Original Article

J Genet Med 2022;19(1):7-13
https://doi.org/10.5734/JGM.2022.19.1.7
ISSN 1226-1769 (Print) 2383-8442 (Online)

Check for updates

Clinical application of genome-wide single nucleotide polymorphism genotyping and karyomapping for preimplantation genetic testing of Charcot–Marie–Tooth disease

Min Jee Kim[1,2], Sun Ok Park[2], Ye Seul Hong[2], Eun A Park[1,3], Yu Bin Lee[4], Byung-Ok Choi[5], Kyung-Ah Lee[1], Eun Jeong Yu[4], and Inn Soo Kang[6,*]

[1]Department of Biomedical Sciences, College of Life Sciences, CHA University, Seongnam, Korea
[2]Laboratory of Reproductive Genetics, CHA Biotech, Seoul, Korea
[3]Fertility Research Lab, CHA Fertility Center Seoul Station, Seoul, Korea
[4]Department of Obstetrics and Gynecology, CHA Fertility Center Seoul Station, CHA University School of Medicine, Seoul, Korea
[5]Department of Neurology, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea
[6]Department of Obstetrics and Gynecology, CHA Fertility Center Daegu, CHA University School of Medicine, Daegu, Korea

Purpose: Preimplantation genetic testing for monogenic disorders (PGT-M) has been successfully used to prevent couples with monogenic disorders from passing them on to their child. Charcot–Marie–Tooth Disease (CMT) is a genetic disorder characterized by progressive extremity muscle degeneration and loss of sensory function. For the first time in Korea, we report our experience of applying single nucleotide polymorphism genotyping and karyomapping for PGT-M of CMT disease.
Materials and Methods: Prior to clinical PGT-M, preclinical tests were performed using genotypes of affected families to identify informative single-nucleotide polymorphisms associated with mutant alleles. We performed five cycles of *in vitro* fertilization PGT-M in four couples with CMT1A, CMT2A, and CMT2S in CHA Fertility Center, Seoul Station.
Results: From July 2020 through August 2021, five cycles of PGT-M with karyomapping in four cases with CMT1 and CMT2 were analyzed retrospectively. A total of 17 blastocysts were biopsied and 15 embryos were successfully diagnosed (88.2%). Ten out of 15 embryos were diagnosed as unaffected (66.7%). Five cycles of PGT-M resulted in four transfer cycles, in which four embryos were transferred. Three clinical pregnancies were achieved (75%) and the prenatal diagnosis by amniocentesis for all three women confirmed PGT-M of karyomapping. One woman delivered a healthy baby uneventfully and two pregnancies are currently ongoing.
Conclusion: This is the first report in Korea on the application of karyomapping in PGT-M for CMT patients. This study shows that karyomapping is an efficient, reliable and accurate diagnostic method for PGT-M in various types of CMT diseases.

Key words: Preimplantation genetic testing, Charcot–Marie–Tooth Disease, Single nucleotide polymorphism.

Received: 18 November 2021, Revised: 22 May 2022, Accepted: 22 May 2022, Published: 30 June 2022
*Corresponding author: Inn Soo Kang, M.D., Ph.D. https://orcid.org/0000-0001-8551-3355
Department of Obstetrics and Gynecology, CHA Fertility Center Daegu, CHA University School of Medicine, 2095 Dalgubeol-daero, Jung-gu, Daegu 41936, Korea.
Tel: +82-53-222-4200, Fax: +82-53-214-6611, E-mail: ikang67pgd@gmail.com
Conflict of interest: The authors declare that they do not have any conflicts of interest.

This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/4.0/) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.
© Copyright 2022 by the Korean Society of Medical Genetics and Genomics

Introduction

Charcot–Marie–Tooth disease (CMT) is a genetically and clinically heterogeneous disorder of the peripheral nervous system. CMT is characterized by progressive limb muscle degeneration and loss of sensory function. It is mainly divided into CMT type 1 and CMT type 2 according to the clinical phenotype, genetic cause, and mode of inheritance [1-3]. To date, more than 80 genes have been reported to be associated with CMT. Of these, 74 CMT-associated genes are known, including inheritance mode and neuropathy type (https://www.ncbi.nlm.nih.gov/books/NBK1358) [4]. We performed preimplantation genetic testing for monogenic disorders (PGT-M) for CMT1A, CMT2A and CMT2S among various CMT types. Seventy to eighty five percent of CMT patients are CMT1A (Online Mendelian Inheritance in Man [OMIM] 118220) with a 1.5 Mb duplication in the p11-p12 region of chromosome 17 containing the *PMP22* (peripheral myelin protein 22) gene [3-7]. Another form of CMT2 that maps to chromosome 1p36.2 are CMT2A (OMIM 609260), which results from a heterozygous mutation in the *MFN2* gene [8]. CMT2S (OMIM 616155) is caused by a homozygous or compound heterozygous mutation in the *IGHMBP2* gene on chromosome 11q13 [9].

PGT-M has been successfully used as an alternative to prenatal testing to prevent couples with specific genetic disorders in the family from passing them on to their child [10]. After 5 days of culture after *in vitro* fertilization (IVF) or intracytoplasmic sperm injection, embryos identified as unaffected by biopsy of five to seven trophoblast cells from the blastocyst are transfer to the uterus. The use of PGT-M allows high-risk patients to avoid termination of affected pregnancy or births of affected child. Conventional polymerase chain reaction (PCR) PGT-M, which uses short tandem repeat (STR) markers, has limited information near the target locus, and optimization of multiplex PCR at the single-cell level requires a significant amount of laboratory work and takes about six months [11].

Karyomapping has been proposed as an alternative to the conventional PGT-M. Karyomapping performs genome-wide linkage analysis by genotyping parents and embryos using over 300,000 single-nucleotide polymorphisms (SNPs) across the entire genome [12,13]. Karyomapping can be used for the diagnosis of any single gene disorders without designing a patient-specific method, significantly reducing the time required for optimization prior to PGT-M. Therefore, unlike conventional PGT-M, detecting single gene disorders is a faster, more efficient and more reliable method [14].

Herein, we applied genome-wide linkage analysis and karyomapping for PGT-M of CMT disease for the first time in Korea.

Materials and Methods

1. Patient description

Four patients were diagnosed with CMT at Samsung Medical Center were referred to our clinic, Genetics Laboratory, CHA Fertility Center, Seoul station for PGT-M.

From July 2020 through August 2021, five cycles of PGT-M using karyomapping in four cases with CMT1 and CMT2 were analyzed retrospectively. PGT-M was requested by two cases for CMT1A, one case for CMT2A, and one case for CMT2S. Patients' demographic and medical information, including laboratory and clinical results, were extracted from medical charts.

In Case 1, CMT1A is an autosomal-dominant disorder, with duplication of *PMP22* detected in the female partner, who had inherited the mutation from her affected father. In Case 2, also having duplication of *PMP22* in the female partner with the CMT1A inherited from the affected father. In Case 3 with CMT2A type, an autosomal dominant disease, the mutation in the *MFN2* c.1082A>T in female partner was inherited from affected father. Case 4 with an autosomal recessive disease, CMT2S, had an affected daughter with compound heterozygous mutations of c.712-9G>A and c.257-2A>G in the *IGHMBP2* gene. It was confirmed that the female partner and the male partner had c.712-9G>A

Table 1. Genetic information for cases with CMT1A, CMT2A and CMT2S

Couple	Affected partner	Female's age (yr)	Type	Inheritance mode	Gene	Variant	Type
1	Female	28	CMT1A	AD	*PMP22*	17p11-p12 duplication	Pathogenic
2	Female	36	CMT1A	AD	*PMP22*	17p11-p12 duplication	Pathogenic
3	Female	30	CMT2A	AD	*MFN2*	c.1082A>T	Pathogenic
4	Female, male	29	CMT2S	AR	*IGHMBP2*	c.712-9G>A, c.257-2A	VUS, likely pathogenic

CMT1A, Charcot–Marie–Tooth disease type 1A; CMT2A, CMT type 2A; CMT2S, CMT type 2S; AD, autosomal dominant; AR, autosomal recessive; *PMP22*, peripheral myelin protein 22, *MFN2*, mitochondrial protein mitofusion-2; *IGHMBP2*, immunoglobulin mu-binding protein 2; VUS, variant of uncertain significance.

and c.257-2A>G of the *IGHMBP2* gene, respectively (Table 1).

2. Work up for haplotype analysis

Patients and family members received appropriate genetic counseling and informed consent was obtained for all procedures to be performed. DNA extraction from ethylene-diamine-tetraacetic acid-treated peripheral blood of all family members was performed within 1 week after blood collection using the Quickgene DNA whole blood kit according to the manufacturer's instructions (SHIMADZU, Neyagawa, Japan).

For preclinical test for CMT, DNA samples from additional family members used as references for phasing of SNP alleles included the couple's sons/daughters or the parents of male and/or female patients. DNA samples from couples and other families were analyzed using the HumanKaryomap-12 Bead Chip according to the manufacturer's protocol (Illumina, San Diego, CA, USA), and karyomapping for each single gene disorder was performed using dedicated software (BlueFuse Multi V4.5, Illumina).

3. Embryo biopsy and whole genome amplification

After IVF, embryos were cultured to the blastocyst stage and biopsied using laser micromanipulation in the morning on day 5 and approximately five to seven trophectoderm (TE) cells were obtained. Morphological grading of blastocysts was assessed according to Schoolcraft and Gardner guideline [15]. The biopsied cells were washed in three to four microdroplets of PBS (Phosphate-Buffered Saline, without magnesium or calcium) and transferred to a sterile PCR tube containing two μL of PBS. After biopsy, all embryos were vitrified and stored. And the biopsied TE cells were subjected to PGT-M. Whole genome amplification (WGA) was performed using the multiple displacement amplification and according to the standard protocol provided by the REPLI-g Single Cell Kit (QIAGEN, Hilden, Germany). WGA could sufficiently obtain genomic DNA by amplifying from the picogram level to the nanogram level for karyomapping experiments.

4. Karyomapping

Infinium Human Karyomap-12 DNA analysis kit (Cat#1500055; Illumina) was used for karyomapping, and fragmentation, precipitation, resuspension, hybridization, washing, and staining were performed according to the manufacturer's protocol. Karyomapping data was scanned using Illumina NextSeq and analyzed using BlueFuse Multi software (Illumina). Using the genotype of the affected family as a reference, haploblock was made using informative SNPs of the male partner and female partner. Using reference, the mutant allele can be identified in the haploblock. According to the genotype of TE cells, heterozygous SNP reads were applied in the 2 Mb region near the mutant gene to identify mutant alleles in the blastocyst [12].

5. Embryo transfer and *in vitro* fertilization outcomes

After genetic counseling on the results of PGT-M, single frozen-thawed unaffected blastocyst was transferred back to the mother's uterus after the endometrium preparation with artificial hormonal cycle. Clinical pregnancy was defined as the presence of a gestational sac or fetal heartbeat on ultrasonogram at 6 to 7 weeks of gestation.

6. Prenatal diagnosis

Amniocentesis was performed under ultrasound guidance according to standard techniques at 16 to 18 weeks of gestation. For prenatal diagnosis using amniotic fluid cells, linkage analysis or direct sequencing, karyomapping, and karyotype analysis were performed to confirm the PGT-M results.

Table 2. The informative SNPs identified by preclinical test for cases with CMT1A, CMT2A and CMT2S

Variable	Case 1	Case 2	Case 3	Case 4
Type	CMT1A	CMT1A	CMT2A	CMT2S
Affected partner	Female	Female	Female	Female, male
Reference	Affected father	Affected father	Affected father	Affected daughter
Target gene	*PMP22*	*PMP22*	*MFN2*	*IGHMBP2*
5' region	19/351	26/351	28/350	29/76
Main region	0/15	0/15	0/6	0/2
3' region	22/233	4/233	9/200	77/202

SNP, single nucleotide polymorphism; CMT1A, Charcot–Marie–Tooth disease type 1A; CMT2A, CMT disease type 2A; CMT2S, CMT disease type 2S; *PMP22*, peripheral myelin protein 22; *MFN2*, mitochondrial protein mitofusion-2; *IGHMBP2*, immunoglobulin mu-binding protein 2.

Table 3. The results of karyomapping according to embryo from cases with CMT1A, CMT2A, and CMT2S

Case	PGT cycle	Embryo	Grade	Karyomapping result		Embryo transfer	Clinical pregnancy	Amniocentesis	Pregnancy outcome
				CMT gene	Chromosome copy number				
1	1	#1	Exp. BL (BB)	Normal	Euploid	Transferred	Singleton	Normal	Live birth
		#2	Exp. BL (BB)	Abnormal	Euploid	Not transferred			
		#3	Mid. BL (BC)	Fail	Fail	Not transferred			
		#4	Mid. BL (BC)	Normal	Euploid	Cryopreservation			
		#5	Exp. BL (BC)	Abnormal	Euploid	Not transferred			
		#6	Hatching BL (BC)	Normal	Euploid	Cryopreservation			
2	1	#1	Mid. BL (BB)	Abnormal	Euploid	Not transferred			
		#2	Mid. BL (BB)	Normal	Partial monosomy 13	Not transferred			
		#3	Exp. BL (BB)	Normal	Euploid	Transferred	Singleton	Normal	Ongoing pregnancy
3	1	#1	Exp. BL (BC)	Normal	Euploid	Cryopreservation			
		#2	Mid. BL (BC)	Abnormal	Euploid	Not transferred			
	2	#1	Exp. BL (BB)	Normal	Euploid	Transferred	Singleton	Normal	Ongoing pregnancy
		#2	Mid. BL (BB)	Normal	Euploid	Cryopreservation			
		#3	Mid. BL (BB)	Normal	Monosomy 8	Not transferred			
		#4	Mid. BL (BC)	Abnormal	Haploid	Not transferred			
4	1	#1	Mid. BL (BC)	Fail	Fail	Not transferred			
		#2	Exp. BL (CB)	Normal	Euploid	Transferred	Biochemical pregnancy	_	_

CMT1A, Charcot–Marie–Tooth disease type 1A; CMT2A, CMT type 2A; CMT2S, CMT type 2S; PGT, preimplantation genetic testing; BL, blastocyst.

Results

1. Preclinical test using DNA of the affected family

The genomic DNA of the affected family was used to identify informative SNPs linked to the mutant allele (Table 2).

Case 1 was CMT1A, and the affected father of the female partner was a reference. The number of informative SNPs associated with *PMP22* gene on 5′ and 3′ flanking regions were 19 and 22, respectively. Case 2 was CMT1A, and the affected father of the female partner was a reference. Twenty-six and four informative SNPs associated with the *PMP22* gene were detected in the 5′ and 3′ flanking regions, respectively. Case 3 was CMT2A, and the affected father of the female partner was a reference. The number of informative SNPs associated with the *MFN2* gene in 5′ and 3′ flanking regions were 28 and nine, respectively. Case 4 was an autosomal recessive disease with CMT2S. The affected daughter of the couple was a reference. The number of informative SNPs associated with the *IGHMBP2* gene in the 5′ and 3′ flanking regions were 29 and 77, respectively.

2. Clinical PGT for CMT

After preclinical testing, PGT-M was applied to four cases with CMT (Table 3). In the DNA sample amplified with WGA, the SNP call rate was 94% on average (recommended SNP call rate of blastocyst biopsy by Illumina, 70-95), which is considered sufficient for karyomapping analysis. A total of 17 embryos were biopsied and 15 embryos were successfully diagnosed (88.2%). Ten out of 15 embryos were diagnosed as unaffected by CMT disease (66.7%). Two out of ten unaffected embryos were found to have chromosomal abnormalities and were excluded from embryo transfer. Five cycles with biopsy resulted in four transfer cycles, in which four embryos were transferred and three clinical pregnancies were achieved (75%).

Case 1, five out of six biopsied embryos were diagnosed successfully. One of three unaffected embryos was transferred, resulting in a singleton pregnancy. PGT-M was confirmed by amniocentesis and the healthy baby was delivered without any complications.

In Case 2, three embryos were biopsied and diagnosed. Two unaffected embryos were identified, and Embryo 2 was excluded due to the deletion of q14.3-qter of chromosome 13 (Fig. 1). Embryo 3 was transferred and successfully implanted. The PGT-M result was confirmed by amniocentesis, and have an ongoing pregnancy at 38 weeks.

https://doi.org/10.5734/JGM.2022.19.1.7 • J Genet Med 2022;19(1):7-13

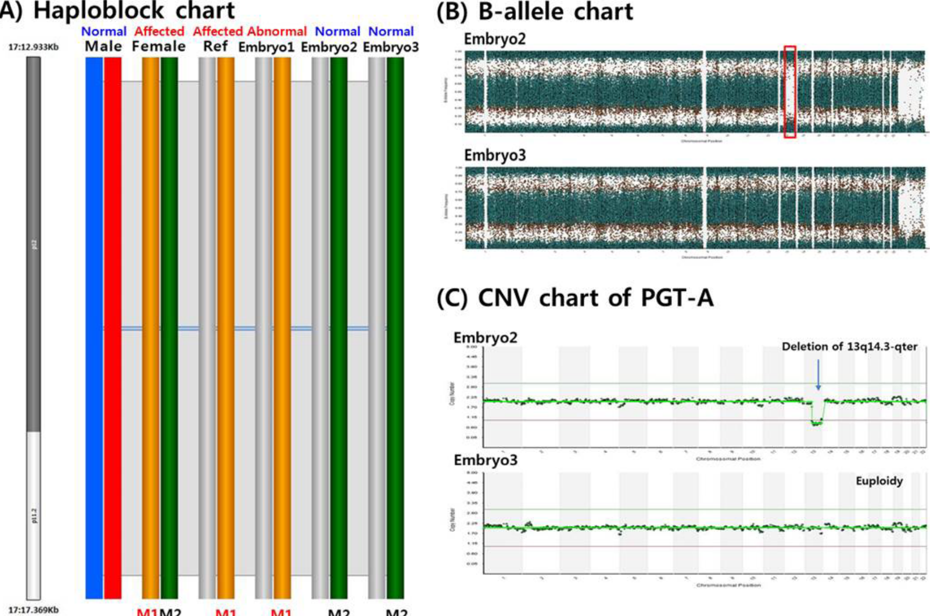

Fig. 1. Results of karyomapping and preimplantation genetic testing for aneuploidy (PGT-A) in case 2 with Charcot–Marie–Tooth disease type 1A. Karyomapping analysis included 2 Mb each of the upstream (5') and downstream (3') sides of the PMP22 gene for linkage analysis. (A) M1 is the mutant allele and M2 is the normal allele. Embryo 1 has mutant allele and embryo 2 and Embryo 3 have normal allele. (B) B allele chart of karyomapping in unaffected embryos. In Embryo 2, segment of chromosome 13 had AA and BB alleles but no AB alleles. A segmental monosomy13 was suggested in Embryo 2. (C) Result of PGT-A shows segmental monosomy on 13q14.3-q31 in embryo 2, and euploidy in Embryo 3. CNV, copy number variation.

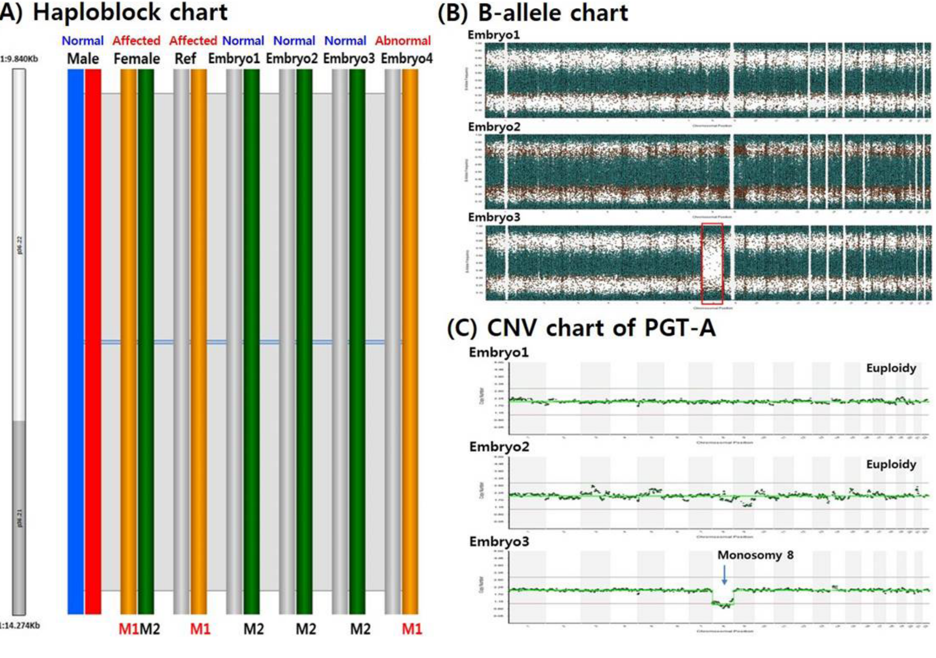

Fig. 2. Results of karyomapping and preimplantation genetic testing for aneuploidy (PGT-A) in case 3 with Charcot–Marie–Tooth disease type 2A. Karyomapping analysis included 2 Mb each of the upstream (5') and downstream (3') sides of the MFN2 gene for linkage analysis. (A) M1 is the mutant allele and M2 is the normal allele. Embryo 1-3 have normal alleles and Embryo 4 has a mutant allele. (B) B allele chart of karyomapping in unaffected embryos. In Embryo 3, chromosome 8 had AA and BB alleles but no AB alleles. A monosomy 8 was suggested in Embryo 3. (C) Result of PGT-A shows monosomy 8 in embryo 3 and euploidy in Embryo 1 and 2. CNV, copy number variation.

Case 3 underwent two IVF cycles and a total of six embryos were biopsied and diagnosed. Four unaffected embryos were identified, and Embryo 3 was excluded due to monosomy 8 (Fig. 2). One unaffected embryo was transferred and the successful pregnancy was achieved. Prenatal diagnosis by amniocentesis using karyomapping and direct sequencing were consistent with the PGT-M results.

For Case 4, two embryos were biopsied and one embryo was diagnosed. One unaffected embryo was transferred, resulting in a biochemical pregnancy.

Discussion

Karyomapping is an advanced method for PGT-M. It dramatically reduces time and work load required for optimization prior to PGT-M compared to conventional PCR methods, allowing us to make a diagnosis of single-gene disorders without designing patient-specific PCR methods [12]. It also reduces the waiting time for patients remarkably. In addition, the allele dropout (ADO) problem, which is one of the most challenging problem of the conventional PCR method, can be overcome with a univer-

sal test using genome-wide SNP markers and haplotyping at the mutation site and adjacent regions [16].

However, karyomapping also has its limitations. Karyomapping with SNP markers cannot be performed without DNA from an appropriate reference from the family. Therefore, karyomapping cannot be applied in couples with *de novo* mutations. And if chromosomal recombination occurs in the main region (causative gene) accurate diagnosis cannot be made [11]. In this report, all CMT patients had references, affected family members, fortunately and so the karyomapping could be applied.

In this study, prior to PGT-M, the preclinical test showed large number of informative SNPs ranging from 30 to 106 in 4 cases, which is considerably larger number than that of STR linked markers in the conventional PCR. Due to large number of informative SNPs, the karyomapping analysis would undoubtedly be much more accurate than PCR method, because large number of SNPs can overcome the problem of ADO of the linked markers. In addition, the probability of false negative results by homologous chromosome recombination in the vicinity of mutation site would be much lowered.

Karyomapping has the additional advantage of detecting the copy number of the chromosome compared to the PCR method [13]. As a result of the PGT-M, chromosomal aneuploidies were found in two out of ten CMT-unaffected embryos. Considering that aneuploidy is a major cause of implantation failure and miscarriage in early pregnancy [17,18], high pregnancy rate in this study, albeit small sample number, may have been achieved by excluding the embryos with abnormal chromosome from the transfer (Table 3).

In the present study, successful pregnancies and live birth were achieved by applying karyomapping in couples with CMT to prevent delivering an affected baby. We confirmed that karyomapping is a highly efficient and accurate diagnostic method for PGT-M in CMT diseases. In addition, the feasibility of simultaneous examination of single gene disorder and chromosomal aneuploidy was confirmed.

In conclusion, we provided further evidence that PGT-M using karyomapping is a reliable and effective clinical technique and is an advantageous option for couples at high risk of transmitting single gene disorders.

Authors' Contributions

Conception and design: ISK. Acquisition of data: MJK, SOP, YSH, EAP, YBL. Analysis and interpretation of data: MJK, SOP, YSH. Drafting the article: MJK, EJY. Critical revision of the article: ISK, EJY. Final approval of the version to be published: all authors.

References

1. Skre H. Genetic and clinical aspects of Charcot-Marie-Tooth's disease. Clin Genet 1974;6:98-118.
2. Patel PI, Lupski JR. Charcot-Marie-Tooth disease: a new paradigm for the mechanism of inherited disease. Trends Genet 1994;10:128-33.
3. Berciano J, Sevilla T, Casasnovas C, Sivera R, Vílchez JJ, Infante J, et al. [Guidelines for molecular diagnosis of Charcot-Marie-Tooth disease]. Neurologia 2012;27:169-78. Spanish.
4. Bird TD. Charcot-Marie-Tooth hereditary neuropathy overview. In: Adam MP, Ardinger HH, Pagon RA, Wallace SE, Bean LJH, Gripp KW, eds. GeneReviews®. Seattle (WA): University of Washington, Seattle, 1993-2022.
5. Braathen GJ. Genetic epidemiology of Charcot-Marie-Tooth disease. Acta Neurol Scand Suppl 2012;193:iv-22.
6. Hoogendijk JE, Hensels GW, Zorn I, Valentijn L, Janssen EA, de Visser M, et al. The duplication in Charcot-Marie-Tooth disease type 1a spans at least 1100 kb on chromosome 17p11.2. Hum Genet 1991;88:215-8.
7. Lupski JR, de Oca-Luna RM, Slaugenhaupt S, Pentao L, Guzzetta V, Trask BJ, et al. DNA duplication associated with Charcot-Marie-Tooth disease type 1A. Cell 1991;66:219-32.
8. Filadi R, Pendin D, Pizzo P. Mitofusin 2: from functions to disease. Cell Death Dis 2018;9:330.
9. Tomaselli PJ, Horga A, Rossor AM, Jaunmuktane Z, Cortese A, Blake JC, et al. IGHMBP2 mutation associated with organ-specific autonomic dysfunction. Neuromuscul Disord 2018;28:1012-5.
10. Grace J, El-Toukhy T, Scriven P, Ogilvie C, Pickering S, Lashwood A, et al. Three hundred and thirty cycles of preimplantation genetic diagnosis for serious genetic disease: clinical considerations affecting outcome. BJOG 2006;113:1393-401.
11. Wang CW, Liu YL, Chen CH. Targeting myotonic dystrophy by preimplantation genetic diagnosis-karyomapping. Taiwan J Obstet Gynecol 2019;58:891-4.
12. Handyside AH, Harton GL, Mariani B, Thornhill AR, Affara N, Shaw MA, et al. Karyomapping: a universal method for genome wide analysis of genetic disease based on mapping crossovers between parental haplotypes. J Med Genet 2010;47:651-8.
13. Natesan SA, Bladon AJ, Coskun S, Qubbaj W, Prates R, Munne S, et al. Genome-wide karyomapping accurately identifies the inheritance of single-gene defects in human preimplantation embryos in vitro. Genet Med 2014;16:838-45.
14. Giménez C, Sarasa J, Arjona C, Vilamajó E, Martínez-Pasarell O, Wheeler K, et al. Karyomapping allows preimplantation genetic di-

agnosis of a de-novo deletion undetectable using conventional PGD technology. Reprod Biomed Online 2015;31:770-5.
15. Gardner DK, Lane M, Stevens J, Schlenker T, Schoolcraft WB. Blastocyst score affects implantation and pregnancy outcome: towards a single blastocyst transfer. Fertil Steril 2000;73:1155-8.
16. Ben-Nagi J, Wells D, Doye K, Loutradi K, Exeter H, Drew E, et al. Karyomapping: a single centre's experience from application of methodology to ongoing pregnancy and live-birth rates. Reprod Biomed Online 2017;35:264-71.
17. Forman EJ, Tao X, Ferry KM, Taylor D, Treff NR, Scott RT Jr. Single embryo transfer with comprehensive chromosome screening results in improved ongoing pregnancy rates and decreased miscarriage rates. Hum Reprod 2012;27:1217-22.
18. Wells D, Delhanty JD. Comprehensive chromosomal analysis of human preimplantation embryos using whole genome amplification and single cell comparative genomic hybridization. Mol Hum Reprod 2000;6:1055-62.