Summary of Safety and Clinical Performance

Density Gradient Media

This Summary of Safety and Clinical Performance (SSCP) is intended to provide public access to an updated summary of the main aspects of the safety and clinical performance of the device. The SSCP is not intended to replace the Instructions For Use (IFU) as the main document to ensure the safe use of the device, nor is it intended to provide diagnostic or therapeutic suggestions to the intended users.

1 Device identification and general information

1.1 Device trade name(s)

Sil-Select Stock
- Sil-Select Stock with phenol red
- Sil-Select Stock with gentamicin
- Sil-Select Stock with phenol red and gentamicin

Sil-Select Plus
- Sil-Select Plus with phenol red
- Sil-Select Plus with gentamicin
- Sil-Select Plus with phenol red and gentamicin

1.2 Manufacturer's name and address

FertiPro NV
Industriepark Noord 32
8730 Beernem
Belgium

1.3 Manufacturer's single registration number (SRN)

BE-MF-000000313

1.4 Basic UDI-DI

5411967DENSG1S5

1.5 Medical device nomenclature description/text

Applicable EMDN code: U08020502 (Materials/solutions for preparation/handling for assisted reproduction)

1.6 Class of device

Class III according to Annex VIII of the MDR

1.7 Year when the first certificate (CE) was issued covering the device

Sil-Select Stock	2009
Sil-Select Plus	2009

1.8 Authorised representative if applicable; name and the SRN

Not applicable

1.9 NB's name and single identification number

BSI Group The Netherlands BV.

NB identification number: 2797

2 Intended use of the device

2.1 Intended purpose

Sil-Select Stock (SIS) is a stock solution for preparing density gradients for semen preparation.

Sil-Select Plus (SIP) is a discontinuous density gradient system which effectively separate spermatozoa from seminal plasma, and select a subpopulation of spermatozoa with good motility, viability and chromatin integrity.

2.2 Indication(s) and intended patient groups

Although seminal plasma helps spermatozoa penetrate cervical mucus, some of its components (e.g. prostaglandins, zinc) are obstacles to the achievement of pregnancy when natural barriers are bypassed in Assisted Reproductive Technologies (ART), such as Intra-Uterine Insemination (IUI), In-Vitro Fertilization (IVF) or Intra-Cytoplasmatic Sperm Injection (ICSI). The separation of human spermatozoa from seminal plasma to yield a final preparation containing a high percentage of morphologically normal and motile cells, free from debris, non-germ cells and dead spermatozoa, is important for clinical practice.

Density gradient centrifugation is a rapid and powerful method for separating motile spermatozoa from other cell types present in human semen (including immotile spermatozoa, debris, contaminating leukocytes and seminal plasma), without causing damage to the gametes.

Direct physical contact occurs between the media products and human sperm. The products do not come into contact with the human body.

2.3 Contraindications and/or limitations

There are no known contraindications and/or limitations identified for Sil-Select Stock / Sil-Select Plus.

3 Device description

3.1 Description of the device

Sil-Select Stock is a stock solution for preparing a density gradient system for semen preparation. Sil-Select Stock consists of silane-coated silica particles in HEPES buffered Earle's Balanced Salt Solution (EBSS).

Sil-Select Plus is a gradient systems for semen preparation, which consists of a defined percentage of silane-coated colloidal silica particles suspended in HEPES-buffered EBSS. During centrifugation, cells move through the discontinuous density gradient to the point in the gradient which matches their own density. By altering the centrifugation conditions (g-force and time) and the physical properties of the colloid, a sperm pellet is formed containing the most robust, good-quality spermatozoa. The inclusion of human serum albumin (which is a medicinal substance derived from human blood plasma) in ART media from FertiPro is approved by the EMA (European Medicine Agency).

Sil-Select Stock and Sil-Select Plus are available with or without gentamicin. The added gentamicin complies with Ph. Eur. Monograph Standard 0331, is EDQM-certified and is approved by the MEB (Medicine Evaluation Board, competent authority the Netherlands)..

Density gradient media can be used in combination with IUI, IVF, ICSI and related ART. The devices are not intended for single use. Multiple single-procedures can be performed with one bottle of Sil-Select Stock / Sil-Select

Plus. The media can be used up to 7 days after bottle opening (when sterile conditions are maintained and the products are stored at 2-8°C).

Sil-Select Stock / Sil-Select Plus are sterilized using aseptic processing techniques (filtration).

In use life time for Density Gradient media is < 27 hours.

3.2 A reference to previous generation(s) or variants if such exist, and a description of the differences

Since May 2018, human serum albumin is included in Sil-Select Plus.

3.3 Description of any accessories which are intended to be used in combination with the device

No accessories for Sil-Select Stock / Sil-Select Plus are identified.

3.4 Description of any other devices and products which are intended to be used in combination with the device

Sil-Select Stock

- FertiCult Flushing medium or Sil-Select Plus Sperm Washing / Insemination medium (both manufactured by FertiPro as class III Medical Devices)

Sil-Select Plus is intended to be used with the following devices:

- Sil-Select Plus Sperm Washing / Insemination medium (except for SIP008 and SIP016) (manufactured by FertiPro as class III Medical Devices)

4 Risks and warnings

4.1 Residual risks and undesirable effects

The only remaining residual risk is the inclusion of human serum albumin (HSA) in Sil-Select Plus. The inclusion of this medicinal substance derived from human blood plasma in the devices is approved by the EMA. A potential risk associated with human serum albumin is the transmission of viral or prion-carried diseases and the batch-to batch variation:

- **Batch-to-batch variation** is still a problem because of the inherent variability in donor blood. Due to this fluctuation, standardization of procedures remains difficult.

  ↔ For this reason, a mouse embryo assay is routinely performed as part of the batch release criteria of HSA (incoming inspection) and human sperm survival assays are routinely performed as part of Density Gradient Media batch release criteria.

- Secondly; with the use of a human-derived protein source, a potential risk exists of **transmitting viral or prion-carried diseases**.

  ↔ HSA is manufactured with a pasteurization procedure that has led to an excellent viral safety record over the 50 years of clinical use. Only Plasbumin-25 or alternatively, Albunorm 25 will be used as a source of albumin, as these products are covered by a valid Plasma Master File, and the EMA has positively evaluated the usefulness, safety and benefit of the inclusion of these products in FertiPro ART-media.

  ↔ On the other hand, despite the rigorous quality controls, all cell culture media should still be treated as potentially infectious. At present, there is no known test method that can offer full assurance that products derived from human blood will not transmit infectious agents. The use of Sil-Select Plus is restricted to the sperm preparation and is not intended to be in direct contact with users or patients. Even so, the instructions for use / MSDS clearly warn that the medium

contains human albumin solution and that protective clothing should be worn.

The major benefit of HSA in density gradient media is clear:

- Inhibition of lipid peroxidation that can be damaging to sperm.
- Detoxification by binding waste products from cell metabolism.
- HAS prevents cell aggregation and adherence to laboratory equipment and promotes the ease of gamete handling and manipulation.

Based on this analysis it is concluded that the benefit of adding HSA to the media outweighs the risk and the overall residual risk related to the use of Sil-Select Plus with inclusion for human serum albumin for semen preparation has been judged acceptable.

With respect to the above, following information is provided to the customer:

- Product composition is clearly indicated on the labels and instructions for use
- Instructions for use contains the following warnings:
  - Standard measures to prevent infections resulting from the use of medicinal products prepared from human blood or plasma include selection of donors, screening of individual donations and plasma pools for specific markers of infection and the inclusion of effective manufacturing steps for the inactivation/removal of viruses. Despite this, when medicinal products prepared from human blood or plasma are administered, the possibility of transmitting infective agents cannot be totally excluded. This also applies to unknown or emerging viruses and other pathogens. There are no reports of proven virus transmissions with albumin manufactured to European Pharmacopoeia specifications by established processes. Therefore, handle all specimens as if capable of transmitting HIV or hepatitis.
  - All blood products should be treated as potentially infectious. Source material used to manufacture this product was tested and found non-reactive for HbsAg and negative for Anti-HIV-1/-2, HIV-1, HBV, and HCV. Furthermore, source material has been tested for parvovirus B19 and found to be non-elevated. No known test methods can offer assurances that products derived from human blood will not transmit infectious agents.

No other known undesirable side-effects are identified.

4.2 Warnings and precautions

Besides the above, attention should be paid to the following warnings and precautions (as described in the instructions for use):

- Do not use the product if:
  - it becomes discoloured (if medium contains phenol red), cloudy or shows any evidence of microbial contamination
  - seal of the container is opened or defect when the product is delivered
  - expiry date has been exceeded
- Do not freeze before use
- Do not re-sterilize after opening
- Products that include gentamicin should not be used on a patient that has a known allergy to gentamicin or similar antibiotics
- Aseptic technique should be used to avoid possible contamination, even when the products contains gentamicin
- Always wear protective clothing when handling specimens

- Any serious incident (as defined in European Medical Device Regulation 2017/745) that has occurred should be reported to FertiPro and the competent authority of the Member State in which the user and/or patient is established

4.3 Summary of any field safety corrective action (FSCA including FSN) if applicable

No field safety corrective actions with regard to Sil-Select Stock / Sil-Select Plus were needed.

5 Summary of clinical evaluation and post-market clinical follow-up (PMCF)

5.1 Real-world evidence analyses

A literature search is performed to investigate whether clinical data obtained during literature search are consistent with the embryological and clinical ART outcomes described in the benchmark papers from the ESHRE (European Society of Human Reproduction and Embryology) (see table below).

- The embryological outcomes must be consistent with the competency limits as reported by the ESHRE Vienna consensus group in 2017 (ESHRE Special Interest Group of Embryology 2017).

  The Vienna consensus report published in 2017 is the result of a 2 day consensus meeting of expert professionals from Sweden, Turkey, UK, Australia, Italy, Spain, Belgium, Austria, Ireland, Canada, USA, and Norway. As a starting point for the discussion, two surveys were organized to collect information on indicators used in IVF laboratories worldwide. During the meeting, the results of the surveys, scientific evidence (where available), and personal clinical experience were integrated into presentations by experts on specific topics. After presentation, each proposed indicator was discussed until consensus was reached within the panel (ESHRE Special Interest Group of Embryology 2017).

Competency limits reported by the ESHRE Special Interest Group of Embryology and Alpha Scientists in Reproductive Medicine in 2017. The Vienna consensus: report of an expert meeting on the development of art laboratory performance indicators (ESHRE Special Interest Group of Embryology 2017).	IVF normal fertilization rate: **≥60%** (lower range: 50%) ICSI normal fertilization rate: **≥65%** (lower range: 60%) ***Since multiple factors can have an influence on the embryology outcomes, (ART policy, approach of the clinic, patients characteristics), a value 10% lower than the competency limit is acceptable.***

- **Clinical ART data** obtained from IVF centers should be consistent with the clinical outcomes described in the benchmark paper from the ESHRE (Wyns et al. 2021).

  Each year, the ESHRE publishes a peer-reviewed report, which collects, analyses and reports ART data generated in Europe. The most recent report includes data from 1382 institutions in 39 countries, with a total of 940 503 treatment cycles (covering the time period from 1 January to 31 December 2017) (Wyns et al. 2021) and is summarized in the table below:

ART in Europe, 2017: results generated from European registries by ESHRE	**In vitro fertilization (IVF):**	**Intra cytoplasmic sperm injection (ICSI):**	**Frozen embryo replacement (FER):**	**Intrauterine insemination (IUI):**
(Wyns C, C De Geyter, C Calhaz-Jorge, MS Kupka, et al. 2021. 'ART in Europe, 2017: results generated from European registries by ESHRE', *Hum Reprod Open*, 2021: hoab026.)	Clinical pregnancy rate per aspiration: **28.3%** *(range: 21.0 – 42.3%)*	Clinical pregnancy rate per aspiration: **26.3%** *(range: 18.4 – 38.2%)*	Pregnancy rate per thawing: **34.4%** *(range: 22.0 – 49.1%)*	**using husband semen (IUI-H):** Delivery rate per cycle: **8.8%** *(range: 4.2 - 24.4%)*

	Clinical pregnancy rate per transfer: **36.2%** *(range: 28.4 – 47.1%)* Delivery rate per aspiration: **21.5%** *(range: 10.4 – 40.9%)* Delivery rate per transfer: **27.5%** *(range: 11.2 – 41.5%)*	Clinical pregnancy rate per transfer: **35.2%** *(range: 27.4 – 45.1%)* Delivery rate per aspiration: **20.2%** *(range: 12.2 – 36.8%)* Delivery rate per transfer: **26.8%** *(range: 17.5 – 37.4%)*	Delivery rate per thawing: **23.3%** *(range: 3.2 – 37.8%)*	**using donor semen (IUI-D):** Delivery rate per cycle: **13.8%** *(range: 4.3 – 42.0%)*

An overview of the articles studying the performance of Sil-Select Stock / Sil-Select Plus is indicated in the table below. Overall it can be concluded from these papers that embryological and ART outcomes, when Sil-Select Stock / Sil-Select Plus is used, are consistent with the outcomes as described in the benchmark papers from the ESHRE (Wyns et al. 2021; ESHRE Special Interest Group of Embryology 2017) and that Sil-Select Stock / Sil-Select Plus is as such able to select for highly motile cells with elevated DNA integrity, without being detrimental for fertilization and embryo development.

Selected articles describing the performance and/or safety of density gradient media[1]		
(Trokoudes et al. 2005)	(Vichinsartvichai et al. 2015)	(Le et al. 2021)
(Dal Canto)	(Tam Le et al. 2019)	(Kaewman et al. 2021)
(Antinori et al. 2008)	(Naji et al. 2018)	(Giebler et al. 2021)
(Berkovitz et al. 2005)	(La Marca et al. 2019)	(Rex et al. 2021)
(Berkovitz et al. 2006)	(Soysal and Ozmen 2018)	
(Fadini et al. 2011)	(Renzini et al. 2017)	
(Honda et al. 2015)	(Fujii et al. 2020)	
(Fadini et al. 2015)	(Dal Canto et al. 2021)	

5.2 Device registries

In addition to the above, ART outcomes of nine IVF clinics located in Europe are included in the clinical evaluation report of Density gradient media (data not publicly available). IVF centers were asked to provide clinical data using Density Gradient media or when ART data is published in national registers, to sign a statement that Density Gradient media were used during their ART procedures during a certain time period. Overall, it could be concluded that the ART outcomes of the IVF centres are consistent or above the national averages of their country or are consistent with the ART outcomes published in the ESHRE paper (Wyns et al. 2021), indicating that Density Gradient media of FertiPro NV not interfere with the general ART procedures.

5.3 Analysis complaints, customer/market feedback, vigilance

No additional actions were initiated, based on the cumulative nature and/or occurrence of all complaints, customer/market feedback and vigilance (if any) during the PMCF analysis.

5.4 An overall summary of the clinical performance and safety

Overall, it can be concluded that Density Gradient media function as stated by the manufacturer. This is established by clinical data (obtained during literature search and from IVF centers using the device). Moreover, there is no evidence from the clinical data, as well as from the registered complains, market/customer feedback and/or vigilance

[1] Four additional articles were retrieved that describe safety and performance of Density Gradient media. Due to reasons of confidentiality, these papers are not listed in the table. Note however that all outcomes described in these additional articles are consistent with the outcomes as described in the benchmark papers from the ESHRE.

that Density Gradient media are toxic for gametes and embryos, nor that the media have a risk for mutagenity, oncogenicity, teratogenity, carcinogenity, cytotoxicity, allergenicity and irritancy for patients and users.

5.5 Ongoing or planned post-market clinical follow-up

Post-market clinical follow-up for Sil-Select Stock / Sil-Select Plus (including PMCF for the HSA and gentamicin component included in some variants of the Density Gradient media) will be performed at least yearly and will include analyses of real-world evidence by performing a literature search, screening of device registers for clinical data, as well as analysis of all complaints, customer/market feedback, vigilance.

The Summary of Safety and Clinical Performance will be updated with information from the post-market clinical follow-up, if this is needed to ensure that any clinical and/or safety information described in this document remains correct and complete.

6 Possible diagnostic or therapeutic alternatives

Good sperm motility and normal sperm morphology are positively related to oocyte fertilization rates in vitro (Kruger et al. 1988).

The WHO manual (6^{h} edition, 2021) 'Examination and processing of human semen' describes different sperm preparation techniques to select motile and morphologically normal spermatozoa from the whole sperm. With respect to density gradients, the WHO manual states: '*Discontinuous density gradients can be used as an effective and adaptable method to collect high-quality sperm for ART. It can provide a good selection of motile sperm, free from other cell types and debris. It is easier to standardize than the swim-up technique, and thus results are more consistent. This technique is used to recover and prepare spermatozoa for use in IVF and ICSI*'.

Devices with similar intended use as Density Gradient media are available on the European Union or international markets.

7 Suggested profile and training for users

Sil-Select Stock / Sil-Select Plus are used in specialized laboratories performing fertilization techniques, including IVF, ICSI and sperm preparation/analysis. The intended users are IVF professionals (lab technicians, embryologists, or medical doctors).

8 Reference to any applicable common specification(s), harmonized standard(s) or applicable guidance document(s)

The following guidance document was used:

- **MDCG 2019-9**: Summary of safety and clinical performance A guide for manufacturers and notified bodies (August 2019).

The following technical standards apply to Density Gradient media:

- **MDR 2017/745**: European Medical Device Regulation 2017/745 of 5 April 2017.
- **(EN) ISO 13485:2016/EN ISO13485:2016/Ac:2018**: Medical devices — Quality management systems — Requirements for regulatory purposes.
- **(EN) ISO 20417:2021**: Medical devices: information supplied by the manufacturer
- **ISO 10993-1:2018/EN ISO 10993-1:2020**: Biological evaluation of medical devices -- Part 1: Evaluation and testing.
- **(EN) ISO 10993-18:2020: Biological evaluation of medical devices – Part 18: Chemical characterization of medical device materials within a risk management process**

- **ISO 13408-1:2008 (Amd 1:2013)/EN ISO 13408-1:2015**: Aseptic processing of health care products – Part 1: general requirements.
- **(EN) ISO 13408-2:2018**: Aseptic processing of health care products – Part 2: Filtration.
- **ISO 13408-6:2005 (Amd 1:2013)/EN ISO 13408-6:2011**: Aseptic processing of health care products – Part 6: Isolator systems.
- **ISO 14644-1:2015/EN ISO 14644-1:2016**: Cleanrooms and associated controlled environments – Part 1: Classification of air cleanliness by particle concentration.
- **(EN) ISO 14644-3:2019**: Cleanrooms and associated controlled environments - Part 3: Test methods.
- **(EN) ISO 14971:2019**: Medical devices – Application of risk management to medical devices.
- **ISO 15223-1: 2021/(EN) ISO 15223-1:2016**: Medical devices - Symbols to be used with medical device labels, labelling and information to be supplied - Part 1: General requirements.
- **(EN) ISO 17665-1:2006**: Sterilization of health care products – Moist heat – Part 1: Requirements for the development, validation and routine control of a sterilization process for medical devices.
- **ISO 23640:2011/EN ISO 23640:2015**: In vitro diagnostic medical devices: Evaluation of stability of in vitro diagnostic reagents (Applicable with exclusion of the following sections: No standard is available for the evaluation of stability of Medical Devices, therefore this standard is used as guideline for the set-up of the stability testing)
- **EN 556-2:2015**: Sterilization of medical devices – Requirements for medical devices to be designated 'STERILE' –Requirements for aseptically processed medical devices.
- **IEC 62366-1:2015 (Amd 1:2020)**: Medical devices - Part 1: Application of usability engineering to medical devices.
- **NBOG BPG 2014-3**: Guidance for manufacturers and Notified Bodies on reporting of Design Changes and Changes of the Quality System.
- **EMA/CHMP/578661/2010**: EMA recommendation on the procedural aspects and dossier requirements for the consultation to the EMA by a notified body on an ancillary medicinal substance or an ancillary human blood derivate incorporated in a medical device or active implantable medical device.
- **ISO 22442-1: 2020/EN ISO 22442-1:2020**: Medical Devices utilizing animal tissues and their derivatives: Part 1: Application of risk management

9 Summary of the safety and clinical performance of the device intended for patients

A summary of the safety and clinical performance of the device intended for patients, is not applicable as the device is for professional use only.

10 Revision history

SSCP revision number	Date issued	Change description	Revision validated by the Notified Body
A.1	19/12/2019	Initial version	Date: not yet Validation language: English
A.2	07/05/2020	CC191017-01: HSSA testing	Date: not yet Validation language: English
A.3	26/11/2020	Implementation remarks BSI during MDR conformity assessment + additions based on the update of the clinical evaluation report performed in 2020	Version A.3 is validated by the Notified Body Validation language: English
A.4	18/11/2021	Update 2021	Date: not yet Validation language: English

11 References

Antinori, M., E. Licata, G. Dani, F. Cerusico, C. Versaci, D. d'Angelo, and S. Antinori. 2008. 'Intracytoplasmic morphologically selected sperm injection: a prospective randomized trial', *Reprod Biomed Online*, 16: 835-41.

Berkovitz, A., F. Eltes, A. Ellenbogen, S. Peer, D. Feldberg, and B. Bartoov. 2006. 'Does the presence of nuclear vacuoles in human sperm selected for ICSI affect pregnancy outcome?', *Hum Reprod*, 21: 1787-90.

Berkovitz, A., F. Eltes, S. Yaari, N. Katz, I. Barr, A. Fishman, and B. Bartoov. 2005. 'The morphological normalcy of the sperm nucleus and pregnancy rate of intracytoplasmic injection with morphologically selected sperm', *Hum Reprod*, 20: 185-90.

Dal Canto, M. 2006. 'IVM – the first choice for IVF in Italy', *Reproductive BioMedicine Online*, 13: 159-65.

Dal Canto, M., A. Bartolacci, D. Turchi, D. Pignataro, M. Lain, E. De Ponti, C. Brigante, M. Mignini Renzini, and J. Buratini. 2021. 'Faster fertilization and cleavage kinetics reflect competence to achieve a live birth after intracytoplasmic sperm injection, but this association fades with maternal age', *Fertil Steril*, 115: 665-72.

ESHRE Special Interest Group of Embryology, ESHRE. 2017. 'The Vienna consensus: report of an expert meeting on the development of art laboratory performance indicators', *Hum Reprod Open*, 2017: hox011.

Fadini, R., R. Comi, M. Mignini Renzini, G. Coticchio, M. Crippa, E. De Ponti, and M. Dal Canto. 2011. 'Anti-mullerian hormone as a predictive marker for the selection of women for oocyte in vitro maturation treatment', *J Assist Reprod Genet*, 28: 501-8.

Fadini, R., G. Coticchio, F. Brambillasca, M. Mignini Renzini, P. V. Novara, C. Brigante, E. De Ponti, and M. Dal Canto. 2015. 'Clinical outcomes from mature oocytes derived from preovulatory and antral follicles: reflections on follicle physiology and oocyte competence', *J Assist Reprod Genet*, 32: 255-61.

Fujii, Y., Y. Endo, S. Mitsuhata, M. Hayashi, and H. Motoyama. 2020. 'Evaluation of the effect of piezo-intracytoplasmic sperm injection on the laboratory, clinical, and neonatal outcomes', *Reprod Med Biol*, 19: 198-205.

Giebler, M., T. Greither, D. Handke, G. Seliger, and H. M. Behre. 2021. 'Lower Spermatozoal PIWI-LIKE 1 and 2 Transcript Levels Are Significantly Associated with Higher Fertilization Rates in IVF', *International Journal of Molecular Sciences*, 22.

Honda, T., M. Tsutsumi, F. Komoda, and K. Tatsumi. 2015. 'Acceptable pregnancy rate of unstimulated intrauterine insemination: a retrospective analysis of 17,830 cycles', *Reprod Med Biol*, 14: 27-32.

Kaewman, P., S. Nudmamud-Thanoi, P. Amatyakul, and S. Thanoi. 2021. 'High mRNA expression of GABA receptors in human sperm with oligoasthenoteratozoospermia and teratozoospermia and its association with sperm parameters and intracytoplasmic sperm injection outcomes', *Clin Exp Reprod Med*, 48: 50-60.

Kruger, T. F., A. A. Acosta, K. F. Simmons, R. J. Swanson, J. F. Matta, and S. Oehninger. 1988. 'Predictive value of abnormal sperm morphology in in vitro fertilization', *Fertil Steril*, 49: 112-7.

La Marca, A., M. Dal Canto, M. Buccheri, M. Valerio, M. Mignini Renzini, A. Rodriguez, and R. Vassena. 2019. 'A novel transnational fresh oocyte donation (TOD) program based on transport of frozen sperm and embryos', *Hum Reprod*, 34: 285-90.

Le, M. T., T. T. T. Nguyen, T. V. Nguyen, H. N. T. Dang, and Q. H. V. Nguyen. 2021. 'Blastocyst transfer after extended culture of cryopreserved cleavage embryos improves in vitro fertilization cycle outcomes', *Cryobiology*, 100: 26-31.

Naji, O., N. Moska, Y. Dajani, A. El-Shirif, H. El-Ashkar, M. M. Hosni, M. Khalil, Y. Khalaf, V. Bolton, and T. El-Toukhy. 2018. 'Early oocyte denudation does not compromise ICSI cycle outcome: a large retrospective cohort study', *Reprod Biomed Online*, 37: 18-24.

Renzini, M.M , C. Brigante, G. Coticchio, M. Dal Canto, I. Caliari, Comi R., De Ponti E., and Fadini R. 2017. 'Retrospective analysis of treatments with recombinant FSH and recombinant LH versus human menopausal gonadotropin in women with reduced ovarian reserve', *J Assist Reprod Genet*, 34: 1645-51.

Rex, A. S., C. Wu, J. Aagaard, and J. Fedder. 2021. 'DNA Fragmentation in Human Spermatozoa and Pregnancy Rates after Intrauterine Insemination. Should the DFI Threshold Be Lowered?', *J Clin Med*, 10.

Soysal, C., and U. Ozmen. 2018. 'Intrauterine insemination in ovulatory infertile patients', *Niger J Clin Pract*, 21: 1374-79.

Tam Le, M., T. Van Nguyen, T.T. Nguyen, T.T.T. Nguyen, Nguyen T.A.T., Nguyen Q.H.V., and Cao N.T. 2019. 'Does polycystic ovary syndrome affect morphokinetics or abnormalities in early embryonic development? Article in Press', *European Journal of Obstetrics & Gynecology and Reproductive Biology*.

Trokoudes, K. M., M. B. Minbattiwalla, L. Kalogirou, K. Pantelides, P. Mitsingas, A. Sokratous, A. Chrysanthou, and S. J. Fasouliotis. 2005. 'Controlled natural cycle IVF with antagonist use and blastocyst transfer', *Reprod Biomed Online*, 11: 685-9.

Vichinsartvichai, P., S. Siriphadung, K. Traipak, P. Promrungrueng, C. Manolertthewan, and S. Ratchanon. 2015. 'The Influence of Women Age and Successfulness of Intrauterine Insemination (IUI) Cycles', *J Med Assoc Thai*, 98: 833-8.

Wyns, C., C. De Geyter, C. Calhaz-Jorge, M. S. Kupka, T. Motrenko, J. Smeenk, C. Bergh, A. Tandler-Schneider, I. A. Rugescu, S. Vidakovic, and V. Goossens. 2021. 'ART in Europe, 2017: results generated from European registries by ESHRE', *Hum Reprod Open*, 2021: hoab026.