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ARTICLE

Provision and quality assurance of preimplantation genetic diagnosis in Europe

Anniek Corveleyn[1,12], Michael A Morris[2,12], Elisabeth Dequeker[1,12], Karen Sermon[3], James Lawford Davies[4,5], Guillermo Antiñolo[6,7], Andreas Schmutzler[8], Jiri Vanecek[9], Nick Nagels[1], Eleni Zika[10], Francesc Palau[6,11] and Dolores Ibarreta*[,10]

[1]*Department for Human Genetics, University of Leuven, Leuven, Belgium;* [2]*Geneva University Hospitals, Service of Genetic Medicine, CMU, Geneva, Switzerland;* [3]*Centre for Medical Genetics, Unversitair Ziekenhuis Brussel (UZ Brussel) of the Vrije Universiteit Brussel (VUB), Brussels, Belgium;* [4]*Institute of Human Genetics, University of Newcastle, UK;* [5]*Clifford Chance LLP, London, UK;* [6]*Centre for Biomedical Research on Rare Diseases, Instituto de Salud Carlos III, Spain;* [7]*Unidad Clinica de Genetica y Reproduccion, Hospitales Universitarios Virgen del Rocio, Seville, Spain;* [8]*Christian-Albrechts University Kiel, Women's Hospital, Kiel, Germany;* [9]*Technology Centre of the Academy of Sciences CR, Prague 6, Czech Republic;* [10]*European Commission, Joint Research Center (JRC), Institute for Prospective Technological Studies (IPTS), Edificio Expo, Seville, Spain;* [11]*Institut de Biomedicina de València – CSIC, C/Jaume Roig, València, Spain*

Preimplantation genetic diagnosis (PGD) is now well established and provided in many European countries. However, regulations, professional standards and accreditation requirements can differ notably. Furthermore, no comprehensive independent data exist either about practice and provision in Europe or about the quality assurance practices and procedures designed to optimize the quality of the results. Consequently, a study was launched to obtain knowledge, currently lacking, of the provision and quality assurance of PGD services and cross-border activities in Europe. An online questionnaire was developed and sent to PGD providers, and expert opinions were obtained through interviews with professionals in specific countries. Information was gathered from 53 centres offering PGD in 17 European countries. There is a diverse array of tests available, with a trend for custom-made services. Although half of the centres have a designated quality manager, just 33% have achieved or are preparing for accreditation or certification. About 66% of the centres responded that they did not participate in external quality assessment, a problem exacerbated by the lack of existing PGD-specific schemes. Approximately 19% of the centres do not keep data on accuracy and 9% do not even follow up until birth. PGD is an expanding activity with an increasing international flow that accounts for approximately one-third of the activity reported. The survey highlights a significant need for improvement in quality assurance in PGD centres. On the positive side, important improvements in the quality management of these services are expected with the European Tissue Directive entering into force.
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***Correspondence: Dr D Ibarreta, Institute for Prospective Technological Studies (IPTS), European Commission, Joint Research Center (JRC), C/Inca Garcilaso S/N, Edificio Expo, Seville E-41092, Spain.**
Tel: 349 544 884 45; Fax: 349 544 884 34;
E-mail: eleni.zika@ec.europa.eu
[12]These three authors have contributed equally to this work.
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Introduction

Preimplantation genetic diagnosis (PGD) involves the genetic testing of embryos generated by *in vitro* fertilization (IVF), principally to assist couples at high risk of transmitting

a severe genetic disease to have unaffected children without needing to resort to prenatal diagnosis and potential termination.[1] Since its initial application for sexing in the early 1990s,[2] PGD has expanded in scope to include, for example, HLA typing for tissue donation or non-medical sex selection.

Although PGD is now a well-established treatment provided in many European countries, no current, comprehensive independent data exist about the practice and provision of PGD in Europe. As a result of differences in regulation, legislation and technical infrastructure between European countries,[3] patients and/or samples are often referred internationally, but data are lacking on the scale and nature of international flow as well as on access to PGD across Europe. The need for thorough quality assurance in assisted reproduction has been underlined for clinics, laboratories and treatment procedures, with the suggestion that European providers should have a formal quality system.[3,4] Despite this call and the recent publication of voluntary best practice guidelines for PGD,[5,6] little information exists on current practice. In response to these concerns, the European Commission's Joint Research Centre launched a study in collaboration with EuroGentest and ESHRE (involving mainly the ESHRE PGD Consortium), in which a survey was conducted to assess the provision of PGD services in Europe, the access by patients and the current state of quality assurance.

Materials and methods

A dynamic online questionnaire, using a dependency approach in which the questions asked change according to the answers given, was developed, containing distinct sections for the three classes of PGD service providers: IVF clinics (which receive patients and perform the medical procedures of IVF), IVF laboratories (which perform the laboratory procedures of IVF, including fertilization and culture) and PGD laboratories (which perform the genetic analyses) (http://www.jrc.es/publications/pub.cfm?id=1531).

Preimplantation genetic screening (PGS, formerly PGD-AS), which is performed to detect embryonic aneuploidy with the aim of improving pregnancy rates in couples undergoing IVF, is distinguished from PGD in Europe and was excluded from this study wherever possible.

Given the current lack of registries or official listings (other than the ESHRE PGD Consortium[7]), the distribution list was broadened to ensure the most comprehensive coverage possible of centres potentially performing PGD, with the effect that the response rate appears low (for details, see http://www.jrc.es/publications/pub.cfm?id=1531). The contact lists were compiled using the ESHRE PGD Consortium list, the ESHRE membership database and some medical genetics laboratories offering PGD previously identified by EuroGentest through their quality assurance survey. The survey was thus distributed to 169 known or likely PGD providers as well as to over 1500 IVF professionals.

On submission, the replies were automatically inserted into a database to simplify analysis. The responses are compared to other sources, where available,[5,7,8] and to expert opinions obtained through interviews conducted with professionals in specific countries.

To complement the information on current practices gathered by the survey and to obtain a more detailed picture of the situation, expert knowledge was sought through interviews conducted with genetic laboratories and IVF clinics offering PGD. Finally, to obtain a full understanding of how PGD is provided in Europe, EU regulations that impact upon the provision of PGD as well as regulatory frameworks in different European countries were examined in detail (manuscript in preparation).

Results

Who provides PGD?

We received 71 responses from the 169 centres approached (Table 1):

- 53 centres provided PGD:
  - 44 performed both PGD and IVF ('IVF + PGD')
  - 9 performed PGD only, receiving samples from an IVF laboratory ('PGD only')

Two categories of PGD non-providers also replied and were excluded from the study:

- 8 centres performed IVF but offered PGD, outsourcing the genetic analysis ('IVF + PGD referred');
- 10 replies were received mistakenly from centres that did not offer PGD (IVF only).

The 53 providers consisted of 141 laboratories and clinics (53 genetics laboratories, 44 IVF clinics and 44 IVF laboratories). They were asked 'How close is the PGD laboratory to the IVF clinic?' and 68 replies were received, as some laboratories and clinics work with multiple partners. In 58/68 cases, both partners were in the same institution or city. In eight cases (five IVF + PGD laboratories, three PGD only), the IVF clinics were in the 'same country' and in two cases (both PGD only), in different countries.

The greatest numbers of providers responded from Spain, Greece, Belgium, the Czech Republic and the United Kingdom, respectively. Overall, the centres were equally distributed between private (41%) and public (46%) settings (Table 1), although 'PGD only' centres were more likely to be in the private sector (78%). Most private centres were located in the Czech Republic, Greece, Spain and Turkey.

Table 1 Distribution of laboratories and PGD cycles per country

	PGD cycles 2005	All centers			PGD only			IVF+PGD			IVF+PGD referred		
		Private	Public	Total	Private	Public	Total	Private	Public	Total	Private	Public	Total
Belgium	~370	0	5	6					5	6			
Czech Republic	~240	3	1	6	1	0	1	2	1	5			
France	~300	0	3	3				0	3	3			
Germany	~110	1	2	4				1	2	3			1
Greece	~260	5	2	7	2	1	3	3	0	3	0	1	1
Netherlands	~150	0	3	3				0	3	3			
Spain	~530	9	1	10	3	0	3	4	1	5	2	0	2
Turkey	~300	3	1	4	1	0	1	2	0	2	0	1	1
United Kingdom	~180	1	4	6				1	3	5	0	1	1
Other	~420	3	6	12	0	1	1	1	5	9	2	0	2
Total numbers	2410 ± 450[a]	25 (41%)	28 (46%)	61[b]	7 (78%)	2 (22%)	9	14 (32%)	23 (52%)	44[b]	4 (50%)	3 (38%)	8[b]

The countries that participated in the survey are listed according to their activity and public/private status. PGD only: laboratory performing only PGD, receiving samples from an IVF laboratory; IVF+PGD: centre offering both laboratory services; IVF+PGD referred: IVF laboratories which act as gateways, collaborating with external PGD laboratories to offer PGD to their patients. Ten responding centres provided IVF only and were excluded from the study.

Bold text indicates the 'Top 7' countries which performed over 150 cycles in 2005. The eight countries with less than three PGD providers are grouped in 'other'.

[a]Forty-nine out of fifty-three PGD providers gave data on the number of cycles, which were calculated from ranges (possible answers '1–10, 11–20, 21–50, 51–100 and >100'); see text for details.

[b]Four IVF+PGD centres had mixed public/private affiliations, and four (three IVF+PGD and one IVF+PGD referred) did not reply.

Independent data are scarce on the scale of PGD use in Europe and consequently the approximate number of PGD cycles performed in 2005 was an important aspect of this survey. The largest centres, declaring over 100 cycles, were in Belgium, Cyprus, the Czech Republic, France, Greece, Spain and Turkey. With the exception of Cyprus (from which only one centre replied), this correlates with those countries with three or more centres (Table 1).

Forty-nine of the 53 PGD providers (92%) gave data on the number of cycles they performed in 2005, selecting from possible answers of '1–10, 11–20, 21–50, 51–100 and >100'. In this way, they declared a total of 2410 ± 450 PGD cycles, significantly above the estimate of approximately 1000 cycles annually worldwide.[3] A potential cause of overestimation in this figure is that although the survey was designed specifically to exclude PGS, some laboratories may nonetheless have included PGS in their self-declared data (this is probably the case for some German and Dutch laboratories; personal communication). An underestimate is more likely, as (a) the 12 large laboratories responding '>100 cycles' are conservatively counted for this calculation as only 100, (b) four centres did not provide numbers and (c) it is possible that not every centre performing PGD in Europe in 2005 was identified.

Thirty-nine of the 53 (74%) providers voluntarily contributed to the European Society of Human Reproduction and Embryology (ESHRE) PGD consortium and reported 1182 PGD (plus 1722 PGS and 80 social-sexing) cycles in 2004, the last data compilation available[7] at the moment of writing; thus, the data are of similar magnitude.

In a recent survey of IVF clinic directors in the United States, 137/186 IVF clinics reported that they have provided PGD services to patients, resulting in a total of approximately 1200 PGD cycles (plus approximately 2200 PGS) in 2005.[8] A major difference in organization is revealed by the fact that in the United States only 14% ($n = 19$) of the IVF–PGD clinics perform some PGD genetic analysis 'in-house', whereas the majority send biopsy samples to external laboratories.

What kinds of tests are available?

Testing for chromosomal anomalies is the most widespread service, offered by 48 laboratories in 17 countries (Figure 1a). Most of these laboratories also perform PGS, which requires similar FISH technology as well as sex selection for X-linked genetic disorders. Technical and clinical details of cytogenetic testing are thoroughly covered elsewhere and were not requested.[7]

Molecular genetic testing for monogenic disorders is offered by 37 laboratories from 15 countries; 34/37 also perform chromosomal testing, whereas 3 perform only monogenic PGD analysis. Thirty-five centres offered mutation detection in probands/parents, that is, performed genetic diagnosis outside PGD.

Similarly, centres were asked if they confirmed the identification of mutations and/or familial translocations when families were referred for PGD, which is important to guarantee the correct identification and assignment of the anomaly. Mutations were confirmed in the majority of cases: 34/49 responding laboratories confirmed 'in all cases', 11/49 in '>50%' and only 3 in '< 50%'. One laboratory 'always trusts an external report' and 4 did not respond.

a

	PGD labs
Chromosomal anomalies	48 (91%)
Sex selection for X-linked disorders	45 (85%)
Social sex selection	1 (2%)
PGS§	37 (70%)
Monogenic diseases	37 (70%)
Adult-onset monogenic diseases	24 (45%)
HLA matching	14 (26%)
Screen probands before PGD	35 (66%)

b

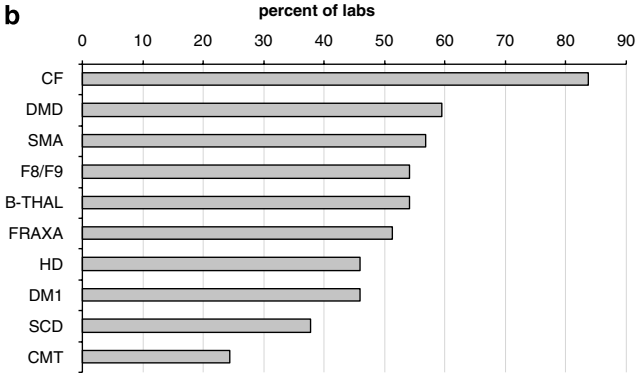

Figure 1 (**a**) PGD services available in the 53 PGD laboratories. § PGS cycles were excluded from the presented data wherever possible (see text). (**b**) Availability of testing for 10 common monogenic diseases. The 10 most common indications for monogenic disease PGD in 2005, in the 37 laboratories providing monogenic PGD. Legend: CF, cystic fibrosis; DMD, Duchenne/Becker muscular dystrophy; SMA, spinal muscular atrophy; F8/F9, haemophilia A/B; B-THAL, β-thalassemia; FRAXA, Fragile X syndrome; HD, Huntington disease; DM1, Steinert myotonic dystrophy; SCD, sickle-cell disease; CMT, Charcot-Marie-Tooth disease.

The 'top 10' monogenic diseases (Figure 1b) are almost identical to those reported in 2004.[7] Most are potentially severe and of early onset and represent common indications for prenatal diagnosis. On average, each laboratory offered 5.1 of the top 10 tests, increasing to 5.6 in those countries reporting over 150 cycles in 2005 (the 'top 7', see Table 1) with Turkey, France and Belgium having the highest counts (9.0, 8.0 and 7.7, respectively). Each of the top 10 tests was offered by an average of 1.6 laboratories per country, indicating that overall there is little redundancy in the provision of services; however, in the top 7 countries, this increases to 2.3 laboratories per test, with Spain and Belgium having the highest overlaps with 4.4 and 2.4 laboratories performing each test, respectively.

Approximately 50 distinct monogenic disorders were cited as indications for PGD (Table 2), with some laboratories responding with 'more than 50 indications' or a 'custom-made analysis of any disease of known genetic cause'. Thirty-five of these disorders appear in the list of 57 conditions reported in 2004.[7]

Overall, these data reveal a clear tendency for laboratories to offer custom-made tests with little specialization in or coordinated repartition of, particular diseases within countries.

Table 2 Indications for monogenic PGD reported in the survey

Autosomal dominant	*Autosomal recessive*	*X-linked*
Achondroplasia	β-Thalassemia	Duchenne/ Becker MD
Angelman/UBE3A	Cystic fibrosis	Fragile X
BRCA1 and BRCA2	Epidermolysis bullosa	Hemophilia A, B
'Cancer predisposition'[a]	Diastrophic dysplasia	Incontinentia pigmenti
Charcot-Marie-Tooth disease	Kell isoimmunization	Lesch–Nyhan syndrome
Crouzon syndrome	Krabbe disease	Leukodystrophy, metachromatic
Ehlers–Danlos syndrome		Lowe syndrome
Familial amyloidosis		Tyrosine hydroxylase deficiency
Hypochondroplasia		
Huntington's disease	San Filippo (MPS III)	Wiscott–Aldrich syndrome
Von Hippel–Landau		
Marfan syndrome	Sickle-cell disease	NARP (mtDNA)
Multiple endocrine neoplasia, 2	Spinal muscular atrophy	Leigh's syndrome
Myotonic dystrophy type 1	Tay–Sachs disease	
Noonan syndrome		
Neurofibromatosis 1 and 2	Polycystic kidney disease	
Pancreatitis, hereditary		
Polycystic kidney disease		
Polyposis coli (APC)[a]		
Retinoblastoma		
Spinocerebellar atrophy 1, 2, 3, 7[a]		
Tuberous sclerosis 1, 2		

Approximately 50 distinct pathologies/indications for which PGD centres offered tests in 2005; in addition, some centres replied 'more than 50 indications' or 'custom-made analysis of any disease of known genetic cause'.
[a]Onset usually in adult life.

The different perception and application of PGD compared with prenatal diagnosis becomes apparent with the widespread availability of testing for adult-onset disorders in 24 of the 37 laboratories doing monogenic PGD. Conditions notably included Huntington (HD) and Charcot-Marie-Tooth (CMT) disease, offered by 17 and 9 laboratories, respectively, plus many familial cancer predispositions. Prenatal testing for HD is available in genetics centres worldwide,[9] although it continues to incite debate because of its late onset and because of issues of consent and privacy – notably, prenatal diagnosis can reveal the status of an untested parent, an issue that can be avoided with PGD. Prenatal diagnosis for CMT is uncommon owing to its relatively benign course and clinical heterogeneity.[10] Similarly, prenatal diagnosis is uncommon for inherited

predispositions to breast, ovarian or bowel cancers, which are not only typically adult-onset but also have incomplete penetrance; PGD offers an alternative that may reduce the ethical questions but nonetheless remains controversial.[11,12] Such testing was authorized in the United Kingdom in 2006, subject to licensing.[13]

The arrival of PGD has also led to the emergence of certain non-diagnostic tests that were almost universally considered as unacceptable in the context of prenatal diagnosis. Embryonic HLA typing, to select HLA-identical donors for siblings with a severe existing disease, represents such a non-diagnostic application of PGD; medical, ethical and legal aspects of preimplantation HLA typing have been discussed recently.[14–16] Typing was provided by 14 laboratories (26%), either simultaneously with PGD for a monogenic disorder treatable with bone marrow transplant or in the absence of a concomitant genetic disorder, for example, in case of acquired haematological disease (performed by 12/14 and 11/14 laboratories, respectively). In the United States, 24% of laboratories perform pre-implantation HLA testing but only 6% do so in the absence of a concomitant inherited disorder.[8]

Non-medical (social) sex selection is a controversial subject in Europe and is prohibited or actively discouraged in some but not all European countries,[17] and only one laboratory responded that it performed social sex selection. This is in marked contrast to the United States, where such testing is considered more acceptable and where 42% of PGD clinics offer non-medical sex selection.[8]

How is the quality of PGD assured?

Assuring patient safety through the quality of results is mission-critical to medical laboratories. Accreditation, according to an internationally recognized standard such as ISO 15189, is the single most effective route to comprehensive quality assurance, attesting to both technical competence and compliance with quality standards. Accreditation is a formal recognition of a laboratory's competence to perform a test. Certification, which attests only compliance, is a less discerning alternative. Licensing is an official or legal permission to perform testing that is required in some countries.

Quality management Despite this and the call for European PGD providers to be certified or accredited,[3,4] the uptake of formal systems of quality management or of their different elements remains low (Table 3). Only 77/141 centres (55%) have a designated quality manager, an essential early step in implementing a quality system, and just 46/141 (33%) have achieved or are preparing for accreditation or certification. Some countries additionally maintain a licensing and inspection system that may fulfil certain aspects of accreditation standards; only 11% of all laboratories and clinics (15/141) were licensed.

Uptake of accreditation is significantly stronger in the private sector: 11/15 accredited laboratories and clinics (73%) and 8/16 (50%) centres working towards accreditation or certification are private. Similarly, larger centres are more likely to adopt a formal quality system: 7/22 (32%) centres that performed over 50 cycles are accredited and 16/22 (73%) have obtained or are preparing for accreditation/certification, compared with only 4/27 (15%) and 10/27 (37%), respectively, of smaller centres. Appropriately, given the different priorities of laboratory testing and patient care, IVF laboratories prefer accreditation to certification, whereas IVF clinics prefer the inverse; genetics laboratories do not follow this choice but opt at similar frequencies for certification or accreditation.

External quality assessment Although accreditation standards require quality control procedures for monitoring the validity of tests, including objective assessment of laboratory performance by external quality assessment (EQA), there is at present no EQA scheme available specifically for PGD. ESHRE recommended that a voluntary EQA scheme be implemented to ensure that related technical aspects, interpretation and reporting of the results are well assessed.[5] Thirty-five of 53 laboratories, and even 4 of the 7 accredited genetics laboratories, responded that they did not participate in EQA, although it was unclear if this concerned EQA in general or specifically for PGD. When asked to rate the importance of EQA, 48/49 laboratories responded with 'very important' or 'important'. A single laboratory replied that EQA was 'irrelevant'.

Table 3 Quality management systems

	All	*IVF clinics (n = 44)*	*IVF laboratories (n = 44)*	*Genetics laboratories (n = 53)*
Quality manager	55%	52% (23)	70% (31)	43% (23)
Accreditation	17%	7% (3)	20% (9)	23% (12)
Certification	17%	27% (12)	9% (4)	15% (8)
Accreditation and/or certification[a]	33%	34% (15)	30% (13)	34% (18)

Providers were asked to indicate if they had a designated quality manager, and if accreditation and/or certification had been obtained or was underway (the latter values are combined under accreditation, certification and accreditation and/or certification). The number of answers in each category appears in parentheses.
[a]Some centres replied positively to both accreditation and certification.

Table 4 Qualifications of directors

	IVF clinics (37)	*IVF laboratories (37)*	*Genetics laboratories (44)*
PhD	8% (3)	65% (24)	57% (25)
MD/ PhD	32% (12)	8% (3)	20% (9)
MD	59% (22)	5% (2)	20% (9)
Other	0	22% (8)	2% (1)

The qualifications of the clinical and laboratory directors were asked; replies were obtained from 84% of clinics and laboratories providing PGD. The number of answers in each category appears in brackets. 'Other' is comprised of different types of pre-doctoral degrees.

Qualifications The presence of directors with a PhD degree has been previously identified as one of the major positive quality indicators in molecular genetics laboratories,[18] and the OECD recommends that the minimum qualification to direct a molecular genetics laboratory be an MD, PhD or equivalent.[19] Current PGD guidelines make no recommendations on this subject.[5,6] The majority of genetics and IVF laboratories were directed principally by PhDs or MDs (Table 4), whereas clinics are logically directed principally by MDs. Eight IVF laboratories (22%) and one genetics laboratory were directed by people with neither PhD nor MD degrees but who had only Masters or similar graduate degrees. Most centres had formal instructions documenting the training of staff (41/53).

Technical aspects A series of technical questions in the survey allowed the identification of common practices in PGD laboratories (Table 5), which were generally in close agreement with the ESHRE guidelines and similar to common practice in the United States.[5,8] The consensus approach was as follows:

- 1 or 2 blastomeres are analysed;
- blastomeres are biopsied on day 3, from the 5 or 6 cell stage onwards;
- biopsy is performed by a biologist/embryologist, either alone or with a technician;
- PGD embryos are transferred 1–2 days after biopsy.

Notable exceptions from consensus practice included the analysis of polar bodies in Austria, Germany and Switzerland, in response to the prohibition of direct embryo testing. Several centres in Estonia, Greece and Turkey only biopsy blastomeres from the seven-cell stage onwards; it is generally true that there is only a very small chance of successful implantation of embryos that have only five or six cells at day 3.

Two-thirds of laboratories performing monogenic PGD (24/36) perform both direct and indirect mutation analysis, improving the reliability of testing by eliminating the risk of errors due to allele drop-out (testing only one of the two alleles owing to a PCR artefact), but 10 (28%) reported that they only perform direct analysis. ESHRE guidelines clearly call for both direct and indirect analysis.[5]

The existence and use of strictly separated working zones within the laboratory is of prime importance to avoid contamination in PCR-based testing, particularly in the context of PGD by single-cell analysis, and are recommended by the ESHRE guidelines. Of the 37 laboratories performing monogenic PGD, 20 (54%) had dedicated pre-pre-, pre- and post-PCR rooms; a further 12 (32%) had either pre- or post-PCR rooms but 5 had no separated facilities. Positive and negative controls for proving reliability

Table 5 Technical aspects of quality assurance in laboratories performing PGD

	All PGD laboratories (n = 53[a])	*Top 7 countries (n = 37)*
1. Cells analysed per embryo per diagnosis?		
1 blastomere	68% (36)	70% (26)
2 blastomeres	57% (30)	59% (22)
1 polar body[b]	4% (2)	—
2 polar bodies	8% (4)	—
(a) On which day do you usually perform blastomere biopsy?		
Day 3	100% (48)	100% (37)
(b) Which embryos do you biopsy?		
From 5 cells	52% (25)	43% (16)
From 6 cells	35% (17)	41% (15)
From 7 cells	13% (6)	16% (6)
(c) Who performs the biopsy of the embryo for PGD?		
Embryologist/biologist	100% (48)	100% (37)
Laboratory technician	25% (12)	22% (8)
(d) On which day do you usually transfer embryos following PGD?		
Day 3	2% (1)	—
Day 4	42% (20)	35% (13)
Day 5	52% (25)	59% (22)
Not specified	4% (2)	5% (2)
2. Do you perform positive/ negative controls?	81% (43)	84% (31)
3. Do you have dedicated rooms for?	*Monogenic laboratories (n = 37)*	*(n = 28)*
Pre-pre-PCR	78% (29)	82% (23)
Pre-PCR	65% (24)	75% (21)
Post-PCR	70% (26)	75% (21)

Complied replies from a series of technical questions designed to determine current practice quality assurance. The number of answers in each category appears in brackets. The Top 7 countries, performing over 150 cycles in 2005, are those indicated in bold in Table 1.
[a]The sum of answers is not necessarily equal to the sum of laboratories, as multiple answers were possible and not all laboratories answered all questions.
[b]Polar bodies were tested only in Austria, Germany and Switzerland.

and identifying potential contaminations were used by 43/53 laboratories (81%), similar to the 76% of US IVF–PGD clinics.[8]

Counselling, informed consent, reporting and follow-up

PGD providers should ensure that their patients receive suitable pre- and post-analytical genetic counselling and that appropriate informed consent is obtained. Fifty of 53 centres (94%) responded that they do provide genetic counselling, which may be at IVF clinics (25/50), medical genetics services (38/50) and/or 'from partners' (8/50). Similarly, 50/53 centres reported that they require informed consent. However, the expert interviews revealed that the relationships between IVF clinics and genetics services may not always be completely transparent, and concerns were raised about whether patients really did receive adequate counselling and whose responsibility this was.

Formal laboratory reports were issued by 50/53 (94%) laboratories, compliant with PGDIS (2004) and ESHRE guidelines (2005). In all laboratories, reports were signed by a clinical scientist (24/50), an MD (7/50) or both (17/50). Of the three centres not producing formal reports, two were small providers (1–10 cycles) who also kept data for only 9–12 months, whereas the majority of the centres kept data for >2 years. The third centre probably performed only PGS, not PGD.

Confirmation of PGD by prenatal diagnosis, which protects against misdiagnosis but presents a small risk to the pregnancy, is 'recommended' or 'suggested' by 44/50 (88%) centres, slightly below the 96% of US IVF–PGD clinics that recommend or require confirmation.

Monitoring and follow-up Monitoring the accuracy of PGD requires follow-up at the very least to the neonatal period and ideally on a longer term, but the survey revealed disappointingly low levels of participation. Only 48/53 (91%) laboratories follow up during pregnancy and a lower proportion follow up to the neonatal period (Table 6). Fifty-two out of 53 centres (98%) keep data on success rate but only 43/53 (81%) do so on accuracy, comparable with the United States.[8]

Table 6 Duration of follow-up of patients after PGD

	n = *53*
During pregnancy	48 (91%)
Neonatal	41 (77%)
Short-term paediatric	22 (42%)
Long-term paediatric	11 (21%)

Providers were asked 'Your laboratory follows up…' and could provide multiple answers to the above categories.

Systematic post-natal follow-up for PGD was generally found to be very limited; it was best in Belgium but weakest in the Czech Republic and Greece where only 2/12 centres followed up beyond the neonatal period. Paediatric follow-up is limited to the larger PGD centres and the expert interviews revealed that the best monitoring is provided by centres that provide both IVF and genetic services at the same location. Lack of expertise and high cost were the main reasons reported for not providing follow-up.

These data highlight a need for more thorough and longer term follow-up and documentation of results, to improve knowledge of the security and accuracy of PGD and thus to increase patient safety. The ESHRE PGD Consortium produces a detailed annual report of PGD activity, including some aspects of quality assurance, from its members who contribute on a voluntary basis; 39/53 (74%) of the surveyed centres were participants. The Consortium hopes to extend its current neonatal follow-up and to encourage monitoring of both PGD technology and long-term follow-up with all those centres that have the infrastructure and financial means.

National and international flow

Although patients and/or samples are often referred internationally, little data were previously available about the scale and nature of international flow. According to the survey, in 2005, over 800 patients were referred internationally, equivalent to approximately one-third of all the cycles identified by the survey; 68% of PGD centres treated patients from abroad and 32% received samples (Table 7).

Table 7 Trans-border flow of main countries treating patients from foreign countries in 2005

	No. of replies (PGD centres)	*Received samples from abroad?*	*Treated patients from abroad?*	*No. of patients from abroad*
Belgium	6	1	5	127
Cyprus	1	1	1	150
Czech Republic	6	1	4	110
France	3	2	3	10[a]
Germany	3	0	2	22
Greece	6	3	3	18
Netherlands	3	0	1	2
Slovakia	1	0	1	20
Spain	8	4	6	332[a]
Turkey	3	2	3	35
United Kingdom	5	0	2	—[a]
Other[b]	8	3	5	6
Total	53	17 (32%)	36 (68%)	832

A series of questions were asked to determine the current level of movement of patients and/or samples.
[a]Replies about number of patients treated annually were incomplete or absent.
[b]Other: Finland, Portugal, Sweden and Switzerland.

Table 8 Reasons for referral from foreign countries

	Number (total = 36)
Legal reasons	24
Test availability	21
Financial reasons	14
Quality/reputation	4
Waiting lists	3
Other	3

Thirty-six out of fifty-three centres received patients and/or samples from abroad. 'Other' reasons included experience, success rates and expertise on certain diseases.

The main receiving countries are Spain, Cyprus, Belgium and the Czech Republic, with a significant flow of patients also to Greece, Germany, Turkey and Slovakia. They all treated patients from a large number of European countries, but also from the US, Lebanon and Israel. Austria, France, Germany, the Netherlands and the UK appeared to be the major referrers (although the numbers of patients referred are not available); they each sent patients and samples to three or more foreign countries in 2005. Belgium and the Czech Republic were the only countries identified that were apparently self-sufficient, referring no patients to foreign countries.

Cross-border movement of patients has been reported to be primarily a direct consequence of the regulatory differences across Europe,[20] and this was partly supported by the survey: legal reasons and test availability were cited as the main drivers for flow (Table 8).

Internationally referred patients and samples clearly represent a large part of the total PGD activity in Europe, which might lead to concerns with regard to medical advice, counselling, monitoring and follow-up. A degree of harmonization may arise through the recent EU Human Tissue and Cells Directive, which also includes PGD practices. The Directive aims to ensure that, in time, patients who travel abroad for treatment will know that they can expect minimum quality and safety standards within any country of the EU. As mentioned above, accreditation to a standard such as ISO 15189 must be considered the target to ensure minimal acceptable standards in all centres providing PGD services to European patients.

Discussion

We have surveyed practices in 53 centres offering PGD from 17 European countries. Approximately 2–3000 PGD cycles were performed in 2005; the most widely available testing was for familial chromosomal anomalies (offered by 91% of laboratories), followed by monogenic diseases and HLA typing for donor matching.

PGD for monogenic testing was available in 37 centres from 15 countries for an average of 2.5 laboratories per country; however, the distribution is unequal with 28 of the laboratories in the 7 countries performing over 150 cycles in 2005. Laboratories within a country tend not to be organized to distribute tests between themselves but to provide a very wide diversity of tests, including even very rare diseases: over 50 disorders were specifically cited as being tested and a trend towards personalized testing was underlined by laboratories indicating that they offered tests for 'more than 50 indications' or even 'custom-made analysis of any disease of known genetic cause'. Each of the 10 most common tests, which would be the best candidates for rational distribution, were offered by an average of 1.6 laboratories per country. A similar lack of rationalization has been previously observed in conventional molecular genetic testing,[21] and consequently it is unsurprising to find it repeated for PGD, particularly as the specialization required for PGD is predominantly in single-cell diagnostics rather than in clinical molecular genetics (mutations are commonly initially characterized by genetics laboratories before referral for PGD). The high proportion of private laboratories active in PGD may also contribute to the tendency to offer a full catalogue of tests.

It is apparent that PGD is not simply used as an alternative to prenatal diagnosis, but it is perceived differently by patients and providers. First, PGD testing of adult-onset disorders (Huntington disease, familial predispositions to cancer, polycystic kidney disease, etc) appears to be more widespread than is the case for prenatal diagnosis. Second, testing may be requested and performed for relatively less severe or less predictable diseases: a quarter of the centres offer PGD for CMT disease, which is not a common prenatal diagnosis. Third, non-diagnostic tests are performed that are not available as prenatal diagnosis: HLA typing to select HLA-identical donors for existing individuals with severe disease was available in one-quarter of laboratories and 11 laboratories even offered this as an isolated test, without also testing for monogenic disease. A single European service responded that they provided non-medical sex selection by PGD, which is in contrast to the United States where it appears to be more acceptable and is offered by 42% of providers.[8]

These findings suggest that PGD and embryo selection may be regarded as less of an ethical problem than prenatal diagnosis (with associated termination of pregnancy); a targeted study of the indications for testing and the experiences of families would be of significant interest.

A very high degree of international exchange was identified, equivalent to approximately one-third of the cycles identified by the survey. In 2005, over 800 patients were referred internationally for PGD, two-thirds of the centres treated patients from abroad and one-third received samples from abroad. Movement was principally due to legal reasons and because of test availability, revealing a deficiency of local access to PGD for patients in some countries. Consequently, given the high costs involved,

there is a risk that PGD may only be available to wealthier couples in many situations.

Although the data reveal that almost all PGD centres provide genetic counselling and require informed consent, concerns were raised by some experts interviewed about whether all couples did indeed receive adequate counselling. It would be valuable to study the availability and quality of genetic counselling in couples who have received or are candidates for PGD, perhaps by a retrospective survey of couples who have undergone PGD.

The survey provides the first independent data on quality assurance in European PGD services. It is important to note that the reliability of PGD in Europe is currently high; no false-negative diagnostic errors were identified by the ESHRE PGD Consortium in 2004, although three misdiagnoses were identified in the latest report.[7,22] Nonetheless, the survey reveals overall that the investment in quality management by PGD centres is at present disappointingly low, notably concerning accreditation and participation in EQA schemes. To maintain the level of quality, it is important that accreditation be actively encouraged or even made mandatory by policy change, as has been recently proposed.[19] The implementation of the EU Human Tissue and Cells Directive should also have a positive impact on quality assurance of testing services, including PGD, by enforcing the introduction of a broad range of quality management elements.

The presence of laboratory directors with a PhD has been identified as a key criterion of quality assurance, and this was the case for 75% of PGD laboratories. It would be valuable to extend these data to specialization and formal training in genetics, which are called for by the OECD Guidelines[19] but which were not surveyed because of their variable availability throughout Europe.

Although accreditation is widely recognized as the single most effective route to comprehensive quality assurance, particularly for medical laboratories, its uptake is very low: only 22% of PGD laboratories are accredited or preparing for accreditation; a small number are licensed or certified (11 and 12%, respectively). Uptake of accreditation is significantly lower in the public sector than in the private sector, which may suggest a lack of support from institutions for investment in diagnostic quality assurance. Only 70% of laboratories and 52% of clinics had designated quality managers.

EQA is the most important instrument for independently determining the precision of a test in a laboratory. The survey highlighted an important weakness in quality assurance for PGD: although 98% of centres regard EQA as very important or important, no purpose-designed EQA schemes are available. The provision of schemes for cytogenetic and molecular PGD should be considered high priority for quality assessment and improvement, and a first scheme for cytogenetic PGD as well as molecular diagnosis is currently under development as a collaboration between UKNEQAS and the ESHRE PGD Consortium.

The replies concerning details of quality assurance procedures reveal a wide variation in approaches to PGD; the modal answers indicate that in the 'average' laboratory, a single blastomere is sampled by an embryologist/biologist from a 5+ cell embryo on day 3; after testing, selected embryos are transferred 2 days later. Molecular analysis for monogenic disorders is performed in facilities with dedicated pre- and/or post-PCR rooms, both by direct and indirect mutation analysis of both samples and positive and/or negative controls.

Some causes for concern were identified of different degrees of severity. Five laboratories performing monogenic PGD had no dedicated pre- nor post-PCR rooms, and one laboratory replied that it neither had any pre- nor post-PCR facilities nor used positive/negative controls and yet still performs PGD for at least eight monogenic diseases. Despite the ESHRE guidelines stating that the use of controls is 'contentious' and merely 'acceptable', this is clearly standard practice and is to be encouraged. Ten laboratories (28%) perform only direct analysis during monogenic PGD, an approach associated with a significant risk of error.

Only 69% of laboratories confirmed anomalies (chromosomal or monogenic) 'in all cases' before undertaking PGD, and one laboratory 'always believes external results', which leaves a risk of not testing for the correct anomaly. This indicates that some laboratories are doing insufficient preliminary work and this identifies an element that should be addressed by follow-up study and quality guidelines.

Similarly unacceptable was the attitude of one laboratory that EQA is irrelevant; quality assurance and improvement is fundamental to patient safety and thus should be of primary importance in medical laboratories. The discovery that 22% of the directors of responding IVF laboratories had neither a PhD nor an MD degree is unexpected, but this may be subject to the precise organization of the laboratory management.

The combined results suggest that there is a lack of knowledge of the value and procedures of laboratory quality assurance. This could be addressed usefully by the provision of specialized initial and continuous education programmes for PGD providers. It is certain that an increased investment in quality assurance will be required to maintain and improve standards and thus ensure patient safety while also increasing public and regulator confidence in PGD providers. The ESHRE PGD Consortium has created an accreditation task force (chaired by KS) to investigate requirements in this area.

Finally, it is evident that systematic long-term follow-up of all children born after PGD is necessary, with the aim of assuring quality of service and patient safety. Such a multicentre, international, longitudinal study should be

considered a priority and the coordination would ideally be assured by an existing specialized group such as the ESHRE PGD Consortium.

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