European Society of Human Reproduction and Embryology

External Review report

"Revised guidelines for good practice in IVF laboratories (2015)"

Review period: 09/09/2015 – 21/10/2015

INVITED REVIEWERS

Open invitation:

- Email sent to members of the SIG Embryology (primary or secondary interest)
- Slide on the ESHRE website

REPORT ON THE REVIEWERS

12 reviewers responded to our invitation and have sent in their comments to the guideline.

Number of reviewers per country

Europe	*10 (83.3%)*
Germany	*2*
Cyprus	*1*
Czech Republic	*1*
Israel	*1*
Italy	*1*
Portugal	*1*
Romania	*1*
Spain	*1*
Sweden	*1*

Non-Europe	*2 (16.6%)*
Abu Dhabi	*1*
China	*1*

LIST OF EXTERNAL REVIEWERS

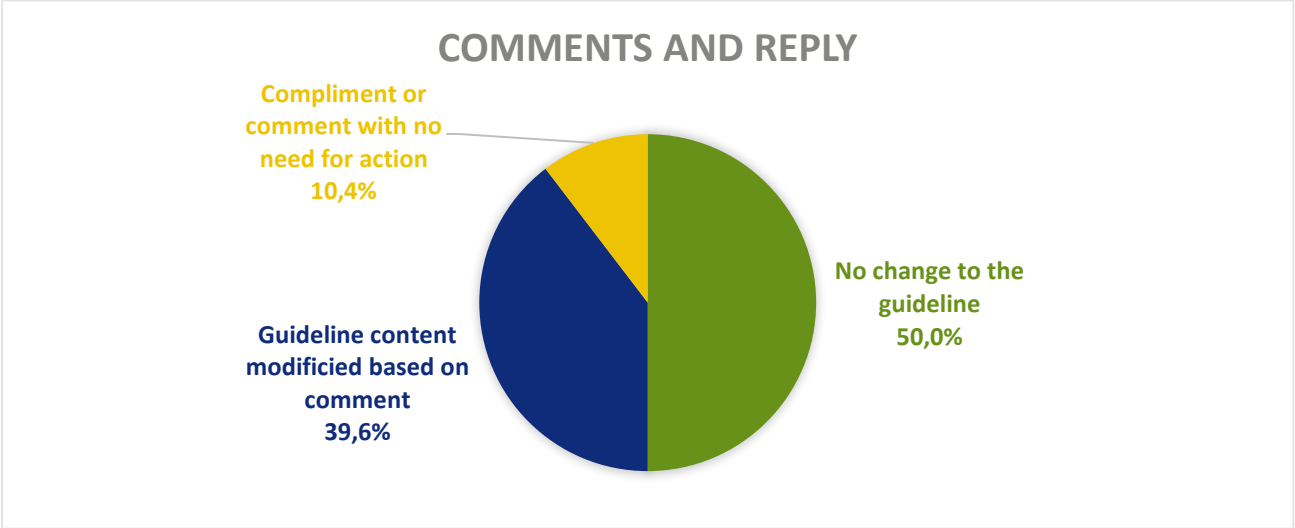
Name	Country	Organisation	COI
Pavel trávník	Czech Republic	REPROMEDA s.r.o.	No
Gianluca di luigi	Italy	University of L'Aquila, MeSVA Department	No
Asina Bayram	Abu Dhabi	IVI GCC Fertility LLC	No form
Michael Scholtes	Germany	IVF center Dusseldorf	No
Eliezer Girsh	Israel	Barzilai Univ.Med.Centre, Ashkelon. Israel	No
Julius Hreinsson	Sweden	IVF Sweden	No
Mario Sousa	Portugal	Institute of Biomedical Sciences Abel Salazar, University of Porto	No
Fang Ma and Qianhong Ma	China	West China Medical Center of Sichuan University, China	No
Sofia Johansson	Cyprus	ISIS CLINIC	No?
Rugescu Ioana Adina	Romania	AER Embryologists Association	(Yes)
Montse Boada	Spain	ASEBIR	No
Verena Nordhoff	Germany	QM Study Group and the Steering Board of the German Society of Human Reproductive Biology (AGRBM):	No

STRATEGY FOR HANDLING COMMENTS

106 comments were received from 12 reviewers.

All comments are summarized in the tables below (per section). Each comment was assessed by the research specialist, the guideline group member responsible for the section, and the chair of the guideline group.

- 11 comments were compliments to the guideline or statements of agreement with the content.
- 95 comments were formulated requesting a change in the guideline, of which 42 were considered valid and resulted in a modification in the guideline text, either the correction of an error, a rephrasing, or a modification of the content of the guideline. The remaining 53 comments were assessed, but did not result in a change in the guideline. A reply to the reviewer was formulated in the tables below.
- The numbers are summarized in the graph below.



COMMENTS (PER SECTION)

SECTION 1: STAFFING AND DIRECTION

Reviewer	Comments	Reply
Michael Scholtes	Page 1, line 8,11: the laboratory staff should be defined according to the number of ovum pick up or IVF/ICSI procedures in the laboratory. Especially a minimum number should be fixed. Line 32: the same, a minimum number of staff should be indicated	We have added a sentence on how to determine the number of staff.
Mario Sousa	Our directives from the portuguese National Council for Reproductive Medicine are different. The Director has to be a medical doctor with specialization in Gynecology & Obstetrics, in Urology, in Endocrinology or Medical Genetics. Our directives for laboratory supervisors are different, it has to have a MD degree and a Senior Clinical Embryologist certification Our directives for Clinical embryologists require at least 5 years of experience and a Clinical embryologist certificate. Regarding the functions, our directives are similar	Thank you for this information on the Portuguese Directives We cannot get such degree of detailed information, however we have added MD in the academic degree for the laboratory director
Rugescu Ioana Adina	1. page 1; line7-8 Clinics that perform up to 150 retrievals and/or cryopreservation cycles per year there should be a minimum of two qualified embryologists. Three embryologists should be required to perform between 151 and 300 cycles per year; four embryologists to perform between 301 and 600 cycles; and from 600 cycles on per year one additional embryologist is needed every 200 additional cycles. Additional laboratory staff may be required if andrological and/or endocrinological duties are assigned. (At least as recommended number of laboratory staff, not necessary as mandatory) 2. page1 line 20 - or similar 3. page1 line 30 - Develop, implement, maintain and improve a.... 4. page1 line31 - Develop, implement, maintain and improve.... 5. page1 line 35 - key performance indicators (KPIs) - I think a minimum process indicators that shell be checked list it is better to be included. 6. page2 line 42 or similar	1. We have added a sentence on how to determine the number of staff, similar to your suggestion, but we decided not to add a table, as calculating the basic size of the staff it is more complex than that per number of cycles 2. We added “or similar” as suggested. 3. By implementation of a QMS, we assume develop, implement and maintain, and therefore we have not adapted the sentence. I don’t think this should be changed 4. By implementation of a QMS, we assume develop, implement and maintain, and therefore we have not adapted the sentence. I don’t think this should be changed 5. KPI in itself is a very complex matter that requires to be specifically addressed in a future paper. 6. We added “or similar” as suggested
Montse Boada	1. Page 1 Line 20 and Page 2 lines 43 and 55: ESHRE senior embryologist certification "or similar"	1. We added “or similar” as suggested

	2. Page 1 Line 24: Add "Promote continual training of staff members and education of new embryologists and students." 3. Page 2. Line 45: Add "Management of laboratory daily incidences solving them efficiently and quickly. 4. Page 2. Lines 48 and 60: staff members "and students"	2. We feel this is covered in 1.1.7 “Management of laboratory staff training and continual scientific and biomedical education” ,and therefore have not added the sentence as suggested 3. We feel this is covered in “daily work, and continuous improvement” ,and therefore have not added the sentence as suggested 4. We added “students” to the sentence, as suggested
Verena Nordhoff	1. page 1 line 17, COMMENT: specify 'a higher academic degree' e.g. as equivalent to M. Sc. or PhD 2. page 1 line 20, COMMENT: also attainment of national certifications as clinical embryologist should be mentioned 3. page 1 line 23, ADD: is obliged to fulfil ESHRE or national requirements 4. page 1 line 34, ADD: fulfilling national and/or ESHRE 'continuous professional development' (CPD)	1. Based on your suggestion we added “(MD, M.Sc., PhD)” 2. We added “or similar” as suggested by another reviewer, and thereby already addressed this comment. 3. We decided not to add the suggested sentence: ESHRE can recommend to follow the national legislation, but we cannot oblige. 4. We believe our more general statement is fine, and have decided not to add more details.

SECTION 2: QUALITY MANAGEMENT

Reviewer	Comments	Reply
Michael Scholtes	Page 2, line 69: long-term follow up of pregnancies and children is mandatory	We agree that long-term follow up of pregnancies and children should be mandatory, but it is under direct responsibility of the clinic or the ART centre, not the laboratory. Therefore, we did not add long term follow up in the document.
Mario Sousa	Our directives are similar. We have internal (each year), external (each year) and National (every two years) certification. For the remainder, all items are in accordance with our guidelines.	Thank you for this information on the Portuguese Directives
Rugescu Ioana Adina	page3 line 108 Participation in Internal Quality Control (IQC) must be mandatory and External Quality Assurance (EQA) programme is recommended.	Both IQC and EQA are recommended in the document. We have modified this to “highly recommended”, but

		not mandatory, as IQC and EQA are not mandatory everywhere.
Verena Nordhoff	page 2 line 82, CHANGE TO: The QMS (not the SOPs) must include provisions for	We have corrected SOPs to QMS.

SECTION 3: LABORATORY SAFETY

Reviewer	Comments	Reply
Pavel Trávník	3.6 Protective measures; Page 6; Line 220: Alcohol-based disinfectants should be avoided. This statement I keep for very shallow from these reasons: 1. Alcohol itself is very good disinfectant 2. I did not find any valid data concerning the injurious influence of very small concentrations of ethanol originating from the possibly in the air present residues of ethanol vapours on the preimplantation embryos 3. There exist a lot of not here prohibited disinfectants containing cytotoxic, mutagenic and other possibly harmful compounds	We have deleted the sentence as requested. There have been reports on the detrimental effects of alcohol (Sally Catt, ESHRE oral presentation 2012). However, due to comments in point 3, we decided to remove the sentence altogether.
Gianluca Di Luigi	1. 3.6 Pag. 6 line 220: Alcohol or IODINE-based 2. Pag.6 line 222: NO PERFUMES	1. We deleted the sentence based on another comment, because it focussed on alcohol when there are other prohibited disinfectants. Therefore, iodine-based was not added. 2. We have changed the sentence to “use of cosmetics should be minimised and perfumes should be avoided.
Michael Scholtes	1. Page 3: in this section transport IVF has not been addressed at all. Should be commented 2. Page 4, 3.2: v.o.c. particle measurement regurlarly	1. The group feels that transport IVF is a specific situation and should not specifically be mentioned, although we assume that all specifications will apply to transport IVF too. 2. We changed VOC filtration to ”VOC control” as filtration is not the only technique that can be used to decrease the VOC levels.
Ma Fang	1. Page 4, line 151: In this part, should we add: Are the maintenance of equipment required as a regular time under mandatory Certification. 2. Page 4, line 163:High-purity gas and inline HEPA and VOC filters should be used----about these, do they must?	1. Thank you for your comment, but we believe this point is covered, and there is no need to add a sentence.

	3. Page 6, line 223:not wear jewellery, watches and wristbands.----should we mention the place where shouldn't wear, like the neck, ear,ankle which might be not significant effect , if on the wrist, how about the protectable cover could work.	2. Thank you for your comment. We changed this sentence to , High-purity gas and inline HEPA and VOC filters are recommended 3. We deleted the sentence based on your comment and added "to diminish possible sources of contamination" to the new sentence .
Rugescu Ioana Adina	1. page3 line125: the minimum work space per person is about 1.5m long by 1.2 m deep. this depth is made up of 70cm workspace plus 50cm space for the seated or standing worker. in addition, a passage space is necessary behind the worker of about 0.5m. A laboratory design with two parallel rows must take into consideration all of above. 2. page 4 line 141 - particle counts as per ISO 14644-1 annex B5 3. page4 line 142 - or HVAC systems (M13; F8 and G4 plus filters - EN779 and EN1822/ ISO14644-3 4. page 4 line 143 - 15Pa as per ISO14644-3 annex B5 5. page 4 line 148 - grade A or ISO5 class 6. page 4 line 150 - grade D or ISO 8 class 7. page 4 line 150 - air velocity/ISO14644-3 annex B4 -0.36 up to 0.54m/s; room air exchange 6-15 exchange/hour; air flow as per ISO 14644-3 annex B13 laminar flow from up to down 8. An embryology laboratory must control the lighting 2700-3000 grade K. Yellow filters for microscopes are recommended. 9. relative humidity; temperature, electromagnetic fields and noise.....are also not mentioned; 10. page5 line 193 (maybe the minimum requirements must be mentioned)	1. We added "minimum work space per person" to the list of considerations, but we decided not to add the details. 2 To 8: We have assessed your suggestion, but decided that the high level of detail is not necessary, and therefore we did not add these details to the section. 9. We have mentioned relative humidity and temperature, other parameters such as electromagnetism may be considered in future editions 10. We state "screened for infectious diseases according to national and international regulations", and assume this is sufficient.
Montse Boada	1. Page 4 Line138: add after fixatives "and other toxic reagents" 2. Page 4. Lines 154, 172: It will be very useful to include a list of the equipment considered "critical" (Incubators and crystorage units Line 172, and someone else??	1. As suggested, we added "and other toxic reagents" 2. We decided not to expand the list of critical equipment, as suggested.
Verena Nordhoff	1. page 4 line 134, CHANGE TO: a system for clean access of personnel and materials to the laboratory is recommended 2. page 4 line 136, DELETE: there are different national regulations which should not be generalized in this guideline 3. page 4 line 141, CHANGE TO: laboratory air could be subjected to .... 4. page 4 line 143, CHANGE TO: positive pressure is optional to minimize air contamination 5. page 4 line 150, CHANGE TO: However, if it is detrimental or not feasible to carry out a specific procedure in a Grade A environment, or the mode and application of the cells implies a significant lower risk of transmitting infection than with transplantation, it can	1. We have changed "necessary" to "highly recommended" as suggested. 2. Thank you for the comment on Hand washing facilities, but after discussion, we decided not to modify the sentence. 3. We decided not to change the sentence and kept "laboratory air should be subjected to.." 4. We decided to leave "positive pressure is recommended"

	be performed in an appropriate environment. In these cases the environment needs to be specified with regard to quality and safety of the recipient. Therefore clean room conditions are not obligatory for the performance of IVF procedures. COMMENT TO THIS CHANGE: Please pay attention to TD 2006/86/EC, Annex I, D Facilities/Premises, 4. c, and 5. 6. page 4 line 163, CHANGE TO: VOC filters are recommended 7. page 4 line 165, CHANGE TO: Heating devices must be installed to maintain the temperature 8. page 5 line 180, CHANGE TO: Cryostorage units should be regularly monitored and alarm systems are recommended 9. page 5 line 209, ADD: preferably with low particle shedding 10. page 5 line 211, CHANGE TO: Use of appropriate vertical laminar flow benches for handling of biological material is recommended. 11. page 5 line 212, CHANGE TO: Use of mechanical pipetting devices is recommended 12. page 5 line 215 Viral-positive wastes should be segregated into a separate bin, labelled and disposed of according to biosafety policies.	5. This sentence was extensively discussed within the guideline group and we feel our reached consensus should be maintained 6. Thank you for your comment. We changed this sentence to , High-purity gas and inline HEPA and VOC filters are recommended 7. We debated this at length in the group and decided to leave it as it is. 8. After discussion we decided to leave the sentence as it was originally in the guidelines; cryostorage units are critical equipment and therefore an alarm system must be put in place. 9. We added “preferably” as suggested 10. We decided not to change this, as the phrasing is consistent with the other items in the list. 11. Similarly, we decided not to change this, as the phrasing is consistent with the other items in the list. 12. We changed the sentence as suggested: “Viral-positive waste should be segregated into a separate bin, labelled and disposed of according to biosafety policies”
Mario Sousa	All items follow our guidelines.	We are happy to hear our recommendations are similar to the recommendations in the Portuguese Directives

SECTION 4: IDENTIFICATION OF PATIENTS AND TRACEABILITY OF THEIR REPRODUCTIVE CELLS

Reviewer	Comments	Reply
Gianluca Di Luigi	4.4 Pag. 6 line 248: Double-checks MUST be done (if necessary it's possible to check with the gynaecologist)	We have changed the sentence to double-checks are necessary
Ma Fang	Page 6, line 232:each patient's unique---after the patient.....Can be required to check the name and the information of the legal husband in the process of recognition	We have reviewed your comment, but decided not to add further details to the sentence.
Montse Boada	Page 6 Line 248: It will be very useful to include a list of "critical" steps/procedures (Insemination Line 358, transfer Line 439, and someone else???	Based on your suggestion, we have added a list of critical steps.
Verena Nordhoff	page 6 line 243 + page 7 graph box, DELETE EXPRESSEION: 'cycle identification code' EXPLANATION: The date of treatment together with the unique Patient ID is sufficient to uniquely identify the cycle	Based on your suggestion, we have changed "cycle identification code" to "date of treatment"

SECTION 5: CONSUMABLES

Reviewer	Comments	Reply
Michael Scholtes	Page 7: 5.5: appropriate refrigeration define temp °C mimimum/maximum	We recommend that "Appropriate refrigeration facilities must be available for storage of media and reagents", but have decided not to further specify a range of temperatures, as these may vary for different media and reagents.

SECTION 6: HANDLING OF BIOLOGICAL MATERIAL

Reviewer	Comments	Reply
Verena Nordhoff	page 8: line 289, CHANGE TO: Handling of biological material should be easy, simple and effective and may be performed in a laminar flow hood which should be equipped with heating stages and pre-warmed heating blocks, using aseptic techniques at all times.	We have changed the sentence to: "and should preferably be performed in laminar flow hoods" in reply to your comment.

SECTION 7: OOCYTE RETRIEVAL

no comments

SECTION 8: SPERM PREPARATION

Reviewer	Comments	Reply
Eliezer Girsh	Comment 1: Sperm from patient should be washed as soon as possible after acceptance to laboratory (of course after its liquification). Prolonged spermatozoa exposure to seminal plasma is not recommended, because seminal fluid is not friendly to spermatozoa. Spermatozoa are negatively affected by seminal plasma as time is prolonged. Comment 2: After sperm is thawed it should be no more refreezed - we do not know yet if second freezing affect or not spermatozoa and how many sperm cells are damaged (nuclear alterations) after the second freeze/thaw cycle, even if they are still moving. At the same point, do not prepare partial thaw of the sperm (for use part of it), because partial thaw and then refreeze can also negatively affect inner nuclear organization, as compaction of DNA, protein degradation, free radicals production).	1. In reply to this comment, we included the following sentence: "Prolonged spermatozoa exposure to seminal plasma is not recommended." 2. We acknowledge the comment and we will consider this information in case we decide to expand the section on sperm preparation. However, we do not agree with adding this information in the current document.
Ma Fang	1. Page 9, line 317, about sperm preparation, the important treatment ob sperm--capacitaion didn't mention, maybe should be added or completed in this part. 2. Page 9, line 338, Could we also consider that how to the treat the men with a difficulty of obtaining sperm.	1. We acknowledge the comment and we will consider this information in case we decide to expand the section on sperm preparation. However, we do not agree with adding this information in the current document. 2. We have mentioned in the introduction of section 8: "A frozen back-up sample should be requested if sperm collection difficulty on the day of oocyte retrieval is anticipated." We feel there is no need to elaborate further on this issue.
Verena Nordhoff	1. page 9 line 338, CHANGE TO: In case of azoospermia on the day of oocyte retrieval, alternative sperm retrieval procedures or oocyte cryopreservation should be considered. COMMENT: Treatment options should not be generalized. 2. page 9 line 341, COMMENT: it should be stated that the WHO gives no recommendation regarding selection of immotile but viable sperm, therefore either this sentence should be deleted as techniques might change over time; or all methods (also the sperm tail flexibility test (STFT) and the laser assisted immotile sperm selection (LAISS) test) should be mentioned.	1. We have assessed your comment and changed the recommendation to: In case of azoospermia on the day of oocyte retrieval and in the absence of a back-up sample, alternative sperm retrieval procedures or oocyte cryopreservation should be considered. 2. We have decided, based on your comment, to remove point 8.6 from the document.

SECTION 9: INSEMINATION OF OOCYTES

Reviewer	Comments	Reply
Gianluca Di Luigi	9.2 Pag. 10 line 380: Polar body position at h 6 ore 12 9.2.2 Pag. 10: MINIMIZE LIGHT EXPOSITION	1. After discussion, we decided not to change the sentence. There are no conclusive data about where the polar body should be. 2. We added "exposure to light should be minimised" in the section 6 (handling biological material)
Asina Bayram	My comment will be about 9.2.1 Preparation of oocytes for ICSI part. It was explained well but nothing was mentioned about the duration between OPU and denudation. I think there should be more explanation about how long we should wait after OPU. For the guideline, given an optimum time will be usefull.	We added a sentence on the timing of denudation: "Current evidence does not suggest that denudation should be performed at a specific time between oocyte recovery and ICSI." However, since denuded oocytes are more vulnerable to pH changes, we recommend to postpone denudation as much as possible to prior to injection
Eliezer Girsh	Comment 1 (for ICSI section 9.2.1): It is recommended to perform denudation of oocytes immediately or close to OPU. In this case immature (MI stage) oocytes could mature before procedure of ICSI. Cumulus cells provide cAMP to oocyte and prevent oocyte maturation. Denudation is shortened the time of oocyte maturation. Comment 2: Micromanipulation should be prepared under laminar flow conditions to keep sterility of injection and holding micro-capillars.	1. We have added a sentence on the timing of denudation, but decided not to add the sentence as suggested. 2. The group feels that very few labs prepare micromanipulation under laminar flow conditions, suggesting there is no need. Therefore this was not added in the section.
Mario Sousa	All items follow our guidelines. However: 9.2.2: We do not microinject oocytes with small size, large size, large polar body, large smooth endoplasmic reticulum tubule aggregates and vacuolization.	We have added that oocytes with a very large polar body should not be injected, but decided not to add further details.
Sofia Johansson	*The timing of the ICSI is not mentioned. If the guideline is still 37-41 hours after hCG it would be good to mention it in this section (page 10, section 9.2.2)*	Thank you for this comment. The group discussed this sentence intensively and decided not to mention anything on hCG, as there are not many references about this subject. Therefore we decided to leave the sentence unchanged.
Montse Boada	Page 10 Line 382 Add: "(wishes prepared with pre-warmed oil and medium, heating stages and minimal time out of the incubator)." It will be very useful to establish the maximum time out of the incubator.	Thank you for your comment, but we decided not to add this information to the text, as it may depend on

		other variables such as type of mineral oil, storage conditions before use, etc.
Verena Nordhoff	1. page 9 line 354, DELETE SENTENCE; EXPLANATION: no recommendation on the addition of a specific sperm concentration should be given. This is the freedom of treatment and lies in the responsibility of each lab. 2. page 9 line 356, DELETE; EXPLANATION: It should be in the responsibility of each lab to decide which kind of medium should be used.	1. We agree that this is the freedom of treatment and the responsibility of the lab, but we think readers would value an indicated range for the concentration. 2. We agree that labs should be able to decide which medium they use, and therefore we only give recommendations on factors they should take into account when choosing a medium (compatibility and glucose)

SECTION 10: SCORING FOR FERTILIZATION

Reviewer	Comments	Reply
Julius Hreinsson	Page 10, line 389. One comment: A normally fertilized oocyte has 2pn but may also contain 3 polar bodies or 1polar body + fragments. The current formulation may contribute to a misunderstanding regarding pb-status.	Thank you for your comment. We agree that there will always be many details and exceptions, but we feel that we should keep details to a minimum to ensure the document to be readable and clear.
Mario Sousa	All items follow our guidelines. However: 10.4: we do not transfer embryos from 1PN zygotes.	We have changed the sentence to : “Embryos derived from ≥ 3PN oocytes should never be transferred or cryopreserved. Even if no transferable embryos derived from 2PN oocytes are available, the use of embryos derived from 1PN oocytes or oocytes showing no PN is not recommended.“
Montse Boada	1. Page 10 Line 393: Add after inverted microscope... equivalent optics "or time-lapse devices" 2. Page 10 Line 394: Delete "At least". 3. Page 10 Line 397: Add another point: "Presence of cytoplasmic halus" 4. Page 11 Line 399: Change "if no transferable embryos derived from 2PN oocytes" by "if no good quality embryos derived from 2PN oocytes" 5. Page 11 Lines 399-340:Add at the end of the sentence "if two polar bodies can be observed". It is not recommended to transfer these embryos if they have only one polar body .	1. We added “or time-lapse device” as suggested 2. Based on this and other comments, we have removed section 10.3 3. Based on this and other comments, we have removed section 10.3 4. We decided not to change transferable to good quality embryos. However, the sentence was revised and reformulated

		5. We have changed the sentence to : "Even if no transferable embryos derived from 2PN oocytes are available, the use of embryos derived from 1PN oocytes or oocytes showing no PN is not recommended.", but we did not add the requested information on one or two PB.
Verena Nordhoff	1. page 10 line 394, CHANGE TO: At least the following characteristics of the 2PN oocytes may be recorded 2. page 11 line 399, INSERT: …circumstances, and only with informed consent with the patients, ...	1. Based on this and other comments, we have removed section 10.3 2. We decided not to incorporate the requested change, but revised and reformulated the entire sentence

SECTION 11: EMBRYO CULTURE AND TRANSFER

Reviewer	Comments	Reply
Montse Boada	Page 11 line 417: It will be very useful to list the standardised check times recommended by ESHRE	Thank you for this suggestion, but we know this information is presented in other papers, namely the consensus (ESHRE and Alpha) on embryology evaluation, so we feel there is no need to repeat it here.

SECTION 12: CRYOPRESERVATION

Reviewer	Comments	Reply
Gianluca Di Luigi	12.8 Pag. 12 line 479: a double-check MUST be done (if necessary it's possible to check with the gynaecologist)	Thank you for your comment, but we decided to leave the sentence as it was.
Mario Sousa	All items follow our guidelines. However: 12.2: we do not use sloww freezing for sperm.	We mentioned the two current methodologies, without formulating any further specifications or recommendations. Therefore we did not change the section.
Verena Nordhoff	1. page 12 line 448, ADD: pronuclear stage oocytes 2. page 12 lines 444-451, COMMENT: methods do change over time, and it should be avoided to include recommendations in such a detailed manner	1. We added pronuclear stage oocytes as suggested.

	3. page 12 line 456, CHANGE TO: It is recommended to avoid direct contact of the biological material with the LN2. COMMENT: To date, there is no published report of cross-contamination between reproductive tissues in cryogenic storage, neither in livestock industries nor in human IVF.	2. We feel referring to slow freezing and vitrification is not too detailed, as these are the two current methodologies. 3. We have considered your comment, and we have rewritten the sentence to : "Safety issues have been raised regarding direct contact of the biological material with the LN2, however we cannot favor at this point closed over open devices. Laboratories will need to adjust their decisions upon their results, risk analysis and regulations in place

SECTION 13: EMERGENCY PLAN

Reviewer	Comments	Reply
Michael Scholtes	Anyone; processing germ cells and cryopreserving germ cells or embryo / doing embryo transfer should mandatory have a data backup programme for bankruptcy, going out of business. A licensed center or public health institutions should guarantee a secure long term storage.	Thank you for this comment. We agree with it, but this issue should be covered in the emergency plan of clinic, not the lab specifically.
Mario Sousa	All items follow our guidelines. However: we also have a regular training on ressuscitation.	We agree with your comment, but training on resuscitation should be covered in the emergency plan of clinic, not the lab specifically.
Rugescu Ioana Adina	page 13 line 484 - to develop ,implement, maintain and improve an emergency plan	We added "and implement" to the sentence.

ANNEX 1 : METHODOLOGY

Reviewer	Comments	Reply
Mario Sousa	We acknowledge the inclusion of our additions.	The review report will be published on the ESHRE website.
Montse Boada	Perfect	Thank you.

GENERAL COMMENTS

Reviewer	Comments	Reply
Pavel Trávník	In general, the Guidelines I keep for very good made and comprehensible.	Thank you.
Gianluca Di Luigi	It's really important to guarantee ad international registry for all the procedures, specially for the heterologous ones (with an international, shared list of donors). If an IVF Centre wants to be considered ad excellent one, it must inevitably demonstrate to perform a line of research inside.	Thank you for this comment. We have formulated recommendations for good practice, but to be feasible for both small and large laboratories, we feel we cannot state that research is a necessity.
Michael Scholtes	These guidelines are necessary for everyone in the field of fertility treatment. Transparency for the general public and especially politicians is needed, for approval as well as public funding of research. The European Parliament should endorse these guidelines	Thank you.
Mario Sousa	We acknowledge the inclusion of our additions. References: -Portuguese Law on Medical Assisted Reproduction. -Guidelines on Medical Assisted Reproduction. -Sá etal, Fertil Steril, 2011, 96(1): 143-149. Smooth endoplasmic reticulum aggregates. -Azevedo et al, Reprod Sci, 2014, 21(12): 1472-1482. 1PN zygotes.	
Ma Fang	This is a brief IVF labs guideline, which can be referred widely and help the IVF lab more standardized and safe as a keep leaning way. Also staff, patients and their family might benefit more .	Thank you.
Rugescu Ioana Adina	maybe a chapter with terms and definitions should be included. it is mandatory to have clear definitions of the structure of the laboratory organization and the responsibilities of the laboratory personnel. This means that the organizational chart should reflect the reality of the institution and should be kept up to date. Organizational charts and job descriptions will give an immediate idea of the way in which the laboratory functions and the relationships between the different departments and posts and must be integrated with the clinic structure. Records of repairs and Routine maintenance and of any non-routine work on equipment should be retained. Maintenance of the equipment may be carried out in two quite distinct ways: preventive maintenance and curative maintenance I could not find control of documents and control of records, also I did not see more on personnel training.	We will prepare glossary and publish it alongside the guideline on the ESHRE website. We have considered your other comment, but we feel most have been discussed in the paper.

Montse Boada	Good work. Congratulations to the GDG!!	Thank you.
Verena Nordhoff	This review was compiled by eight embryologists: the QM Study Group and the Steering Board of the German Society for Human Reproductive Biology (AGRBM). COMMENT TO Laboratory safety, page 4 line 150: In Germany more and more IVF clinics face the problem, that the authorities claim clean room conditions by referring to the EUTD. However, in our opinion the exceptions (TD 2006/86/EC, Annex I, D Facilities/Premises, 4. c, and 5.) are fully effective for IVF labs. Therefore, our society of embryologists, the AGRBM, has changed their guidelines in order to explain, why clean rooms are NOT necessary in ART. We would very much appreciate if the ESHRE guidelines make this point very clear to support the indispensable argumentation against the requirement for clean rooms.	Thank you for submitting your comments. We have tried to provide good practice recommendations that could be followed by all IVF labs, without providing too many (often country-specific) rules and regulations.