2010; 12(6): 861-68.

8. Campuzano O, Alcalde M, Allegue C, Iglesias A, García-Pavía P, Partemi S, Oliva A, Pascali VL, Berne P, Sarquella-Brugada G, Brugada J. Genetics of arrhythmogenic right ventricular cardiomyopathy. *Journal of medical genetics*. 2013; 50(5): 280-89.
9. Matsusue A, Kashiwagi M, Hara K, Waters B, Sugimura T, Kubo SI. An autopsy case of sudden unexpected nocturnal death syndrome with R1193Q polymorphism in the SCN5A gene. *Legal Medicine*. 2012; 14(6): 317-19.
10. Marcus FI, Edson S, Towbin JA. Genetics of arrhythmogenic right ventricular cardiomyopathy. *Journal of the American College of Cardiology*. 2013; 61(19): 1945-48.
11. Palmisano BT, Rottman JN, Wells QS, DiSalvo TG, Hong CC. Familial evaluation for diagnosis of arrhythmogenic right ventricular dysplasia. *Cardiology*. 2011; 119(1): 47-53.
12. Sheikh F, Ross RS, Chen J. Cell-cell connection to cardiac disease. *Trends in cardiovascular medicine*. 2009; 19(6): 182-90.
13. Rook MB, Evers MM, Vos MA, Bierhuizen MF. Biology of cardiac sodium channel Nav1.5 expression. *Cardiovascular research*. 2011; 93(1): 12-23.
14. TFELT-HANSEN JA, Winkel BG, Grunnet M, Jespersen T. Inherited cardiac diseases caused by mutations in the Nav1.5 sodium channel. *Journal of cardiovascular electrophysiology*. 2010 Jan 1;21(1):107-15.
15. Remme CA, Wilde AA, Bezzina CR. Cardiac sodium channel overlap syndromes: different faces of SCN5A mutations. *Trends in cardiovascular medicine*. 2008; 18(3): 78-87.
16. Ruan Y, Liu N, Priori SG. Sodium channel mutations and arrhythmias. *Nature Reviews Cardiology*. 2009; 6(5): 337-48.
17. Wang DW, Desai RR, Crotti L, Arnestad M, Insolia R, Pedrazzini M, Ferrandi C, Vege A, Rognum T, Schwartz PJ, George AL. Cardiac sodium channel dysfunction in sudden infant death syndrome. *Circulation*. 2007; 115(3): 368-76.
18. Cheng J, Tester DJ, Tan BH, Valdivia CR, Kroboth S, Ye B, January CT, Ackerman MJ, Makielski JC. The common African American polymorphism SCN5A-S1103Y interacts with mutation SCN5A-R680H to increase late Na current. *Physiological genomics*. 2011; 43(9): 461-66.
19. Hwang HW, Chen JJ, Lin YJ, Shieh RC, Lee MT, Hung SI, Wu JY, Chen YT, Niu DM, Hwang BT. R1193Q of SCN5A, a Brugada and long QT mutation, is a common polymorphism in Han Chinese. *Journal of medical genetics*. 2005; 42(2): 7-8.
20. Lahtinen AM, Noseworthy PA, Havulinna AS, Jula A, Karhunen PJ, Kettunen J, Perola M, Kontula K, Newton-Cheh C, Salomaa V. Common genetic variants

associated with sudden cardiac death: the FinSCDgen study. *PloS one*. 2012; 7(7): 41675-83.

21.Vatta M, Dumaine R, Varghese G, Richard TA, Shimizu W, Aihara N, Nademanee K, Brugada R, Brugada J, Veerakul G, Li H. Genetic and biophysical basis of sudden unexplained nocturnal death syndrome (SUNDS), a disease allelic to Brugada syndrome. *Human molecular genetics*. 2002; 11(3): 337-45.

22.Marangoni S, Di Resta C, Rocchetti M, Barile L, Rizzetto R, Summa A, Severi S, Sommariva E, Pappone C, Ferrari M, Benedetti S. A Brugada syndrome mutation (p. S216L) and its modulation by p. H558R polymorphism: standard and dynamic characterization. *Cardiovascular research*. 2011; 91(4): 606-16.

23. Park JK, Martin LJ, Zhang X, Jegga AG, Benson DW. Genetic variants in SCN5A promoter are associated with arrhythmia phenotype severity in patients with heterozygous loss-of-function mutation. *Heart Rhythm*. 2012; 9(7): 1090-96.

24.Wang Q, Chen S, Chen Q, Wan X, Shen J, Hoeltge GA, Timur AA, Keating MT, Kirsch GE. The common SCN5A mutation R1193Q causes LQTS-type electrophysiological alterations of the cardiac sodium channel. *Journal of medical genetics*. 2004; 41(5): 66-66.

25.Shinlapawittayatorn K, Du XX, Liu H, Ficker E, Kaufman ES, Deschênes I. A common SCN5A polymorphism modulates the biophysical defects of SCN5A mutations. *Heart Rhythm*. 2011; 8(3): 455-62.

26.Watanabe H, Nogami A, Ohkubo K, Kawata H, Hayashi Y, Ishikawa T, Makiyama T, Nagao S, Yagihara N, Takehara N, Kawamura Y.

Electrocardiographic characteristics and SCN5A mutations in idiopathic ventricular fibrillation associated with early repolarization. *Circulation. Arrhythmia and electrophysiology*. 2011; 4(6): 874-81.

27.Rijnbeek PR, Van Herpen G, Kapusta L, ten Harkel AD, Witsenburg M, Kors JA. Electrocardiographic criteria for left ventricular hypertrophy in children. *Pediatric cardiology*. 2008; 29(5): 923-28.

28.Sun A, Xu L, Wang S, Wang K, Huang W, Wang Y, Zou Y, Ge J. SCN5A R1193Q polymorphism associated with progressive cardiac conduction defects and long QT syndrome in a Chinese family. *Journal of medical genetics*. 2008; 45(2): 127-28.

29.Friedlander Y, Siscovick DS, Weinmann S, Austin MA, Psaty BM, Lemaitre RN, Arbogast P, Raghunathan TE, Cobb LA. Family history as a risk factor for primary cardiac arrest. *Circulation*. 1998; 97(2): 155-60.

30.Nakajima T, Kaneko Y, Saito A, Irie T, Tange S, Iso T, Kurabayashi M. Identification of six novel SCN5A mutations in Japanese patients with Brugada syndrome. *International heart journal*. 2011; 52(1): 27-31.

31.Miller SA, Dykes DD, Polesky HF. A simple salting out procedure for extracting DNA from human nucleated cells. *Nucleic acids research*. 1988; 16(3): 1215-15.

32.Huang H, Zhao J, Barrane FZ, Champagne J, Chahine M. Nav1. 5/R1193Q polymorphism is associated with both long QT and Brugada syndromes. *Canadian Journal of Cardiology*. 2006; 22(4): 309-13.