Series of Cardiology Research

Open Access
Research Article
Vol 6 Iss 1

Expanding Application of Preimplantation Genetic Testing for Cardiac Disease

Kuliev A*, Pakhalchuk T, Prokhorovich M and Rechitsky S

Reproductive Genetic Innovations, Chicago, Illinois, USA

***Correspondence:** Anver Kuliev, Reproductive Genetic Innovations, Chicago, Illinois, USA

Received on 12 December 2023; Accepted on 09 April 2024; Published on 26 April 2024

Summary

Predisposition to cardiac disease (CD) is currently becoming a common indication for preimplantation genetic testing (PGT). It provides an option for the couplers at risk to avoid the birth of an offspring with a predisposition to CD, as there are no appropriate strategies to prevent CD in the carries of gene mutations predisposing to this condition. The present paper describes the progress in the application of PGT for CD, with its extended application also to carriers of predisposing genes detected through expanded carrier screening (ECS). We present here our experience of 147 PGT cycles for CD, resulting in the birth of 73 children free of predisposing genes to CD, which is a part of our overall PGT series of over one thousand PGT for monogenic disorders (PGT-M). The accumulated experience, presented below, demonstrates considerable progress in using PGT to avoid the birth of children with a genetic predisposition to CD.

Keywords: cardiac disease, preimplantation genetic testing, gene mutations, PGT cycles

Abbreviations: CD: cardiac disease; PGT: preimplantation genetic testing; ECS: expanded carrier screening; PGT-M: preimplantation genetic testing for monogenic disorders; ART: assisted reproductive technology; PB: polar bodies; WGA: whole genome amplification; ADO: allele dropout

Introduction

At present, there are no appropriate strategies to prevent cardiac disease (CD) in the carries of gene mutations predisposing to this condition, making avoidance of the birth of such offspring quite attractive to couples at risk, through the option of preimplantation genetic testing for monogenic disorders (PGT-M), currently performed for increasing number of patients [1–5]. The available experience shows that the pathway of referral of couples with an inherited predisposition to PGT-M has recently been changing with the current shift to direct referral through the information on the internet, and lately through the application of expanded carrier screening (ECS), which identifies

at-risk couples who can benefit from PGT-M even without a known family history. Thus, the framework of avoiding the birth of affected children also involves ECS for the identification of individuals at risk, providing them with an option of avoiding the birth of a child with a predisposition to CD, and, instead, having an offspring free from the genes predisposing to the disease by utilizing PGT. Accordingly, it is reasonable that the testing for genes predisposing to CD is included in the available ECS programs, in addition to the provision of information on the availability of PGT to couples, with a family history of CD, so they could make their choices for a possible carrier screening and PGT.

Performed already for almost twenty years [1, 2], predisposition to CD is currently not a rare indication for PGT-M, as it not only ensures the avoidance of inheritance of CD predisposing genes but also provides an opportunity to have an offspring without predisposing genes of their own, despite having to undergo assisted reproductive technology (ART) [2–5]. It is also of note that predisposing to CD gene mutations is mainly of the autosomal-dominant mode of inheritance, with a 50% risk of passing these genes to their offspring, so the application of ECS programs allows detecting the at-risk individuals even before they have contracted the disease, in addition to those diagnosed with CD and those with family history of CD.

We have previously reported our experience of PGT for CD, as part of our overall PGT-M experience [4, 5]. The present paper is an update of 147 PGT-M cycles for CD presently applied, also, prospectively to an increasing number of at-risk couples detected through ECS, resulting in the birth of 73 children free of CD predisposing genes in one of the world's largest series of PGT-M cycles performed for patients at risk of producing offspring predisposed to CD.

Materials and Methods

A total of 147 PGT cycles for 91 couples (Table 1) at risk for producing a progeny with genes predisposing to CD were performed. The most frequent were hypertrophic cardiomyopathy, CMH4, caused by MYBPC3 mutation (30 cycles performed for 19 patients), and dilated cardiomyopathy, CMD1A, caused by LMNA gene mutation (19 cycles performed for 7 patients). PGT for remaining mutations was performed in under a dozen cycles.

Disease	**Gene**	**# Pt**	**# Cycle**	**# Transfers**	**# Embryos transferred**	**Pregnancy**	**Birth**
ACYL-CoA dehydrogenase, very long-chain, deficiency of; ACADVLD	ACADVL (AR)	5	6	8	12	2	2
Cardioencephalomyopathy, fatal infantile, due to cytochrome c oxidase deficiency 1; CEMCOX1	SCO2 (AR)	2	5	5	10	3	3
Cardiofaciocutaneous syndrome 4; CFC4	MAP2K2 (AD)	1	2	1	1	1	1
Cardiomyopathy, dilated, 1A; CMD1A	LMNA (AD)	6	19	17	26	11	10
Cardiomyopathy, dilated, 1DD; CMD1DD	RBM20 (AD)	2	3	5	5	3	3
Cardiomyopathy, dilated, 1E; CMD1E	SCN5A (AD)	1	2	2	2	1	1
Cardiomyopathy, dilated, 1G; CMD1G	TTN (AD)	4	4	4	6	3	3
Cardiomyopathy, dilated, with woolly hair, keratoderma, and tooth agenesis; DCWHKTA	DSP (AD)	5	7	6	7	6	3
Cardiomyopathy, familial hypertrophic, 1; CMH1	MYH7 (AD)	7	11	8	10	4	3
Cardiomyopathy, familial hypertrophic, 2; CMH2	TNNT2 (AD)	1	3	2	4	2	2
Cardiomyopathy, familial hypertrophic, 4; CMH4	MYBPC3 (AD)	19	30	21	32	15	13

Cardiomyopathy, familial hypertrophic, 7; CMH7	TNNI3 (AD)	4	5	7	7	3	2
Cardiomyopathy, familial hypertrophic, 8; CMH8	MYL3 (AD)	1	2	1	1	0	0
Cardioskeletal myopathy with neutropenia and abnormal mitochondria	TAZ (XLR)	2	2	3	3	2	2
Coenzyme Q10 deficiency, primary, 7; COQ10D7	COQ4 (AR)	1	1	1	1	1	1
Emery-Dreifuss muscular dystrophy 1, X-linked; EDMD1	EMD (XLR)	3	5	5	8	4	4
Holt-Oram syndrome; HOS	TBX5 (AD)	6	9	9	10	5	5
Loeys-Dietz syndrome 1; LDS1	TGFBR2 (AD)	2	5	4	6	2	1
Long QT syndrome 1; LQT1	KCNQ1 (AD)	6	8	6	7	4	4
Long QT syndrome 2; LQT2	KCNH2 (AD)	3	3	2	2	1	1
Long QT syndrome 8; LQT8	CACNA1C (AD)	1	1	1	1	1	1
Myopathy, myofibrillar, 1; MFM1	DES (AD)	2	3	2	3	2	2
Noonan syndrome 1; NS1	PTPN11 (AD)	7	11	10	11	7	6
	23 Genes	91	147	130	175 1.34	83 63.8%	73

Table 1: PGT-M for inherited predisposition to cardiac disease. AD: autosomal dominant; AR: autosomal recessive; XLR: X-linked recessive.

PGT cycles were performed using a standard IVF protocol, coupled with micromanipulation procedures of polar bodies (PB) or embryo biopsy, described in detail elsewhere [5, 6]. Details of PGT guidelines were reported previously [7, 8]. The present standards of the procedure involve whole genome amplification (WGA) of biopsied PB or embryo biopsy samples, followed by multiplex nested PCR analysis of the mutations in question, together with closely linked genetic markers in a multiplex heminested system. Most cases are currently performed by blastocyst biopsy followed by WGA [5, 6]. For each family, heterozygous alleles and haplotypes not shared by parents were selected. This allowed detecting and avoiding misdiagnosis due to preferential amplification and allele dropout (ADO), and a possible aneuploidy or uniparental disomy of chromosomes in which the tested mutations are located, which may affect diagnostic accuracy of PGT-M. In PGT-M cycles, involving an advanced reproductive age maternal partner, aneuploidy testing was also performed by the next generation technologies (NGS) (Illumina Inc) for 24-chromosome aneuploidy testing [5, 9].

Results and Discussion

The table (Table 1) presents our cumulative experience of 147 PGT cycles performed in 91 couples at risk for producing offspring with a genetic predisposition to different CDs, caused by 23 genes. This is one of the largest series available for PGT for CD, resulting in the transfer of 175 predisposition-free embryos in 130 cycles (1.34 average embryos per transfer), yielding 83 (63.8%) clinical pregnancies and the birth of 73 healthy children free of predisposing gene mutations and demonstrating the practical utility of PGT for this group of inherited conditions.

The largest CD group was familial hypertrophic cardiomyopathy (CMH), including patients with CMH1, CMH2, CMH4, CMH7, and CMH8, determined by a mutation in MYH7, TNNT2, MYBPC3, TNNI3, and MYL3 genes respectively. Neither of these couples had previous progeny but had a family history of premature or sudden death. Among them, the most frequent was HCM4, caused by a mutation in the MYBPC3 gene, located on chromosome 11 (11p11.2) and encoding the cardiac isoform of myosin-binding protein C. This is localized exclusively in the heart muscle, with a high risk of cardiac failure and sudden death. Of 30 PGT cycles performed for CMH4, the single most common in our experience, 32 embryos free of the predisposing gene were detected for transfer in 21 cycles, resulting in the birth of 13 children with no risk of developing CD in their lifespan.

Another common was also dilated cardiomyopathy, CMD1A, an autosomal dominant disease caused by different mutations in the LMNA gene located on chromosome 1. Of 19 PGT cycles performed for CMD1A, 26 embryos free of the predisposing gene were detected for transfer in 17 cycles, resulting in the birth of 10 children with no risk of developing CD. This CD is characterized by ventricular dilation and impaired systolic function, resulting in heart failure and arrhythmia that may cause premature or sudden death. While the large phenotypic variability of patients may be determined by various mutations in the LMNA gene, differences from one family to another may also be observed within the same mutation.

The list of Mendelian disorders for which PGT-M was performed now comprises over 600 different conditions, with the most frequent ones shifting to common conditions with genetic predisposition, such as cancer, late-onset neurodegenerative conditions, and CD. The risk of having offspring with severe late-onset common disorders of strong genetic predisposition is an increasingly accepted indication for PGT-M. The spectrum of referral to PGT-M has also changed, with the current shift to direct referral through ECS [10]. It is understood that the preference of PGT in contrast to prenatal diagnosis is because prenatal diagnosis could lead to pregnancy termination that was not considered well justified based on genetic predisposition alone. That is, the tested fetus or embryo was not certain to become affected by CD. On the other hand, choosing the embryos free of genetic predisposition would obviate the need for considering pregnancy termination, as only potentially normal pregnancies are established. PGT for such conditions gained acceptability on ethical grounds because only a limited number of embryos (presently only one) is selected for transfer anyway. Thus, it is not surprising that the number of PGT requests for inherited predisposition for common late-onset conditions has been increasing overall. One of the main reasons to request PGT for inherited CD is that they often manifest despite pre-symptomatic diagnosis, while PGT may provide the only alternative for some at-risk couples to reproduce and avoid clinical termination of an affected fetus.

As in PGT for other common disorders, an inherited predisposition to CD may not be realized throughout a patient's entire lifetime. This can make the application of PGT for CDs controversial, perhaps explaining the still limited application of PGT for these conditions. But it is of note that most inherited CDs are dominant, and without available cure to prevent the realization of CD, the first and sometimes only clinical occurrence being a premature or sudden death, provoked by factors such as excessive exercise.

Among the conditions in the family history of the couples at risk that may indicate a possible need for PGT may be a heart attack and sudden death at a young age, family members with pacemakers or internal cardiac defibrillators, arrhythmia, and cardiac surgery. The likelihood that the offspring of these patients will develop the same heart disease will depend on the mode of inheritance, but penetrance is difficult to predict because many inherited cardiac conditions escape diagnosis, manifesting at different ages. As mentioned, disease may also be induced by certain factors, such as medications or excessive exercise that may lead to cardiac arrest or sudden death. All these justly requests for PGT-M, as in some cases, even a common, apparently "milder" disease susceptibility gene may contribute to premature death or major disability. Personal experience may alter a perception of the severity of the condition, in favor of a family member toward a decision to undertake PGT. As mentioned, among the patients presenting for PGT-M were not only those diagnosed with CD but also those with information of such conditions in their extended families, with the most recent addition of carriers of CD predisposition gene mutations detected through ECS [10–13]. Without prospective detection of the carriers of CD predisposing genes, more than half of the patients who may develop CD due to inherited predisposition may be missed with major implications for family members.

Because symptoms of inherited CD may be easily overlooked, findings in the family history may alone provide the reason to test for the presence of predisposing gene mutations or the need for PGT. However, in many instances, information on family history is not available or incomplete, so ECS may be the only opportunity to detect predisposition to CD, which could also pick up de novo predisposing variants. In addition, as mentioned, because most of these conditions are dominant, the detection of predisposing genes even in one family member is an indication of PGT. This may be a life-saving procedure for the offspring of individuals at risk. With future advances in the

identification of genes predisposing to inherited CD, PGT might appear as a useful tool for couples at risk for producing offspring with inherited CD that have a high probability of premature or sudden death during their life span.

According to our overall PGT-M experience, an increase in referrals for PGT-M through positive ECS was observed also for other genes, compared to the baseline referrals through the traditional approach, with the number of prospective PGT-M cases for the last seven years more than doubling through a referral from ECS [10]. This may become the major source for performing PGT-M in the near future allowing to offer PGT-M prospectively before the birth of an affected child.

With a low treatment efficiency for therapeutic interventions and a high (25–50%) risk of recurrence in each pregnancy, PGT-M may be a practical tool for primary prevention of CD. However, at-risk couples usually do not know about their at-risk status until they have an affected child, so reduction of affected birth prevalence cannot be done in the absence of screening programs, which have been offered initially for special ethnic groups or selected genetic conditions [11, 12]. But in contrast to such screening programs, ECS is offered in a pan-ethnic fashion, extending surveillance to couples of all ethnicities and involving an increasing number of genetic conditions, including CD-predisposing genes [13–15], which may have a growing impact on a wider PGT-M application. Targeted gene panels further increase the number of genetic disorders amenable to ECS, leading to PGT-M application prospectively, representing a form of primary prevention, as it allows for avoiding affected pregnancies, and establishing unaffected pregnancies from the onset. Several hundred additional genes are becoming considered plausible candidates for inclusion into ECS, with expanded panels now consisting of hundreds of genes, resulting in an increase in detection rate [14–16]. This will shift the application of PGT-M from a retrospective to a prospective approach, thus beyond the family level, as a tool for primary prevention of inherited predisposition to CD.

References

1. He J, McDermott DA, Song Y, et al. Preimplantation genetic diagnosis of human congenital heart malformation and Holt-Oram syndrome. Am J Med Genet A. 2004;126A(1):93-98.

2. Kuliev A, Pomerantseva E, Polling D, et al. PGD for inherited cardiac diseases. Reprod Biomed Online. 2012;24(4):443-53.

3. Kuliev A, Pakhalchuk T, Rechitsky S. Pre-implantation genetic diagnosis (pgd) for heart disease determined by genetic factors. Interv Cardiol. 2016;8(1):595-99.

4. Kuliev A, Pakhalchuk T, Prokhorovich M, et al. Preimplantation Genetic Testing for Inherited Predisposition to Cardiac Disease. Ann Heart. 2021;5(1):96-99.

5. Kuliev A, Rechitsky S, Simpson JL. Practical Preimplantation Genetic Testing. 3rd ed. Switzerland: Springer Nature; 2020.

6. Kuliev A, Rechitsky S, Verlinsky O. Atlas of Preimplantation Genetic Diagnosis. 3rd ed. London: CRC Press; 2014.

7. Preimplantation Genetic Diagnosis International Society (PGDIS). Guidelines for good practice in PGD: programme requirements and laboratory quality assurance. Reprod Biomed Online. 2008;16(1):134-47.

8. ESHRE PGT Consortium Steering Committee, Carvalho F, Coonen E, et al. ESHRE PGT Consortium good practice recommendations for the organisation of PGT. Hum Reprod Open. 2020;2020(3):hoaa021.

9. Rechitsky S, Pakhalchuk T, San Ramos G, et al. First systematic experience of preimplantation genetic diagnosis for single-gene disorders, and/or preimplantation human leukocyte antigen typing, combined with 24-chromosome aneuploidy testing. Fertil Steril. 2015;103(2):503-12.

10. Simpson JL, Rechitsky S, Kuliev A. Before the beginning: the genetic risk of a couple aiming to conceive. Fertil Steril. 2019;112(4):622-30.

11. ACOG Committee on Obstetrics. ACOG Practice Bulletin No. 78: hemoglobinopathies in pregnancy. Obstet Gynecol. 2007;109(1):229-37.

12. Committee Opinion No. 691: Carrier Screening for Genetic Conditions. Obstet Gynecol. 2017;129(3):e41-e55.

13. Edwards JG, Feldman G, Goldberg J, et al. Expanded carrier screening in reproductive medicine-points to consider: a joint statement of the American College of Medical Genetics and Genomics, American College of Obstetricians and Gynecologists, National Society of Genetic Counselors, Perinatal Quality Foundation, and Society for Maternal-Fetal Medicine. Obstet Gynecol. 2015;125(3):653-62.

14. Miyatake S, Koshimizu E, Fujita A, et al. Detecting copy-number variations in whole-exome sequencing data using the eXome Hidden Markov Model: an 'exome-first' approach. J Hum Genet. 2015;60(4):175-82.

15. Haque IS, Lazarin GA, Kang HP, et al. Modeled Fetal Risk of Genetic Diseases Identified by Expanded Carrier Screening. JAMA. 2016;316(7):734-42.

16. Hogan GJ, Vysotskaia VS, Beauchamp KA, et al. Validation of an Expanded Carrier Screen that Optimizes Sensitivity via Full-Exon Sequencing and Panel-wide Copy Number Variant Identification. Clin Chem. 2018;64(7):1063-073.

To access the full-text version of this article, please scan the QR code: