# Artificial insemination with donor sperm (AID): heterogeneity in sperm banking facilities in a single country (Belgium)

A. THIJSSEN<sup>1,2,\*</sup>, N. DHONT<sup>1</sup>, E. VANDORMAEL<sup>1</sup>, A. COX<sup>1</sup>, E. KLERKX<sup>1</sup>, E. CREEMERS<sup>1</sup>, W. OMBELET<sup>1,2</sup>

<sup>1</sup>Genk Institute for Fertility Technology, Department of Obstetrics and Gynaecology, Ziekenhuis Oost-Limburg, Schiepse Bos 6, 3600 Genk, Belgium; <sup>2</sup>Faculty of Medicine and Life Sciences, Hasselt University, Martelarenlaan 42, 3500 Hasselt, Belgium.

Correspondence at: annelies.thijssen@uhasselt.be

#### Abstract

Due to the high inflow of foreign patients seeking cross-border reproductive care in Belgium and the increased number of lesbian couples and single women who call for artificial insemination with donor sperm (AID), Belgian sperm banks nowadays face a shortage in donor sperm. However, since there is no central registration system for sperm donors in Belgium, no figures are currently available supporting this statement. Therefore a study was performed to obtain a detailed overview of the sperm banking facilities in Belgium. Questionnaires were sent to all Belgian centres for assisted reproduction with laboratory facilities (n = 18) to report on their sperm banking methods. The results showed that 82% of the centres rely partially or completely on foreign donor sperm. Moreover, four of the thirteen centres that have their own sperm bank use imported donor sperm in > 95% AID cycles. Our results show that in 63% of the Belgian AID cycles imported Danish donor sperm is being used.

Donor recruitment is mainly performed through the centre's website (61%) or by distributing flyers in the centre (46%) and 9 to 180 potential donors have been recruited per centre in 2013. Eventually, 15 to 50% of these candidate donors were accepted. Different criteria for donor acceptance are handled by the centres: donor age limits range from 18-25 to 36-46 years old, and thresholds for sperm normality differ considerably. We can conclude that a wide variation in methods associated with sperm banking is observed in Belgian centres.

Key words: Artificial insemination, donor sperm, Belgium, questionnaire, sperm banking.

#### Introduction

According to the Belgian Register for Assisted Procreation (BELRAP), the number of initiated artificial insemination cycles with donor sperm (AID) in Belgium has increased from 8766 AID cycles in 2008-2009 (BELRAP, 2013) to 13048 in 2010-2011 (BELRAP, 2014). Although this increase of AID cycles may partially be attributed to additional centres reporting to the BELRAP in recent years, Belgian centres also reported a substantial increase in the number of patients relying on sperm donation (personal communications).

Where in the past the use of donor sperm was restricted to hetero couples with an azoospermic husband or when the male partner carried an inheritable genetic disease, nowadays more lesbian couples and single women call for donor insemination (BELRAP, 2009; 2013; 2014).

Secondly, Belgian centres for Reproductive Medicine are overwhelmed with patients seeking cross-border reproductive care (CBRC) (BELRAP, 2009; 2013; 2014; Pennings et al., 2009), trying to avoid restrictive laws in their home country (Fig. 1) (Pennings, 2004; Ferraretti et al., 2013). For example, in the Netherlands and the United Kingdom, anonymous sperm donation is abolished since 2004 and 2005 respectively (Staatsblad, 2002; HFEA, 2004). Furthermore, the treatment of lesbian couples and single women with donor sperm is restricted in France and Germany (Journal officiel de la République Française, 1994; Berlin Sperm Bank, 2011). On the contrary, Belgian legislation allows for anonymous as well as non-anonymous



*Figure 1:* Rise in the percentage of patients with no male partner (lesbian couples and single women) and no social security (foreign patients), applying for AID in Belgium.

AID: Artificial Insemination with Donor sperm. No social security indicates the percentage of patients treated for AID in Belgium that are not connected to a Belgian social security fund, mostly indicating foreign patients.

(Based on results adapted from reports of the Belgian Register for Assisted Procreation (BELRAP, 2009; 2013; 2014)).

sperm donation to be performed on hetero and lesbian couples as well as single women (Fig. 2) (Belgisch Staatsblad, 2007). However, in contrast to countries like Spain and Denmark where the government supports men in becoming a sperm donor, the Belgian government does not allow any form of advertisement for the recruitment of sperm donors (Belgisch Staatsblad, 2008). Moreover, according to the Belgian legislation only six women can become pregnant from a single sperm donor (Belgisch Staatsblad, 2007).

Nowadays, Belgian sperm banks face a shortage in donor sperm (Belga, 2014). However, since there is no central registration system for sperm donors in Belgium, there are currently no figures supporting this statement. In line of the setup of our own sperm bank, we performed a study to obtain a detailed overview of the sperm banking facilities in Belgium. To our knowledge no such study has been performed in Belgium or in any other country before. Questionnaires (see Addendum) were sent to all Belgian centres for assisted reproduction with laboratory facilities for intra-uterine insemination (IUI) and in vitro fertilisation (IVF) (B-centres) to report on their sperm banking methods, with special attention to the methods and criteria used for recruitment, screening and selection of potential sperm donors, procedures for sperm washing and freezing and costs associated with sperm donation.



*Figure 2:* Legislation in Belgium and neighbouring countries Legislation allows for hetero couples as well as lesbian couples and single women to be treated with donor insemination + sperm donation may be anonymous as well as nonanonymous, Legislation only allows hetero couples to be treated with donor insemination, Legislation only allows non-anonymous sperm donation. BE: Belgium; DE: Germany; FR: France; NL: Netherlands; UK: United Kingdom.

#### **Materials and Methods**

Questionnaires involved a mix of open and closed questions (i.e. multiple choice), with the possibility of making additional remarks (see addendum).

At a first stage, a short questionnaire was sent by e-mail to all Belgian centres for assisted reproduction with laboratory facilities for IUI and IVF, the socalled B-centres (n = 18). Only B-centres are allowed to have a sperm donor bank. This short questionnaire was designed to obtain basic information on the use of AID in Belgium; i.e. the number of AID cycles performed by the centre, the origin of the donor sperm (Belgian or imported donor sperm), to what ratio the centres use Belgian versus imported donor sperm, for which patients (hetero/lesbian couples or single women) donor insemination is performed, whether the centre performs anonymous or nonanonymous sperm donation and, in case the centre has its own sperm bank, methods for the recruitment of potential donors. The short questionnaire was distributed in English to all Belgian centres.



*Figure 3*: Number of AID cycles per year. Overview of het total number of AID cycles performed by the centres each year.

Based on the results of the first questionnaire, the centres having their own sperm bank were selected (n = 13). A more detailed questionnaire was sent by e-mail to these centres to collect more information on their procedures for sperm banking. Additional questions included specific donor recruitment strategies and numbers, screening and selection criteria for candidate donors, the freezing and washing procedures for donor semen and the payment of donors. The detailed questionnaires were sent in Dutch to the Flemish speaking centres and in French to the French speaking centres and they were distributed between December 2013 and April 2014.

All data were stored and analysed in Excel spreadsheets (Microsoft Excel 2010).

#### Results

# *Response rate and geographical distribution of the participating centres*

Belgium counts eighteen B-centres for Reproductive Medicine (n = 18). For both questionnaires, a response rate of 100% was accomplished. The different centres for Reproductive Medicine are distributed between the three Belgian regions as follows: eight centres in Flanders, four centres in Brussels and six centres in Wallonia.

## Size of the participating centres

The results from the first questionnaire showed that all but one (17/18 or 94,4%) of the B-centres for Reproductive Medicine perform AID. The size of the centres can be determined by the number of AID



*Figure 4:* The percentage of centres treating hetero and lesbian couples and (no) single women.



*Figure 5:* The percentage of centres performing only anonymous or anonymous + non-anonymous donor inseminations.

cycles they perform each year. These are clearly shown in Figure 3.

#### Patients treated with AID

All three patient groups, i.e. hetero and lesbian couples and single women, can apply for AID in thirteen out of the seventeen (76%) centres. The remaining centres (n = 4) do not offer AID to single women and only perform AID for hetero and lesbian couples (Fig. 4).

#### Anonymous or non-anonymous donor insemination

About half of the centres (8/17 or 47%) always perform anonymous donor inseminations. The other centres (9/17 or 53%) are open to both anonymous and non-anonymous sperm donation, although they often state that the request for non-anonymous donor insemination is extremely rare (Fig. 5).

59



Figure 6: Origin and use of donor sperm

# The origin and use of donor sperm

Only two out of the seventeen centres (12%) rely completely on their own sperm bank. The other fifteen centres (15/17 or 88%) rely partially (11/17 or 64%) or entirely (4/17 or 24%) on the import of foreign donor sperm (Fig. 6). Of the 11 centres using imported donor sperm as well as Belgian donor sperm from their own sperm bank, three centres use Belgian donor sperm in > 95% of the AID cycles they perform, in four centres the ratio for Belgian and imported donor sperm used for AID is fifty/fifty and another four centres use imported donor sperm in  $\ge$  95% of their AID cycles (Fig. 6).

Imported donor sperm in Belgium is exclusively obtained from Denmark, i.e. Nordic Cryobank (Nordic Cryobank ApS, Frederiksberg, Denmark) or Cryos International sperm bank (Cryos International ApS, Aarhus, Denmark), and none of the centres imports donor sperm from other Belgian sperm banks. Our results indicate that 63% of the donor inseminations performed in Belgium are accomplished with imported donor sperm from Denmark.

# Donor recruitment

Donor recruitment is mostly done by providing information on donor insemination and the possibility of becoming a sperm donor on the centres website (8/13 or 61%) or by distributing flyers in the centre (6/13 or 46%). Other ways to recruit new potential donors are oral advertisement (3/13 or 23%) or asking partners of women who became pregnant after infertility treatment and with no male factor infertility involved (normal semen sample) to become a donor (solidarity principle) (4/13 or 31%). Most centres use a combination of different donor recruitment strategies.

In 2013 the total number of candidate donors applying to the Belgian centres for a first semen analysis was 602, with one centre reporting 9 candidate donors and another centre reporting 180 candidate donors. The average acceptance rate of the Belgian candidate donors is 32% (range 15% to 50%). In 2013 a total number of about 260 candidate donors were accepted in Belgium (Table I).

To make sure a donor will only donate in one specific centre, five out of the thirteen sperm banks (38%) make use of a contract with the donor. This implicates that eight sperm banks (62%) do not make use of such a written contract.

# Criteria for sperm donors

The minimum age for a candidate sperm donor was reported at 18 years for 11 out of the 13 sperm banks (85%). The other two centres reported a minimum age of 21 and 25 years old respectively. The maximum donor age differs substantially between the centres, with a minimum of 36 and a maximum of 46 years old (Fig. 7, Table I)).

All sperm banks perform the statutory infection screenings: hepatitis B and C, human immunodeficiency virus (HIV)1,2, syphilis and chlamydia. Furthermore, all centres but one (12/13 or 92%) also screen for cytomegalovirus (CMV) infections. Of the centres who screen for CMV infection, the majority (9/12 or 75%) accepts CMV+ donors, with 8 of them also matching CMV+ donors



Figure 7: Maximum donor age limits used by the centres

with CMV+ recipients. In addition, one centre also reported to screen for Neisseria gonorrhoea. Next to the infection screening, all centres also perform a genetic screening for cystic fibrosis (CF) and determine the karyotype of the candidate sperm donor. Additionally, one centre reported to test for Y chromosome microdeletions and two other centres reported to test for hemoglobinopathy. A psychological screening of the candidate donors is only performed in three out of the 13 Belgian sperm banks (23%).

The minimum criteria for sperm quality parameters differ substantially between the different sperm banks. The lower limit for native sperm concentration ranges from 15 up to 60 million spermatozoa per ml. Additionally, the threshold for good forward progressive motility has to be at least 30% in one centre and up to 70% in another centre. Minimum criteria for sperm morphology range from 4% to 25% morphologically normal spermatozoa. Minimum criteria for inseminating motile counts (IMC) after sperm freezing and thawing, range from 1 up to 5 million (Table I).

Before donor acceptance, the majority of the sperm banks (10/13 or 77%) also perform a test procedure for sperm washing. Two centres perform the washing procedure prior to sperm freezing, seven centres after sperm freezing and one centre performs a washing procedure of the sperm sample both before and after sperm freezing.

# Handling of donor sperm

The majority of the sperm banks (9/13 or 69%) always freeze donor semen in its fresh state. One centre (1/13 or 8%) indicated to perform both freezing of fresh and concentrated semen and another centre always concentrates the semen sample prior to freezing. The remaining two centres freeze the sperm sample after capacitation. In addition, all centres reported to make use of straws as a carrier material for sperm freezing (Table I).

The majority of the sperm banks (11/13 or 85%) store semen samples from donors that were not yet serologically tested in a separate quarantine container. Of the centres that accept CMV+ sperm donors, only one (1/9 or 11%) stores these samples separately.

# Donor payment

A Belgian donor is paid  $\notin$  66 per donation on average, with a minimum of  $\notin$  50 and a maximum of  $\notin$  100 per donation (Table I).

# Discussion

Belgium is a small country, with only 13 sperm banking facilities. It is well known that country regulations concerning all different aspects of gamete donation vary a lot because of political, ethical, socio-cultural and religious differences. The results we obtained only describe the Belgian situation.

Belgian sperm banks nowadays face a shortage in donor sperm. Therefore, this study aimed to present an overview of the sperm banking facilities in Belgium with special attention to the methods and criteria used for recruitment, screening and selection of potential sperm donors, procedures for sperm washing and freezing and costs associated with sperm donation.

Since three quarter of the Belgian centres offer donor insemination to lesbian couples and single women as well as hetero couples, and anonymous donor insemination is available in all Belgian centres, foreign patients frequently come to Belgium seeking CBRC. According to Pennings et al. (2009), most of these patients are coming from France (38%), the Netherlands (29%), Italy (12%) and Germany (10%). Patients coming from the UK constitute only 2% of all foreign patients treated in Belgium, since they are more likely to go to other European countries like Spain (Tremlett, 2006). More specifically for sperm donation, Pennings et al. (2009) reported that 80% of the treatment cycles were performed for French women, 13% for Dutch women and 3% for Italian women. Since 2004, there has been a steady increase in the number of French patients coming for donor insemination (Pennings et al., 2009; Rozee Gomez & de La Rochebrochard, 2013). For Dutch patients, the number for donor insemination almost doubled between 2004 and 2005, following the abolishment of donor anonymity in the Netherlands (Pennings et al., 2009; Staatsblad, 2002).

Our results showed that in 2013 about 63% of the total number of donor inseminations performed in

61

Belgium were accomplished with imported donor sperm from Denmark. This figure seems to be alarming, but one has to keep in mind that in the period 2010-2011 63.3 % of AIDs in Belgium were performed in foreign patients (8262/13048 AID cycles) (BELRAP, 2014).

Out of the 13 centres having their own sperm bank, 4 centres still use imported donor sperm in over 95% of the AID cycles. Some of them stated they are keeping their own sperm bank for 'times of need', in case the Belgian government would decide to prohibit the import of foreign donor sperm.

In addition to the shortage in donor sperm, Belgian legislation only allows pregnancies with sperm from a single donor to a maximum of six different women (Belgisch Staatsblad, 2007). Recently, a comment was added to this law, stating that a lesbian couple applying for donor insemination accounts for one women (Belgisch Staatsblad, 2014). The maximum of six women is rather low in comparison to most of our neighbouring countries, allowing a maximum of 25 children per sperm donor in the Netherlands, 15 children in Germany and a maximum of 10 families in the United Kingdom (Fig. 2). Only France has, with a maximum of 5 children, a lower rate for the number of children that are allowed to be born from a single donor. Because there is no central registration system for sperm donors in Belgium, it is questionable if this law is actually implemented in practice. Some centres ask their sperm donors to donate sperm exclusively in their centre by signing a contract in order to avoid that the same donor is going to different centres.

Legislation in Belgium also makes it difficult to recruit sufficient sperm donors, mainly because any form of public advertisement for the recruitment of sperm donors is prohibited by law (Belgisch Staatsblad, 2008). Therefore, the centres mainly use their website or the distribution flyers in their centre to inform potential donors on the possibility of sperm donation. According to many AID-centres, a rise in the number of sperm donor applicants is observed whenever sperm donation is mentioned in the media, although this has never been properly investigated and/or documented. If so, this would be a good argument for asking the government to start with awareness campaigns. The National Gamete Donation Trust (NGDT) in the United Kingdom is a very good example of how to organise recruitment for gamete donation. The NGDT is the national body running the National Gamete Donation Services. They work with potential recipients, UK licensed fertility clinics, the media and support organisations to raise awareness of the need for gamete donors. They realised to have an

increase of sperm donors, although anonymous donation is forbidden in the UK (National Gamete Donation Trust, http://www.ngdt.co.uk).

Belgian donors are paid between  $\in 50$  and  $\in 100$  per donation. This payment is only meant to cover the transportation costs or loss of salary for the donor when he comes to donate his sperm (Belgisch Staatsblad, 2007). The reason why some centres pay much more than other centres cannot be found.

Apart from the low number of candidate donors applying to the centres, the average acceptance rate for sperm donors in Belgium is also rather low. On average between 15 and 50% of the sperm donor applicants meet all criteria to become a sperm donor. We couldn't find a good reason why the acceptance rate differed so much between centres.

The threshold values for sperm quality differ strongly between centres. Different cut-off values for normality are used mostly due to different methodology for sperm analysis and different criteria used, especially for sperm morphology (Ombelet et al., 1997). We observed that in all centres the criteria for defining normal sperm quality are substantially higher than the criteria posed by the World Health Organization indicating a normal semen sample (WHO, 2010). However, the sometimes higher threshold for sperm normality of a certain centre does not always correspond to a lower acceptance rate in that specific centre (Table I). On the other hand, higher values of sperm parameters do not necessarily result in better pregnancy rates (Hu et al., 2012; Ombelet et al., 2014).

The Belgian law implies that a sperm donor tests negatively for hepatitis B and C, HIV1,2, syphilis and chlamydia. In addition, all but one of the Belgian centres also screen for CMV infection. With CMV being the leading cause of congenital viral infection, potentially leading to foetal death, severe birth disease or subsequent development of neurological or sensory impairment, it is indeed an important parameter to take into account (Pass et al., 1980; Stagno et al., 1977). Different strategies for handling CMV+ sperm donors have been proposed by different agencies. According to the American Society for Reproductive Medicine (ASRM) guidelines, gametes from CMV seropositive donors should only be used for seropositive recipients (American Society for Reproductive Medicine, 2013). On the other hand, the British Andrology Society (BAS) recommends that only CMV seronegative men should be allowed to donate sperm (British Andrology Society, 1999). This recommendation would however lead to an even greater shortage in sperm donors since about 50-95% of all people at reproductive age are CMV

53         20         21         20         100         80         10         19         180           17         33         50         30         15         50         20         46         NA         N           21         18         18         25         18         18         18         18         18         18           21         18         18         25         18 <th>9 NA</th> <th></th> <th></th> <th></th> <th>-</th> <th>_</th> <th></th> <th></th> <th></th> <th>;</th>	9 NA				-	_				;
17     33     50     30     15     50     20     46     NA     N       21     18     18     25     18     18     18     18     18     18       44     46     43     45     40     44     45     45     45       15     40     50     60     15     40     30     20     20     30       32     50     50     60     15     40     30     20     30     30       32     50     50     60     15     40     30     20     30     30       32     50     50     60     15     40     30     20     30     30       32     50     50     60     42     50     70     40     30     30       18     10     10     4     10     4     10     4     10       1     2     2     1     1     1     1     NA       1     1     1     1     1     1     10       1     1     1     1     1     1     1       1     1     1     1     1     1       1	NA	180	19	10	80	100	20	21	20	53
21       18       18       25       18       19       19       10       10       10       10       10       10       10       10 <td< td=""><td></td><td>NA</td><td>46</td><td>20</td><td>50</td><td>15</td><td>30</td><td>50</td><td>33</td><td>17</td></td<>		NA	46	20	50	15	30	50	33	17
21         18         18         25         18         10         10         10<										
44     46     43     45     40     44     45     45     45     45     45       15     40     50     60     15     40     30     20     20     30       32     50     50     60     15     40     30     20     20     30       44     10     10     4     10     4     10     4     10       2     2     1     1     1     1     1     NA       2     2     1,5     5     1     1     1     NA       3     5     5     5     5     5     5     5       3     5     5     5     5     5     5     5       4     10     4     10     4     10     1       5     5     5     5     5     5     5       5     5     5     5     5     5     5       5	18	18	18	18	18	18	25	18	18	21
15       40       50       60       15       40       30       20 <td< td=""><td>40</td><td>45</td><td>45</td><td>45</td><td>44</td><td>40</td><td>45</td><td>43</td><td>46</td><td>44</td></td<>	40	45	45	45	44	40	45	43	46	44
15       40       50       60       15       40       30       20       20       20       30         32       50       50       60       42       50       70       40       30 <t< td=""><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></t<>										
15       40       50       60       15       40       30       20       20       20       30 <td< td=""><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></td<>										
32         50         50         60         42         50         70         40         30         33           4         10         10         10         10         4         4         10         30         33           2         2         2         1,5         5         1         1         4         10         4         10         30         33           2         2         2         1,5         5         1         1         1         NA         10         4         10         10         1         1         1         1         1         1         NA         10         1         <	20	20	20	30	40	15	60	50	40	15
441010410410410221,55111NA111,55111NA111,55111NA1111111NA11<	50	30	40	70	50	42	60	50	50	32
2     2     2     1,5     5     1     1     1     NA       fresh     fresh     fresh     fresh     fresh     fresh     fresh     fresh     fresh       straws     straws     straws     straws     straws     straws     straws     straws	4	10	4	10	4	4	10	10	4	4
2     2     2     1,5     5     1     1     1     NA       ifesh     fresh     fresh     fresh     fresh     fresh     fresh     fresh     ca       straws     straws     straws     straws     straws     straws     straws     straws     straws										
freshfreshconcfreshfreshfreshfreshfreshcastrawsstrawsstrawsstrawsstrawsstrawsstrawsstrawsstraws	5	NA	1	1	1	5	1,5	2	2	2
freshfreshfreshfreshfreshfreshcastrawsstrawsstrawsstrawsstrawsstrawsstrawsstraws										
straws	capac	fresh	fresh	fresh	fresh	fresh	conc	fresh	fresh	fresh
	straws	straws	straws	straws	straws	straws	straws	straws	straws	straws
60         50         50         85         75         50         75         50         7	70	50	75	50	75	85	50	50	50	60
20		50 5 5 capac capac straws 70	30         50           10         4           NA         5           fresh         capac           straws         straws           50         70           able).         50	40         30         50           4         10         4           1         NA         5           fresh         fresh         capac           straws         straws         straws           75         50         70           A = not available).         70	70         40         30         50           10         4         10         4           1         1         NA         5           fresh         fresh         fresh         capac           straws         straws         straws         straws           50         75         50         70           cedure (NA = not available).         70         70	50         70         40         30         50           4         10         4         10         4           1         1         1         1         4           1         1         1         1         4           1         1         1         1         4           1         1         1         1         4           1         1         1         1         5           fresh         fresh         fresh         fresh         capac           straws         straws         straws         straws         straws           75         50         75         50         70           ashing procedure (NA = not available).         70         70         70	42     50     70     40     30     50       4     4     10     4     10     4       5     1     1     1     NA     5       fresh     fresh     fresh     fresh     fresh     capac       straws     straws     straws     straws     straws     straws       85     75     50     75     50     70	60         42         50         70         40         30         50           10         4         4         10         4         10         4           1,5         5         1         1         1         NA         5           1,5         5         1         1         1         NA         5           conc         fresh         fresh         fresh         fresh         fresh         straws           straws         straws         straws         straws         straws         straws         straws           50         85         75         50         75         50         70	50         60         42         50         70         40         30         50           10         10         4         4         10         4         10         4           2         1,5         5         1         1         1         NA         5           fresh         conc         fresh         fresh         fresh         fresh         fresh         straws           straws         straws         straws         straws         straws         straws         straws         straws           50         50         75         50         75         50         70         70	50 $50$ $60$ $42$ $50$ $70$ $40$ $30$ $50$ 4         10         10         4         4         10         4         10         4           2         2         1,5         5         1         1         1         NA         5           fresh         fresh         fresh         fresh         fresh         fresh         fresh         straws           straws

positive (Perol, 1994; Bresson et al., 2003). Although we found that most Belgian sperm banks screen potential donors for CMV infection and either reject or accept the CMV+ donor if he can be matched to a CMV+ acceptor, one centre does not match their CMV+ samples and another centre doesn't screen for CMV at all. The Belgian law is not clear on whether a candidate sperm donor should even be tested for CMV since it only states that: "In certain situations, further tests are needed, depending on the history of the donor and the characteristics of the donated human body material (e.g. CMV)." (Belgisch Staatsblad, 2009). Literature makes it even more confusing, because there is no consensus on whether CMV is only present in the seminal plasma (Lupton et al., 2014) and thus could be eliminated by sperm washing or also in the spermatozoa themselves (Naumenko et al., 2011). Additionally, research by Doerr et al. (1987) showed that the sero-immunoglobulin status (i.e. IgG or IgM positive) is not indicative for whether or not CMV is shed in the semen. Other researchers therefore recommend to test each sperm sample for presence of CMV via PCR, rather than testing the donor (Kaspersen et al., 2012). Furthermore, CMV seropositive women are only partially protected against a new CMV infection (Boppana et al., 2001; Fowler et al., 2003).

According to the results of our study we can conclude that a wide variation in methods associated with sperm banking is observed in Belgian centres. Donor recruitment strategies, screening of donors, acceptance rates, thresholds for acceptable sperm quality, sperm preparation techniques, freezing methods and even the financial reimbursement per sperm sample differ substantially between the centres. Furthermore, we have shown that there is indeed a shortage of non-imported donor sperm by demonstrating that about two-third of the donor inseminations in Belgium are performed with Danish donor sperm. In order to increase the number of candidate donors, the Belgian government should be more supportive, for example by allowing advertisement for sperm donor recruitment or by organizing awareness campaigns themselves as they do in the United Kingdom (National Gamete Donation Trust).

### Acknowledgements

We would like to thank all the B-centres for Reproductive Medicine in Belgium for their co-operation in this study: Centre de Fécondation In Vitro, C.H. Interregional Edith Cavell (CHIREC), **BRAINE L'ALLEUD**; Centre Liégeois pour l'Etude et le Traitement de la Stérilité, Clinique Saint Vincent, **ROCOURT**; Centre de FIV, Centre Hospitalier Regional de la Citadelle, LIEGE; Centrum voor Reproductieve Geneeskunde, Universitair Ziekenhuis Brussel, BRUSSEL; Clinique de Procréation Médicalement Assistée, Hôpital Universitaire Saint-Pierre - U.L.B., BRUXELLES; Centrum voor Reproductieve Geneeskunde, ZNA Middelheim, ANTWERPEN; Leuvens Universitair Fertiliteitscentrum, K.U.Leuven Gasthuisberg, LEUVEN; Vrouwenkliniek - afdeling reproductieve geneeskunde, Universitair Ziekenhuis Gent, GENT; CRG Brugge-Kortrijk, Algemeen Ziekenhuis Sint-Jan, BRUGGE; Centrum voor Reproductieve Geneeskunde, Universitair Ziekenhuis Antwerpen, EDEGEM; Unit Reproductieve Geneeskunde, Regionaal Ziekenhuis Heilig Hart, LEUVEN; Service de Gynécologie, Cliniques Universitaires Saint-Luc -U.C.L., BRUXELLES; Centre de FIV de l'ULB, Hôpital Erasme, BRUXELLES; Centrum voor Fertiliteitstherapie, Algemeen Ziekenhuis Jan Palfijn, GENT; Service Gyn/Obst, GHdC, Clinique Notre Dame, CHARLEROI; Centre d'Infertilité, Centre Hospitalier de l'Ardenne, LIBRAMONT; Service PMA, Centre Hospitalier Régional de Namur, NAMUR, Genk Institute for Fertility Technology, Ziekenhuis Oost-Limburg -St. Jan, GENK.

This study is part of the Limburg Clinical Research Program (LCRP) UHasselt-ZOL-Jessa, supported by the foundation Limburg Sterk Merk, Hasselt University, Ziekenhuis Oost-Limburg and Jessa Hospital.

#### References

- American Society for Reproductive Medicine. Recommendations for gamete and embryo donation: a committee opinion. Fertil Steril. 2013;99:47-62.
- Belga. Nog steeds tekort aan spermadonoren in België. De Standaard [Online]. 22 January 2014. Available: http://www.standaard.be/cnt/dmf20140122\_00942263
- Belgisch Staatsblad. Wet betreffende de medisch begeleide voortplanting en de bestemming van de overtallige embryo's en de gameten. 6 juli 2007.
- Belgisch Staatsblad. Wet inzake het verkrijgen en het gebruik van menselijk lichaamsmateriaal met het oog op de geneeskundige toepassing op de mens of het wetenschappelijk onderzoek. 19 december 2008.
- Belgisch Staatsblad. Koninklijk besluit tot vaststelling van de kwaliteits- en veiligheidsnormen voor het doneren, wegnemen, verkrijgen, testen, bewerken, bewaren en distribueren van menselijk lichaamsmateriaal, waaraan de banken voor menselijk lichaamsmateriaal, de intermediaire structuren voor menselijk lichaamsmateriaal en de productieinstellingen moeten voldoen. 28 september 2009.
- Belgisch Staatsblad. Wet houdende diverse bepalingen inzake gezondheid. 10 april 2014.
- BELRAP, 2009. Report 2007 of the College of Physicians in Reproductive Medicine - Non-IVF [Online]. http://www. belrap.be/Public/Reports.aspx
- BELRAP, 2013. Report 2008-2009 of the College of Physicians in Reproductive Medicine - Non-IVF [Online]. http://www. belrap.be/Public/Reports.aspx
- BELRAP, 2014. Report 2010-2011 of the College of Physicians in Reproductive Medicine - Non-IVF (revised on 15-01-2014) http://www.belrap.be/Public/Reports.aspx
- Berlin Sperm Bank, 2011. Information for Donors: legal aspects [Online]. http://www.berlin-spermbank.com/donors/legalaspects.html

- Boppana SB, Rivera LB, Fowler KB et al. Intrauterine transmission of cytomegalovirus to infants of women with preconceptional immunity. N Engl J Med. 2001;344:1366-71.
- Bresson JL, Clavequin MC, Mazeron MC et al. Risk of cytomegalovirus transmission by cryopreserved semen: a study of 635 semen samples from 231 donors. Hum Reprod. 2003;18:1881-6.
- British Andrology Society. British Andrology Society guidelines for the screening of semen donors for donor insemination (1999). Hum Reprod. 1999;14:1823-6.
- Doerr HW, Rentschler M, Scheifler G. Serologic detection of active infections with human herpes viruses (CMV, EBV, HSV, VZV): diagnostic potential of IgA class and IgG subclass-specific antibodies. Infection. 1987;15:93-8.
- Ferraretti AP, Goossens V, Kupka M et al. Assisted reproductive technology in Europe, 2009: results generated from European registers by ESHRE. Hum Reprod. 2013;28:2318-31.
- Fowler KB, Stagno S, Pass RF. Maternal immunity and prevention of congenital cytomegalovirus infection. JAMA. 2003;289:1008-11.
- HFEA. The Human Fertilisation and Embryology Authority (Disclosure of Donor Information) Regulations 2004. 14 June 2004.
- Hu L, Liao AH, Song S et al. Evaluation of donor semen quality provided by six sperm banks: a retrospective study of 1877 artificial insemination cycles. Andrologia. 2012;44 Suppl 1:499-504.
- Journal officiel de la République française. LOI no 94-654 du 29 juillet 1994 relative au don et à l'utilisation des éléments et produits du corps humain, à l'assistance médicale à la procréation et au diagnostic prénatal (1). 29 juillet 1994.
- Kaspersen MD, Larsen PB, Kofod-Olsen E et al. Human herpesvirus-6A/B binds to spermatozoa acrosome and is the most prevalent herpesvirus in semen from sperm donors. PLoS One. 2012;7:e48810.

- Lupton J, Vernamonti J, McCabe C et al. Cytomegalovirus and human immunodeficiency virus in semen of homosexual men. Fertil Steril. 2014;101:350-8.
- Naumenko VA, Tyulenev YA, Yakovenko SA et al. Detection of human cytomegalovirus in motile spermatozoa and spermatogenic cells in testis organotypic culture. Herpesviridae. 2011;2:7.
- National Gamete Donation Trust, http://www.ngdt.co.uk
- Ombelet W, Pollet H, Bosmans E et al. Results of a questionnaire on sperm morphology assessment. Hum Reprod. 1997;12: 1015-20.
- Ombelet W, Dhont N, Thijssen A et al. Semen quality and prediction of IUI success in male subfertility: a systematic review. Reprod Biomed Online. 2014;28:300-9.
- Pass RF, Stagno S, Myers GJ et al. Outcome of symptomatic congenital cytomegalovirus infection: results of long-term longitudinal follow-up. Pediatrics. 1980;66:758-62.
- Pennings G. Legal harmonization and reproductive tourism in Europe. Hum Reprod. 2004;19:2689-94.
- Pennings G, Autin C, Decleer W et al. Cross-border reproductive care in Belgium. Hum Reprod. 2009;24:3108-18.
- Perol Y. Infection à cytomegalovirus et grossesse. Lett Gynécol. 1994;192:10-8.
- Rozee Gomez V, de La Rochebrochard E. Cross-border reproductive care among French patients: experiences in Greece, Spain and Belgium. Hum Reprod. 2013;28:3103-10.
- Staatsblad. Wet donorgegevens kunstmatige bevruchting. 28 mei 2002.
- Stagno S, Reynolds DW, Huang ES et al. Congenital cytomegalovirus infection. N Engl J Med. 1977;296:1254-8.
- Tremlett G. Spain becomes the destination of choice for fertility tourists from Britain. The Guardian. 2006.
- WHO. WHO Laboratory Manual for the Examination and Processing of Human Semen, 5th edition. Cambridge University Press, Cambridge, UK. 2010.

Questionnance, Municipal Inserninguon white donor spermitivally in perendin	<b>Ouestionnaire:</b> Ar	tificial insemination	with donor sperm	(AID) in Belgium
---	--------------------------	-----------------------	------------------	------------------

General information	
Name of the clinic: City	
1. Does your centre perform AID?	
2. If yes, how many cycles per year?           □ 0-200         □ 200-400         □ 400-600         □ More than 6	500 🗆 Unknown
3. Where do you obtain your donor sperm from? Multiple answers are po Own sperm bank Belgian sperm bank II in Which centre?	ossible nternational sperm bank Which centre? Cryos Nordic Other:
<ul> <li>4. In your centre, what ratio is used for AID of: Total = 100%</li> <li>Donor sperm from own centre:%</li> <li>Imported Belgian donor sperm:%</li> <li>Imported international donor sperm:%</li> </ul>	
5. Which patients can be treated with AID in your centre? Multiple answer Hetero couples Lesbian couples Singles	ers are possible
6. In your centre AID is Always anonymous Never anonymous Both anonymous a	and non-anonymous
<ul> <li>7. This question is only for centers with their own sperm bank :</li> <li>Can you indicate how you recruit the sperm donors?</li> <li>Orally (e.g. during lectures or couples coming to or previously treated If yes, describe how (for example: solidarity principle)</li> <li>Through website</li> <li>Advertisement in papers or magazines <ul> <li>If yes, where and how?</li> <li>Flyers in centre</li> <li>Other (specify):</li> </ul> </li> </ul>	d in the IVF centre)
1. Number of cycles	
<ul> <li>How many cycles with donor sperm do you perform in your centre ea</li> </ul>	ch year?
2011:  0-200 2012:  0-200	
□ more than 600 □ more than 6	00
<ul> <li>2. Donor Recruitment</li> <li>How many candidate donors apply to your centre each year for a first candidate donors a year</li> <li>How many of the recruited candidate donors are accepted?</li> <li></li></ul>	semen analysis? n your centre only?

	□ Sperm quality <i>Fresh semen:</i> Conc: ≥ m/ml Mot: ≥ % Morph: ≥ % Vol: ≥ ml <i>After thawing:</i> IMC ≥
•	Are candidate sperm donors psychologically screened before acceptance as a donor?
020	□ NO □ Tes
•	(Multiple answers are possible)         HCV       Syphilis         HBV       Chlamydia
•	If a candidate sperm donor tests positive for CMV, the candidate donor is:         Refused         Accepted, but matched with a CMV+ acceptor         Other (specify):
•	Is there a quarantine nitrogen vessel for donor samples that are not yet serologically screened?
•	If your centre accepts CMV+ sperm donors, is there a separate quarantine nitrogen vessel for CMV+ sample storage?
•	Is there a chromosomal test performed on the (candidate) sperm donor? (Multiple answers are possible) No Yes CF-screening Karyotype Other (specify):
•	Is there a test procedure for sperm washing performed on the semen sample?
	<ul> <li>□ No □ Yes</li> <li>□ Before freezing □ After freezing □ Before and after freezing</li> </ul>
4.	Freezing and washing procedures
•	How is the sperm sample frozen?
	<ul> <li>Fresh</li> <li>Washed</li> <li>Concentrated</li> <li>Gradient centrifugation</li> <li>Swim-up</li> <li>Other (specify):</li> </ul>
٠	Which carrier material is used for sperm freezing?
	□ Straws □ Vials □ Other (specify):
5.	Cost
•	Is the sperm donor reimbursed? □ No □ Yes € per donation
٠	If yes, when is the sperm donor reimbursed?
	<ul> <li>After every donation</li> <li>At the end of the donation series</li> <li>Other (specify):</li> </ul>