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Mitochondrial DNA haplogroup H as a risk factor for idiopathic dilated cardiomyopathy in spanish population

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Abstract
Idiopathic dilated cardiomyopathy (IDC) is a structural heart disease with strong genetic background. The different single nucleotide polymorphisms (SNPs) that constitute mitochondrial haplogroups could play an important role in IDC progression. The aim of this study was to test frequencies of mitochondrial haplogroups in healthy controls (n = 422) and IDC patients (n = 304) of a Caucasian Spanish population. To achieve this, ten major European haplogroups were identified. Frequencies and Odds Ratios for the association between IDC and haplogroups were calculated in both groups. We found that compared to healthy controls, the prevalence of haplogroup H was significantly higher in IDC patients (40.0% vs 50.7%, p-value = 0.040).

Abbreviations
SNP, Single Nucleotide Polymorphism; IDC, Idiopathic Dilated Cardiomyopathy; ROS, Reactive Oxygen Species; mtDNA, Mitochondrial DNA; ETC, Electron Transport Chain; SBE, Single Base Extension; OR, Odds Ratio; CI, Confidence Interval; RFLP, Restriction Fragment Length Polymorphisms

Keywords
Mitochondrial haplogroups; Idiopathic dilated cardiomyopathy; Oxidative stress; Reactive oxygen species; Single nucleotide polymorphisms

1. Introduction

Dilated cardiomyopathy is a myocardial disorder characterized by dilation and dysfunction of the left ventricle. Between 20 and 48% of dilated cardiomyopathy cases are heritable (Baig *et al*., 1998; Mestroni *et al*., 1994 ; Michels *et al*., 1992). However, dilated cardiomyopathy is also present in the absence of congenital, valvular, coronary artery disease or any systemic disease known to cause myocardial dysfunction. In this case the cardiomyopathy is defined as idiopathic dilated cardiomyopathy (IDC). Accumulating evidence has implicated the oxidative stress in the progression of IDC, but the molecular mechanisms are unknown. An increased concentration of reactive oxygen species (ROS) plays a key role in IDC promoting the apoptotic death of myocytes, endothelial cells and fibroblasts (Cesselli et al., 2001), and ultimately leads to myocyte hypertrophy and interstitial fibrosis (Kinugawa et al., 2000). The clinical consequence of oxidative stress is the reduction of contractile function in IDC patients (Burton *et al*., 1984 ; Gupta and Singal, 1989). An in-depth understanding of its basic pathophysiologic mechanisms is necessary to provide early prognosis and better therapies for preventing and curing this disease.

Human mitochondrial DNA (mtDNA) encodes 37 genes, but only 13 of these genes are transcripted into 13 polypeptides. They constitute essential subunits of the mitochondrial oxidative phosphorylation enzyme complexes, which provide the principal source of ATP (DiMauro and Schon, 2003 ; Wallace, 2005). The function of mitochondrion-encoded proteins is affected by amino acid substitutions, but they can also be indirectly affected by mutations in mtDNA control regions. mtDNA mutations have accumulated throughout human history and they are present in groups of human populations: the mitochondrial haplogroups. Each mitochondrial haplogroup is defined as a collection of haplotypes characterized by specific single nucleotide polymorphisms (SNPs). SNPs are present in indigenous populations and this has been attributed to genetic drift and/or possible climate selection (Mishmar *et al*., 2003 ; Ruiz-Pesini *et al*., 2004).

The polymorphic variants are directly associated with the disease, but others could affect indirectly to its development. Pello et al. (2008) described specific SNPs involved in the assembly of components of the Electron Transport Chain (ETC). In fact, another study showed that mitochondrial haplogroups are associated with differences in the concentration of superoxide, and other reactive oxygen species produced by the ETC (Marcuello et al., 2009). The variation of oxidative stress levels in cells will ultimately have an effect in morbidity, mortality and longevity among individuals with different haplogroups (Cai *et al*., 2009 ; Herrnstadt and Howell, 2004).

Specific haplogroups may constitute either a risk or a protective factor in the origin of complex diseases or age-related diseases such as Parkinson's (van der Walt et al., 2003), Alzheimer's disease (Santoro *et al*., 2010 ; van der Walt *et al*., 2004), osteoarthritis (Rego-Pérez *et al*., 2008 ; Rego-Pérez *et al*., 2010) and several cancers (Fang *et al*., 2010 ; Li *et al*., 2011). Mitochondrial haplogroups have also been associated with an increased risk of developing various cardiovascular diseases. In particular, the haplogroup T is more frequent in hypertrophic cardiomyopathy patients (Castro et al., 2006). Furthermore, haplogroup H1 (Rosa et al., 2008), K (Chinnery et al., 2010) and the Asian haplogroup N9b (Nishigaki et al., 2007) are protective factors against ischemic stroke. In a previous study, we found haplogroups H and J to be risk and protective factors for ischemic cardiomyopathy development, respectively (Fernández-Caggiano et al., 2012).

In this work, a case–control study was performed to assess the possible association of mitochondrial haplogroups in a Spanish population of 304 IDC patients and 422 controls. In addition, the frequencies of different SNPs that characterize the mitochondrial haplogroups were analyzed in order to determine whether any of them constitutes a risk factor for IDC development.

2. Methods

2.1. Ethics statement

The present study was conducted according to the Spanish Law for Biomedical Research (Law 14/2007 — 3rd of July) and complied with the Declaration of Helsinki. The study and the use of archive samples for this project were approved by the Research Ethics Committee of Galicia. The DNA National Bank Institution which provided DNA samples received approval from their own ethical committee. Written informed consent was obtained from all individuals. All the samples were collected anonymously.

2.2. Patients and controls

DNA samples from 726 unrelated Caucasian Spanish individuals (422 healthy controls and 304 IDC patients) were used in this study. The IDC group included 224 patients obtained from the A Coruña University Hospital Cardiology Unit and 80 provided by the DNA National Bank (University of Salamanca, Spain). The control group was an age and sex matched population of 422 donors from A Coruña University Hospital Blood Bank. Individuals in this group represented both genders and had no history of IDC. We included in the group of patients those who met all the criteria established by The American Heart Association clinical standards (Radford et al., 2005). These criteria included patients with heart failure and reduced systolic function with a dilatation of the ventricular chambers diagnosticated by 2-dimensional echocardiography. Patients did not have any previous evidence for myocardial infection neither reported familial cardiomyopathies. The ischemic origin was discarded if any of these conditions were present: 1) at least one major epicardial coronary artery with more than 70% obstruction by coronary angiography; 2) history of acute myocardial associated with wall motion abnormalities by echocardiography and 3) stress testing diagnostic of coronary artery disease. Clinical parameters about all the individuals were collected. Hypercholesterolemia was considered a risk if total cholesterol levels were ≥ 220 mg/dl. Body mass index was expressed as weight in kilograms divided by height in square meters. Hypertension was defined as systolic blood pressure ≥ 140 mm Hg, diastolic blood pressure ≥ 90 mm Hg or by the use of antihypertensive medication. Smokers were defined as current smokers. Diabetes mellitus was defined as a self-reported disease, use of antidiabetic drugs, or a nonfasting plasma glucose ≥ 11 mmol/l.

2.3. Assignment to mDNA haplogroups

Haplogroup analysis was based on the use of single base extension (SBE) for the assessment of European mtDNA haplogroups. The SBE assay permitted us to identify six SNPs that determine the most frequent European haplogroups (H, T, K, U, J, V), while the less common haplogroups (W, I, X) were identified by polymerase chain reaction–restriction fragment length polymorphisms (PCR–RFLPs). The samples obtained in this study were haplogroup-typed using a previously described assay (Fernández-Caggiano et al., 2012). Fragments containing the six analyzed SNPs were amplified using the twelve primers listed in Supplementary Table 1.

2.4. Statistical methods

Statistical analysis was performed using SPSS 17.0 software. The Chi-square test was used to assess haplogroups and allele frequencies between controls and patients. For the haplogroup analysis, each haplogroup was compared against all the other haplogroups pooled into a single group. The less frequent haplogroups I, W and X, which account for less than 10 controls/patients, were re-grouped based on common-ancestor criteria. The haplogroup HV was re-grouped as "others". Odds Ratio (OR) and 95% Confidence Intervals (CI) were calculated for each haplogroup. Comparisons between haplogroups in IDC patients and controls were computed by applying the Bonferroni's adjustment. Thus, significant p-values obtained from Chi-square test were multiplied by the number of outcomes ($k = 8$ for mtDNA haplogroups and $k = 4$ for mtDNA clusters) tested and the differences were

considered significant if, after the adjustment, $p < 0.05$ (2-tailed test). Haplogroup frequencies between controls in this study and in other European studies were also analyzed using the same tests. Binary logistic regression adjustment was used to test the influence of hypercholesterolemia, hypertension, diabetes mellitus and the smoking habit. Differences were considered significant at $p < 0.05$ (2-tailed test).

3. Results

3.1. Clinical parameters stratified by mitochondrial haplogroups

A total of 726 Caucasian Spanish subjects were included in this study, 304 had been previously diagnosed with IDC (case group) and 422 were controls with no history of IDC. The subjects were selected to match sex and age of the patients' population. Considering the age variable influence we selected subjects of similar ages (66.33 ± 11.7 years) to the IDC group (59.5 ± 15.1 years) for the control group. This prevented the inclusion of young individuals predisposed to IDC in the control group. The lower proportion of women developing IDC was also taken into account and we selected our control group (22.0% of women) to have frequency similar to our case group (26.5% of women). Clinical parameters stratified by mitochondrial haplogroups are listed in Table 1. The distribution of haplogroups for the major IDC risk factors was no different between controls and patients.

Table 1. Mitochondrial haplogroup frequencies (%) stratified by classical risk factors for IDC development.

		Hypercholesterolemia		Hypertension		Diabetes		Smoking habit	
		C	IDC	C	IDC	C	IDC	C	IDC
Haplogroups	H	44 (36.7)	41 (37.6)	49 (40.2)	45 (40.2)	20 (35.7)	24 (39.3)	25 (48.1)	42 (47.2)
	U	22 (18.3)	20 (18.3)	14 (11.5)	18 (16.1)	6 (10.7)	7 (11.5)	10 (19.2)	18 (20.2)
	J	15 (12.5)	15 (13.8)	17 (13.9)	17 (15.2)	8 (14.3)	9 (14.8)	4 (7.7)	9 (10.1)
	T	13 (10.8)	10 (9.2)	16 (13.1)	12 (10.7)	6 (10.7)	6 (9.8)	4 (7.7)	5 (5.6)
	K	7 (5.8)	10 (9.2)	8 (6.6)	7 (6.2)	7 (12.5)	5 (8.2)	4 (7.7)	5 (5.6)
	V	2 (1.7)	1 (0.9)	3 (2.5)	3 (2.7)	2 (3.6)	2 (3.3)	2 (3.8)	2 (2.2)
	I WX	8 (6.7)	4 (3.7)	7 (5.7)	2 (1.8)	2 (3.6)	2 (3.3)	1 (1.9)	2 (2.2)
	O	9 (7.5)	8 (7.3)	8 (6.6)	8 (7.1)	5 (8.9)	6 (9.8)	2 (3.8)	6 (6.7)
	Total	120	109	122	112	56	61	52	89

C. Controls. IDC. Idiopathic dilated cardiomyopathy patients. O. Others. No significant differences were found for the frequencies of major cardiovascular risk factors between controls and IDC patients stratified by mitochondrial haplogroups.

3.2. Haplogroup H and cluster HV: risk factors for IDC development

Samples were genotyped for the most common European descent mitochondrial haplogroups and the resulting frequencies are shown in Table 2. The obtained frequencies ranged from 50.7% for the most common haplogroup H, to 1.6% for the less prevalent haplogroup V. Using the rapid and effective multiplex SBE assay, 92.5% of the samples were assigned to the most common European mtDNA haplogroups (H, U, J, K, T, V and HV). The less frequent haplogroups (X, I, W) accounted for 3.6% of the samples and were assigned using the conventional PCR–RFLP assay. The haplogroup frequencies for our control group did not differ substantially from those reported in previous studies that analyzed different European populations (Supplementary Table 2) (Kofler *et al.*, 2009; Mancuso *et al.*, 2004 ; Torroni *et al.*, 1996).

Table 2. Frequencies and OR of mitochondrial haplogroups and clusters in controls and patients with IDC.

		No. of individuals (%)		Total	OR [95% CI]	p-Value
		Control (n = 422)	IDC patients (n = 304)			
Haplogroups	H	169 (40.0)	154 (50.7)	323 (44.5)	1.54 [1.14–2.07]	**0.040**[a]
	U	73 (17.3)	45 (14.8)	118 (16.3)	0.83 [0.55–1.24]	0.415
	J	47 (11.1)	31 (10.2)	78 (10.7)	0.91 [0.56–1.46]	0.717
	T	47 (11.1)	28 (9.2)	75 (10.3)	0.81 [0.49–1.32]	0.459
	K	28 (6.6)	20 (6.6)	48 (6.6)	0.99 [0.55–1.79]	1.000
	V	13 (3.1)	5 (1.6)	18 (2.5)	0.52 [0.19–1.49]	0.239
	I WX	20 (4.7)	6 (2.0)	26 (3.6)	0.40 [0.16–1.02]	0.067
	OTHERS	25 (5.9)	15 (4.9)	40 (5.6)	0.82 [0.43–1.59]	0.623
Clusters	HV	188 (44.5)	166 (54.6)	354 (48.8)	1.50 [1.11–2.01]	**0.030**[a]
	JT	94 (22.3)	59 (19.4)	153 (21.1)	0.84 [0.58–1.21]	0.358
	KU	101 (23.9)	65 (21.4)	166 (22.9)	0.86 [0.61–1.23]	0.422
	I WX	20 (4.7)	6 (2.0)	26 (3.6)	0.40 [0.16–1.02]	0.067

OR. Odds Ratio. 95% CI. Confidence Intervals. IDC. Idiopathic dilated cardiomyopathy patients.
[a] Bonferroni corrected p-value. The significant p-values obtained in the Chi-squared test were multiplied by the number of outcomes (k = 8 for mtDNA haplogroups and k = 4 for mtDNA clusters) tested. Significant differences (p-value < 0.05) are indicated in bold.

The haplogroup H was significantly overrepresented in IDC patients (OR = 1.54 [95% CI = 1.14–2.07], p = 0.040) when each haplogroup was compared against all the rest pooled together (Table 2). These results indicate that haplogroup H constitutes a risk factor for IDC in our study population.

Because the haplogroups analyzed share a common ancestry and several SNPs have been conserved during evolution, we examined the frequencies of clusters HV, JT, KU and IWX. Cluster HV was found to be a risk factor for IDC (OR = 1.50 [95% CI = 1.11–2.01], p = 0.030) (Table 2).

Hypercholesterolemia, hypertension, diabetes and smoking habit were significantly and independently associated with IDC after the multivariate logistic regression analysis (Table 3). Our results supported previous studies showing that IDC development was associated with hypercholesterolemia (OR = 1.51 CI = [1.08–2.11], $p < 0.05$), hypertension (OR = 1.40 CI = [1.00–1.95], $p < 0.05$), diabetes (OR = 1.83 CI = [1.20–2.78], $p < 0.05$) and cigarette smoking (OR = 3.11 CI = [2.09–4.61], $p < 0.001$). The haplogroup H continued to be a risk factor compared with haplogroup U (OR = 0.62 CI = [0.39–0.98], $p < 0.05$), haplogroup V (OR = 0.25 CI = [0.07–0.92], $p < 0.05$) and the cluster IWX (OR = 0.31 CI = [0.12–0.81], $p < 0.05$) (Table 3). These results support the idea that patients with IDC are overrepresented by haplogroup H compared with the haplogroups U and V and the cluster IWX.

Table 3. Multivariate analysis of the study groups.

	B	SEM	OR [95% CI]	p-Value
Hypercholesterolemia	0.411	0.172	1.51 [1.08–2.11]	**0.017**
Hypertension	0.334	0.169	1.40 [1.00–1.95]	**0.048**
Diabetes	0.604	0.213	1.83 [1.20–2.78]	**0.005**
Smoking habit	1.133	0.202	3.11 [2.09–4.61]	**0.001**
Haplogroup H	–	–	1	–
Haplogroup U	− 0.476	0.230	0.62 [0.39–0.98]	**0.039**
Haplogroup J	− 0.449	0.270	0.64 [0.38–1.08]	0.097
Haplogroup T	− 0.411	0.271	0.66 [0.39–1.13]	0.130
Haplogroup K	− 0.357	0.327	0.70 [0.37–1.33]	0.274
Haplogroup V	− 1.400	0.674	0.25 [0.07–0.92]	**0.038**
Haplogroup IWX	− 1.178	0.491	0.31 [0.12–0.81]	**0.016**
Others	− 0.638	0.361	0.53 [0.26–1.07]	0.077

B. Regression coefficient. SEM. Standard error of the mean. OR. Odds Ratio. 95% CI. Confidence Intervals. Significant differences (p-value < 0.05) are indicated in bold.

3.3. mtDNA alleles m.7028C and m.14766C were risk factors for IDC

The frequencies for eight SNPs characteristic of European mtDNA haplogroups were also analyzed. Two phylogenetically associated SNPs were found overrepresented in IDC patients: the SNP m.7028C>T, which characterizes haplogroup H (OR = 1.54 [95% CI = 1.14–2.07], p = 0.005) and SNP m.14766C>T (OR = 1.48 [95% CI = 1.10–1.99], p = 0.010), which is associated with cluster HV (Table 4). Although the SNP m.7028C>T does not produce an amino acid change in p.MT-CO1, the nucleotide change in 14766 *locus* produces a non-synonymous amino acid change in cytochrome b (p.Thr7Ile).

Table 4. Polymorphisms relative to the revised Cambridge reference sequence found in each nucleotide position analyzed.

Nt position	Locus	Nt change	Amino acid change	No. of individuals (%)		OR [95% CI]	p-Value
				Controls	IDC patients		
7028	Cytochrome c oxidase subunit 1	C>T	Syn	169 (40.0)	154 (50.7)	1.54 [1.14–2.07]	**0.005**
14766	Cytochrome b	C>T	p.Thr7Ile	188 (44.5)	165 (54.3)	1.48 [1.10–1.99]	**0.010**
10398	NADH dehydrogenase subunit 3	A>G	p.Thr114Ala	97 (23.0)	62 (20.4)	0.86 [0.60–1.23]	0.415
4580	NADH dehydrogenase subunit 2	G>A	Syn	14 (3.3)	5 (1.6)	0.49 [0.17–1.37]	0.238
12308	tRNA leucine 2	A>G	Syn	101 (23.9)	65 (21.4)	0.86 [0.61–1.23]	0.422
4216	NADH dehydrogenase subunit 1	T>C	p.Tyr304His	94 (22.3)	59 (19.4)	0.84 [0.58–1.21]	0.358
10034	tRNA glycine	T>C	Syn	3 (0.7)	3 (1.0)	1.39 [0.28–6.94]	1.000
14470	NADH dehydrogenase subunit 6	T>C	Syn	16 (3.8)	4 (1.3)	0.34 [0.11–1.02]	0.064

Nt position. Nucleotide position. Nt change. Nucleotide change. Syn. Synonymous. OR. Odds Ratio. 95% CI. Confidence Intervals. IDC. Idiopathic dilated cardiomyopathy. Significant differences (p-value < 0.05) are indicated in bold.

4. Discussion

We found significant association of mitochondrial haplogroup H and the incidence of idiopathic dilated cardiomyopathy in our Spanish population. To the best of our knowledge, this is the first time that haplogroup H was found as a risk factor for IDC.

To this date, IDC cannot be associated to single gene defects. While a few common susceptibility alleles for IDC have been identified from candidate-gene approaches, they have not been confirmed yet in replicative populations (Rampersaud et al., 2010), which is a common problem of single gene based analyses.

Because cardiac tissue has high-energy requirements, mitochondrial mutations have been hypothesized to contribute to IDC development. Although to identify which mtDNA mutations actually cause IDC is a challenge yet, some mtDNA mutations have been reported in IDC (Li *et al*., 1997; Mahjoub *et al*., 2007; Marin-Garcia *et al*., 2000 ; Santorelli *et al*., 1999). However, we did not report any risk association between analyzed SNPs in our work and those considered as possibly relevant for the dilated cardiomyopathy pathogenesis in these studies.

Gallardo et al. (2012) described haplogroup H as a risk factor for the progression to end-stage heart failure in a Spanish population. In agreement with our study, they observed that the frequency of haplogroup H in 148 patients with idiopathic dilated cardiomyopathy was 51.4% when they divided the allograft recipients according to etiology. However, in this study it was not possible to confirm in a statistical way the haplogroup H as a risk factor for idiopathic dilated cardiomyopathy. This was probably due to the low number of patients with this disease included in the study (n = 148). Our data from 304 patients supported in a statistical way the haplogroup H as a risk factor for idiopathic dilated cardiomyopathy. A recent study showed mitochondrial haplogroup H as a risk factor for early onset myocardial infarction (Palacín et al., 2011). Conversely, Rosa et al. (2008) reported sub-haplogroup H1 to be a protective factor for ischemic stroke. Therefore, the difference in the phenotype could be due to other polymorphisms present in sub-haplogroup H1. Taken together, these studies suggest that no single specific SNP is responsible for the risk effect; instead it is due to a

particular set of polymorphisms within haplogroup H. Although distribution in our control population did not differ from those in other European studies (Kofler *et al*., 2009; Mancuso *et al*., 2004; Palacín *et al*., 2011 ; Torroni *et al*., 1996), an exhaustive work carried by Benn et al. (2008) in a Danish population found no differences between mitochondrial haplogroups and risk for myocardial infarction or ischemic stroke. Therefore, our Spanish population cannot be directly extrapolated to other Northern European populations. The differing results between studies could be explained by geographic specificity for some mtDNA SNPs and clades.

There are many SNPs that characterize haplogroup H. In this study we analyzed those which permitted us to classify the different European mitochondrial haplogroups. Among them, the allele m.7028C was found overrepresented in our study. However, the SNP m.7028C>T causes a synonymous amino acid change, and therefore this SNP is not responsible for the phenotypic effect that defines haplogroup H as a risk factor for IDC development. On the other hand, the m.14766C allele constituted a risk factor for the IDC development in our Spanish population. The SNP m.14766C>T causes the amino acid substitution of a threonine for an isoleucine at site 7 in cytochrome b. A computational approach accomplished by Beckstead et al. (2009) indicated the possibility that the region around cytochrome site 7 becomes more open, less globular and less compact due to the presence of a threonine. This could affect the efficiency of the ETC., which is expected to be higher in haplogroup H cells. Accordingly, results emerging from different studies have provided insights concerning different energy efficiency between haplogroups. Haplogroup H has higher oxygen consumption than haplogroup J for example (Marcuello *et al*., 2009 ; Martínez-Redondo *et al*., 2009), which is associated with higher efficiency in the electronic respiratory chain and low ATP and ROS production. Consequently, cells with mitochondrial haplogroup H undergo more mitochondrial oxidative damage. Since the heart has the highest oxygen uptake rate in body, we speculate that minor differences in energy efficiency might lead to major physiological effects.

It is well known that oxidative stress is increased in patients with dilated cardiomyopathy or acute myocardial infarction (Hill and Singal, 1996). On the subcellular level, the reactive oxygen species attack biomolecules such as contractile proteins (Canton *et al*., 2011 ; Kaneko *et al*., 1994) or ion channels (Liu et al., 2010). Additionally, the change of intracellular redox balance may lead to activation of stress sensitive signaling pathways. Several reports showed evidence of increased oxidative stress in dilated cardiomyopathy patients (Cesselli *et al*., 2001; Kato *et al*., 2010; Shah *et al*., 2011; Yücel *et al*., 1998 ; Yücel *et al*., 2002). Furthermore, it has been demonstrated that under experimental conditions therapy with antioxidant drugs is able to arrest the development of this disease (Cappola *et al*., 2001; Kawakami *et al*., 2009 ; Nishioka *et al*., 2007). This provides further evidence for a significant role of reactive oxygen species in IDC development. From our results, we suggest that individuals with haplogroup H might have a slight impaired intracellular redox balance that possibly influences IDC development.

A complete understanding of the genetic basis of IDC has not been achieved based on currently available data. Most recent efforts have been devoted to IDC gene discovery or to preliminary studies of mutation frequency in modest sized IDC cohorts. Besides, it must be taken into account that other factors are involved on the IDC development. Mitochondrial haplogroups may act synergistically with proteins and environmental components present in the cell. Although this work showed significant results, a limitation of the present study is the lack of a replication study in another population. A replica of our study is quite demanding, due to difficulties in enrolling another comparable large number of patients. Nevertheless, IDC patients and controls have been recruited in a relatively large geographic area thus avoiding possible bias related to founder effect or population heterogeneity. We believe that additional studies similar to the present one, will allow in the future meta-analyses assessing actual risk scores and equations for the different mitochondrial haplogroups in the development of IDC.

5. Conclusion

Our results show suggestive evidence for the association of mitochondrial haplogroup H as risk factor for idiopathic dilated cardiomyopathy development in a Caucasian Spanish population. Further analysis of the full sequenced mtDNA in these haplogroups and their phenotypic analysis might yield additional insights towards therapeutic targets for IDC pathogenesis.

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