January 2023

Unexplained infertility Guideline Group

Unexplained Infertility

European Society of Human Reproduction and Embryology

Developed in collaboration with the Monash University led NHMRC Centre of Research Excellence in Women's Health in Reproductive Life (CREWHIRL).

REVIEW REPORT

CREWHiRL
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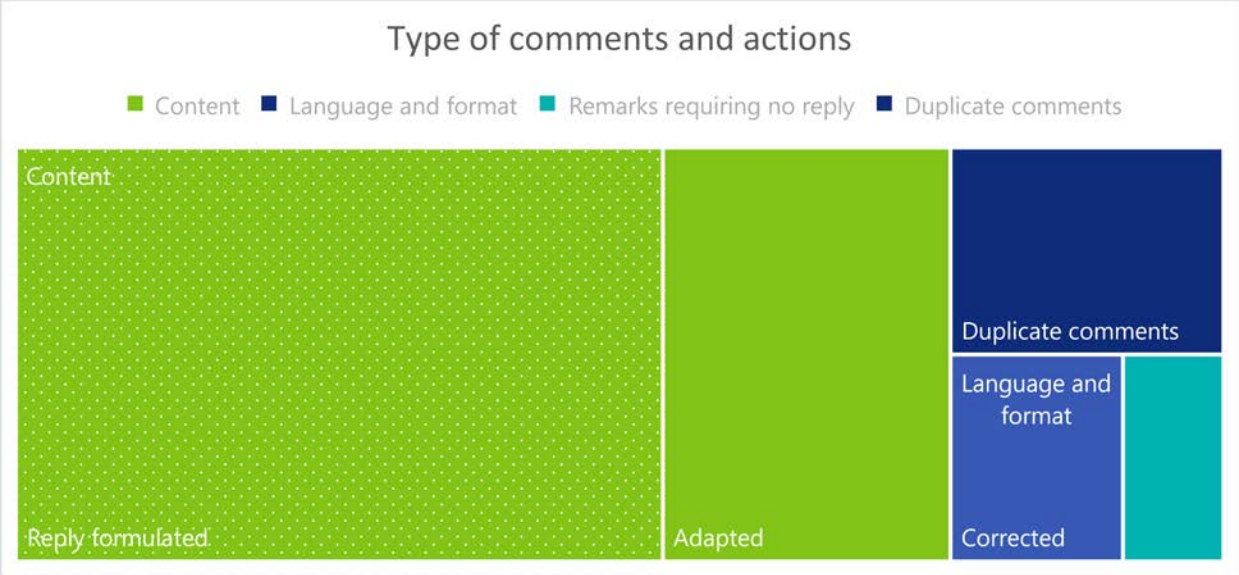
The draft of the evidence-based guideline "Unexplained Infertility" was published for public review for 6 weeks, between 12 December 2022 and 30 January 2023.

This report summarizes all reviewers, their comments[1] and the reply of the guideline development group and is published on the ESHRE website as supporting documentation to the guideline.

During the stakeholder review, a total of 336 comments (including 38 duplicates) were received from 41 reviewers.

The comments were focussed on the content of the guideline (260 comments), language and style (24 comments), or were remarks that did not require a reply (14 comments). All comments to the language and format were checked and corrected where relevant.

The comments to the content of the guideline (n= 260) were assessed by the guideline development group and where relevant, adaptations were made in the paper (n= 80; 31 %). Adaptations included revisions and/or clarifications of the text, and amendments to the recommendations. For a number of comments, the working group considered them outside the scope of the paper or not appropriate/relevant (n= 180; 69 %).



[1] Two comments were not included in this review report because of inappropriate language.

Experts that participated in the stakeholder review

The list of representatives of professional organization, and of individual experts that provided comments to the guideline are summarized below.

Representatives of professional organisations

Organisation	Country	Representative
German Society of Andrology (DGA e.V.) **German Society of Urology, working group Andrology**	Germany	Sabine Kliesch
Institute of Reproductive Medicine, Kolkata	India	Pratip Chakraborty
Centre for Human Reproductive Science, Birmingham Health Partners, The University of Birmingham	UK	Jackson Kirkman-Brown, Meurig Gallagher
Reproductive medicine Amsterdam UMC, The Netherlands and the Netherlands Cochrane Gynaecology&Fertility	The Netherlands	J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn
European Academy of Andrology	Europe	Dimitrios G. Goulis Giovanni Corona
German Society for Reproductive Medicine (DGRM) and URZ	Germany	Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.
Bundesverband Reproduktionsmedizinischer Zentren (BRZ)	Germany	Ulrich A. Knuth, Michael Ludwig
German Society of Human Reproductive Biology	Germany	Verena Nordhoff
Unexplained Infertility Guideline Australian Adaptation Committee	Australia	

Individual experts

Reviewer	Country
Joel Bernstein	Australia
Gustavo Botti	Argentina
Maruf Siddiqui	Bangladesh
Jean Calleja-Agius	Malta

Hunida Elmegrab	Libya
Bulent Tandogan	Turkey
Carlos Calhaz-Jorge	Portugal
Liliana Ramos	The Netherlands
Michael Morris	Switzerland
Adam Balen	UK
George Lainas	Greece
Mahmoud A Abdel-Aleem	Egypt
Marco Sbracia	Italy
Mario Sousa	Portugal
Maria Elisabetta Coccia	Italy
Michael Grynberg	France
Exalto N. **Emanuel MH.**	The Netherlands
Ben Mol	Australia
Mitranovici Melinda Ildiko	Romania
Panayotidis Costas	Greece
Nusrat Mahmud	Bangladesh
Aboubakr Mohamed Elnashar	Egypt
Mira Töyli	Finland
Petya Andreeva	Bulgaria
Kalmantis Konstantinos	Greece
Åsa Magnusson	Sweden
Christina Bergh, **Jan Bosteels** **Frank Broekmans** **Astrid Cantineau** **Arri Coomarasamy** **Vinh Dang** **Annemieke Hoek** **Joop Laven** **Rong Li** **Abha Maheswari** **Ben W. Mol** **Anja Pinborg** **Annika Strandell** **Chris Venetis** **Lan Vuong** **Madelon van Wely**	Sweden Belgium The Netherlands UK Vietnam China Australia Denmark
Maria Schubert	Germany
Monica Varma	India

Christophe Blockeel	Belgium
Priya Bhide	UK
Lars Björndahl	Sweden

Reviewer comments and replies

Reviewer	Page	Line	Comment	Action / Reply
TITLE				
Sabine Kliesch			Finally, we suggest to either rename these guidelines into 'female unexplained infertility` which reflects the current content of the guideline or to include further evidence-based aspects and experts in the andrological field. For the latter we are happy to contribute with suggestions on further experts in the field.	The guideline covers all aspects of UI, male and female, so the GDG sees no need to change the title.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn			We think it is appropriate to name the guideline "diagnostic work-up and treatment for unexplained infertility"	The GDG discussed your suggestion. However, the guideline covers all aspects of UI, so no need to elaborate in the title. Furthermore, the titles of all ESHRE evidence-based guidelines are restricted to the name of the condition
INTRODUCTION				
Jackson Kirkman-Brown Meurig Gallagher	/	/	The abbreviation UI is liable for confusion with the term uterine insemination (as in IUI) we would instead suggest UEI	The GDG understand where possible confusion could emerge, however, UI is the predominantly used abbreviation for unexplained infertility in literature.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell			To provide clinicians (and infertile couples?) with evidence-based information on the optimal diagnostic work-up for infertile couples based on the examinations and procedures available to date, in order to correctly confirm the diagnosis of UI. Suggest to replace the word confirm by establish. The word confirm suggest that there is already a suspicion of unexplained infertility. Instead, the guideline should start with the question how to establish the diagnosis of UI.	This was adapted as suggested.

Chris Venetis Lan Vuong Madelon van Wely		
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	To provide clinicians with evidence-based information on the optimal therapeutic approach considering issues like live birth rates, safety, burden of testing and treatment, and individualization. Individualization could be made more explicit throughout the guidelines (for example values, preferences and prognosis)	This was adapted as suggested.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	An important additional aim would be to provide clinicians with evidence-based information on the impact of knowledge on the causes of the infertility: knowing for the sake of knowing. This is partly formulated in recommendation 55 but could be formulated as a separate aim. This new aim also requires separate PICOS. This is a classic reference: Asch DA, Patton JP, Hershey JC. Knowing for the sake of knowing: the value of prognostic information Med Decis Making 1990 Jan-Mar;10:7-57. doi: 10.1177/0272989X9001000108.	This is included in the concept of patient satisfaction and quality of life (among the important outcomes). The last chapter reviews the value in terms of QoL of knowing the cause of infertility for both men and women. Furthermore, in the diagnostic PICO questions, the reliability/accuracy of each test was included in the outcomes

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely			It is not defined if the guideline aims at high-resource settings, low-resource settings or both.	This was better clarified as suggested.
George Lainas	5	72	Reference	A reference was added to the text.
Mario Sousa	5	72	About 30%-40% of infertile couples are considered affected by “unexplained infertility” (UI). This controversial diagnosis is made when no abnormalities of the female and/or male reproductive systems are clearly identified. In my experience this is not true. When standard female and male examination and testing is performed, less than 10% of the couples do not show infertility criteria (detailed personal and family story, detailed body examination, spermiogram, testicular Doppler ultrasound, serologies with cervix and male urethral swabs for Chlamydomonas and Mycoplasma, hemogram with blood groups, biochemical status including major immunology and thrombophilia factors, hormones, karyotypes, endovaginal ultrasound, hysterosonography).	We agree that the prevalence of "Unexplained Infertility" is highly variable, depending on different epidemiological data and, basically, on the diagnostic workout performed. After analysing percentages provided by ASRM and ACOG, we modified the sentence into "about 30%". Thank you
Jean Calleja-Agius	5	77	Fallopian – not with a capital letter	This was adapted as suggested.
Jean Calleja-Agius	5	78	Add time frame – 1 year	The time frame is not part of the ICMART definition.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy	6	Line 110-114	Suggest: To provide clinicians and infertile couples….	Due to the medical terminology and contents, the guideline is for clinicians. A patient leaflet will be available on the ESHRE website

Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				
Carlos Calhaz-Jorge	6	112	“correctly confirm the diagnosis of UI”. As UI is an exclusion diagnosis maybe “correctly identify couples with UI” could be more exact.	"confirm" diagnosis was replaced by "establish"
Liliana Ramos	6	122	The same sentence is written twice, delete one of them (the studied population in these sections is couples with unexplained infertility specifically)	The sentence is providing information about the population studied in two different chapters.
Liliana Ramos	6	131	The term MAR refers to “Medical Assisted” Reproduction (add Medical assisted)	This was adapted as suggested.
Jean Calleja-Agius	6	132	Reproduction – small letter	This was adapted as suggested.
Mol BW	6	142	….important outcomes in this guideline are ……live full-term singleton birth . What is wrong with live full-term twins? The problem with twins is harm from preterm birth; so either want to establish singleton clinical pregnancies…. full-term twins are not a main problem for someone with infertility.	Live birth in general, as well as multiple pregnancies/multiple births are listed among the critical outcomes (together with the singleton birth) without any negative connotation.
Mol BW	6	142	An important outcome would be knowledge on the causes of the infertility: knowing for the sake of knowing. This is a classic reference: Asch DA, PattonJP, Hershey JC. Knowing for the sake of knowing: the value of prognostic information Med Decis Making 1990 Jan-Mar;10:7-57. doi: 10.1177/0272989X9001000108.	This is included in the concept of patient satisfaction and quality of life (among the important outcomes). The last chapter reviews the value in terms of QoL of knowing the cause of infertility for both men and women. Furthermore, in the diagnostic PICO questions, the reliability/accuracy of each test was included in the outcomes
Jean Calleja-Agius	6	145	I think you can remove the word ‘value’	The choice of terminology here was extensively discussed by the GDG. In the end, the term "benefit" to the couples was

				preferred over simple "effectiveness" as it was deemed more appropriate and inclusive of different aspects
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	6	Line 145	The guideline focuses on outcomes of relevance, accuracy, acceptability, reliability, feasibility, value (in terms of cost-benefit ratio) for the diagnostic tools (page 6). What do you mean with value (in terms of cost-effectiveness)?	The choice of terminology here was extensively discussed by the GDG. In the end, the term "benefit" to the couples was preferred over simple "effectiveness" as it was deemed more appropriate and inclusive of different aspects
Jean Calleja-Agius	7	148	add fullstop	This was adapted as suggested.
METHODOLOGY				
Priya Bhide	/	/	The methodology for this guideline (literature search, data collection, data analysis, forest plots) is unavailable and should be included	This information is included in the annexes of the guideline, which will be published on the ESHRE website together with the guideline.
I. DEFINITION				
Aboubakr Mohamed Elnashar	/	/	Definition of unexplained infertility is required. What are basic investigations? Age to be considered or not?	All these questions are covered by the guideline.
Carlos Calhaz-Jorge	8	R1	I suggest to remove “at least”	The recommendation has been removed for reasons unrelated to this comment.
Unexplained Infertility Guideline Australian Adaptation Committee	8	R 1	The group highlight the recommendation to have sexual intercourse every 2-3 days in the first half of the cycle. We recommend saying the first 21 days as cycle length may vary. It would be helpful to define a regular menstrual cycle	The recommendation has been removed for reasons unrelated to this comment. Thank you for your suggestion. The definition of normal menstrual cycle is out of the scope of this specific section.

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	12	Line 161-162	The important question "AFTER HOW MANY MONTHS OF UNPROTECTED INTERCOURSE SHOULD A COUPLE BE DEFINED AS INFERTILE?" is not approached in an empiric way. For example: In 12 months 80% of the couples are pregnant, in 24 months 90% of the couples are pregnant; so 50% of the remaining couples who are not pregnant in one year become pregnant between 12 en 24 months . WHO definition of infertility is changed from 12 months till 24 months see ref. Ref;European Journal of Obstetrics & Gynecology and Reproductive Biology 214 (2017) 204–208 .Ever growing demand for in vitro fertilization despite stable biological fertility—A European paradox	Of course we acknowledge that a number of these couples will conceive between 12-24 months, but here we are referring to the initiation of investigations. According to prognostic indicators, which have been well documented, the decision as to when to initiate treatment is taken.
George Lainas	12	168	Why in the absence of evidence make any reccomandation frequency of sexual intercourse? Better avoid formulating a frequency and timepoint oriented recommendation. In addition, while the narrative question refers on whether the frequency of intercourse will affect the diagnosis of UI, the recommendation as formulated, provides sexual intercourse frequency instructions.	The recommendation has been removed.
Jackson Kirkman-Brown Meurig Gallagher	12	169	The evidence supports a minimum of once every three days – there is no evidence to suggest twice daily is a problem so the answer should be reframed to reflect daily-3 days. Couples should equally not be concerned that daily is 'too frequent' which has been neglected here.	Lines 179-182 have addressed the issue of more frequent intercourse. However, for further clarity, lines 182-184 have been amended.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	12	169	Recommendation – the background text does not support the clear recommendation considering the frequency (every 2-3 days) and time frame (whole first half of the cycle) and does not correspond to preferences and possible cause of stress through recommendation. Suggesting to add as written in the background text: "Couples with UI are advised to have sexual intercourse "at least" every 2-3 days "prior to the fertile days in the menstrual cycle to the extent that such suits their own preferences" OR "acknowledging that this can lead to stress in individual couples""	The recommendation has been removed.
Christina Bergh Jan Bosteels Frank Broekmans	12	184	Couples with UI are advised to have sexual intercourse every 2-3 days at least in the first half of the cycle.	The recommendation has been removed.

Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely			This is a treatment advise and should be positioned at that part of the guideline. Please reformulate the definition.	
Mario Sousa	12	185	According to the International Committee for Monitoring Assisted Reproductive Technology (ICMART) definition of infertility, couples should have at least 12 months of regular, unprotected sexual intercourse before fertility interventions may be initiated. The WHO definition applies here. I just add that this is for females < 35y, being 6m if ≥35y	The GDG could not find any justification to diagnose UI after 6 months if female age >35.
Lars Björndahl			There are clear indications, that the WHO reference limits (2021) are based on a very mixed population. As early as 1968 Tietze showed that most couples succeeded in starting a pregnancy within 3-4 months after discontinuing use of contraceptives. Quite recently it was also shown that men in couples achieving a pregnancy rapidly showed much higher semen examination results compared to the WHO reference limits. It can therefore be argued that the 12 month period before starting an infertility investigation and treatment is reasonable, but not to base "normal reference limits" from results from men in couples who have struggles to start a pregnancy for more than 4 months!	That's very interesting information but we cannot really incorporate it into the guidelines.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn			We miss an advise on when to start fertility workup. After a year of unprotected regular intercourse and regular cycle?	This is indeed the advice, based on the ICMART definition. This is also stated in the guideline.
Jackson Kirkman-Brown Meurig Gallagher	13	188-196	It would be desirable to also reflect that male age is known to have an effect within this text – people tend to think of the 'Rod Stewart' celebrity with young partner and neglect that both male and female age increasingly impact as both age. Otherwise this is potentially disproportionally stigmatizing to women.	The sentence "To a much lesser extent and at more extreme ages, male age could affect fertility potential" was added to the text.

Mira Töyli	13	196	I would like the age related decline in fertility to be discussed a bit further. Since diagnosis of UI is to rule out factors causing infertility, there are no tests or markers for poor egg quality. As stated in the draft wrong positive is 10% in women under 35 years and 80% in women over 40. I would like the GDG discuss that can UI be used in women over 40? And how much of subfertility is caused by advanced female age and poor egg quality in age group 35-40 and not by UI.	As female age is the best indicator of egg quality and prognosis for pregnancy, the GDG has drawn the line for the definition of UI at 40 years old. Under this age ovarian insufficiency is much less likely.
Sabine Kliesch	13	198	i) The definition of unexplained infertility in this guideline mentions normal sperm count as prerequisite. Two aspects have to be considered here: 1st in male infertility, the lack of identified etiologic male (and female) factors may lead to the diagnose of unexplained - (no pathologic findings) or idiopathic (pathologic findings in basic semen analysis) male infertility. The common aspect being the lack of identified etiologic factors. If the guideline group considers normozoospermic men to be included only, this would neglect a large proportion of infertile men, and would thus be not representative. 2nd the reduction of impaired fertility to the WHO definition of semen results and thus mainly on the number, motility and morphology of spermatozoa is far too short. Male fertility involves many aspects more such as sexual problems (erectile dysfunction/premature ejaculation), infections of the male genital tract, chromosomal disorders on the male side that are not reflected by sperm count (such as balanced translocations), sperm dysfunction e.g. due to sperm-channeldysfunction and DNA damage at the sperm level as well as hormonal dysfunctions such as pituitary, hypothalamic or testicular disorders. The guideline summarizes andrological aspects of unexplained infertility in an unfortunate and absolutely incomplete, if not incompetent fashion, telling the reader that no andrological examination or intervention is required as long as the sperm quality is normal. No reflection on repeated semen values, no reflection on the phenotyping of the male by clinical aspects are taken into consideration or reflected by the selected manuscripts. In principle, the situation of unexplained infertility and primarily normal semen parameters is actually where andrological aspects start and do not end. Fertility and infertility is a couple issue and male aspects of that go far beyond sperm analysis. Aspects of diagnostics and treatment in men have been unfortunately ignored in this guideline.	We thank Sabine Kliesch for the detailed appraisal. We agree that there are several male factors that are contributory towards infertility. This guideline is specifically for UI and the GDG adhered to the definition by ICMART (2017), hence the various male factor related causes for infertility are not within the scope of this guideline. It has been acknowledged that the potential for the diagnosis of UI is dependent upon the methodologies used and/or those methodologies available (lines 79-90).
Jackson Kirkman-Brown Meurig Gallagher	13	198-205	We strongly commend the team on this decision and support it.	Thank you.
Unexplained Infertility Guideline Australian	13	198	The definition deals largely with female factors and it is presumed male erectile problems or libido issues would exclude the diagnosis from being made?	A paragraph was added to the definition section with regards to the importance of clinical history taking.

Adaptation Committee				
Jean Calleja-Agius	13	207-209	Rephrase as a sentence	This was adjusted in the text.
II. DIAGNOSIS				
Marco Sbracia			The recommendation on this issue is really poor and should completely re-written.	It is unclear to the GDG which issue the reviewer is referring to. Therefore, the GDG is unable to follow-up on this.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn			What is unexplained subfertility, how is it diagnosed?	The definition of unexplained infertility is covered in chapter one, diagnosis is discussed in chapter two.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn			We miss an advise on when to start fertility workup. After a year of unprotected regular intercourse and regular cycle?	According to the ICMART definition of infertility, fertility work-up may be initiation after 12 months of regular, unprotected intercourse. To clarify this, a sentence was added to the definition section.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis			In addition to the above. There is a whole list of tests that are recommended not to be required in the context of unexplained infertility. As an unwanted consequence, this suggest that these tests are justified in other settings. We would like to ask to provide more literature on where these tests are useful. Alternatively, the suggestions on what not to do can be omitted.	Other settings where these tests can be useful is considered outside the scope of the guideline.

Lan Vuong Madelon van Wely				
II.1 OVULATION				
Joel Bernstein	7	R2	Point 2. Confirmation of ovulation https://www-bmj-com.rcog.idm.oclc.org/content/bmj/321/7271/1259.full.pdf While this is a rather old article I believe that the recommendation not to routinely test for ovulation should be amended as timing using fertile period calculations or apps is often inadequate. This would also require removing point #3 1 Infertile couples have usually been trying to conceive for some time and are stressed 2 Intercourse often does not occur as frequently as they report 3 Mentally, timed intercourse (with LH surge monitoring) provides them with a plan of action 4 It can be used in between more aggressive forms of treatment IUI OI or IVF provided Fallopian tubes patent and uterus normal 5 Especially cost effective where female age >40 6 Some patients do not wish to proceed with more “invasive treatment”	Ovulation testing can indeed be performed by couples and centres facilities performing MAR to pinpoint ovulation timing prior to treatment procedures such as timed intercourse, IUI and natural cycle FET. However, precise ovulation timing was not part of the scope of this PICO question, which was solely focused on ascertaining the occurrence of ovulation (for diagnostic purposes when attempting to define whether the cause of infertility is unexplained or not), without attempting to pinpoint the exact timing.
Unexplained Infertility Guideline Australian Adaptation Committee	8	R2	It would be helpful to define a regular menstrual cycle ie 26-35. There is a lot of published data on the cycle length	Thank you for your suggestion. The definition of normal menstrual cycle is out of the scope of this specific PICO question.
Unexplained Infertility Guideline Australian Adaptation Committee	8	R3	Do you mean there is no need for repeated luteal phase tests?	Such is actually addressed in the recommendation 2, which states "In women with regular menstrual cycles, tests for confirmation of ovulation are not routinely recommended".
Jean Calleja-Agius	14	235	Rephrasing of the PICO question: What is a reliable and convenient measure to confirm regular ovulation?	Despite acknowledging that the suggested wording changes seem minor and well-intended (i.e. to make the reading clearer), the data retrieval and synthesis was based on the original PICO question and changing it after such is generally not recommended.
George Lainas	15	235	I doubt if ‘’confirmation of ovulation’’ is a valid term, regarding the role of urinary LH measurements, ultrasound monitoring or mid-luteal progesterone measurement. The role of these methods is probably related to programming the timing of intercourse	The data synthesis showed in multiple instances the application of these strategies outside of the context of timed intercourse,

				namely for confirmation of ovulation (the studies assessed were described in the appendix). Moreover, specifically for midluteal progesterone measurement, this assessment generally occurs at a timing in which timed intercourse would no longer be recommended.
Hunida Elmegrab	15	295	About confirmation of ovulation ultrasound, urinary LH and with estrogen level at time of ovulation to confirm normal ovulation	It is true that consecutive serum estrogen/progesterone/LH and ultrasound assessments are routinely performed in many facilities performing MAR to pinpoint ovulation timing prior to treatment procedures such as timed intercourse, IUI and natural cycle FET. However, precise ovulation timing was not part of the scope of this PICO question, which was solely focused on ascertaining the occurrence of ovulation, without attempting to pinpoint the exact timing. When actual timing is not considered, estrogen measurements have not been, to our knowledge, assessed as an instrument to confirm regular ovulation.
Hunida Elmegrab	17	305	We should explain to the doctor and to the patient what is normal menstrual cycle? because there are mis understanding at this point	Thank you for your suggestion. The definition of normal menstrual cycle is out of the scope of this specific PICO question.
George Lainas	16	288	No data on ultrasound evidence are provided.	In all relevant studies, ultrasound was used as the reference test.
Aboubakr Mohamed Elnashar	17	305	Regular menstrual cycles should be defined, all instigations are based on it	Thank you for your suggestion. The definition of normal menstrual cycle is out of the scope of this specific PICO question.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	17	306	Studies for UI and use of fertility awareness-based methods are lacking but the benefit especially under cost-effectiveness analysis is proven in couples with infertility. The recommendation of exclusive suggestion of LH-tests, ultrasound or progesterone is not supported by the quoted studies and clinical practical recommendations. Suggestion for Recommendation to add “methods as”:	Following your suggestion, the recommendation has been amended.

	In women with regular menstrual cycles, if confirmation of ovulation is warranted, “methods as” urinary LH measurements, ultrasound monitoring or mid-luteal progesterone measurement can be used. Thijssen A, Meier A, Panis K, Ombelet W. 'Fertility Awareness-Based Methods' and subfertility: a systematic review. Facts Views Vis Obgyn. 2014;6(3):113-23. PMID: 25374654; PMCID: PMC4216977. Najmabadi S, Schliep KC, Simonsen SE, Porucznik CA, Egger MJ, Stanford JB. Cervical mucus patterns and the fertile window in women without known subfertility: a pooled analysis of three cohorts. Hum Reprod. 2021 Jun 18;36(7):1784-1795. doi: 10.1093/humrep/deab049. PMID: 33990841; PMCID: PMC8487651. Stanford JB, Willis SK, Hatch EE, Rothman KJ, Wise LA. Fecundability in relation to use of mobile computing apps to track the menstrual cycle. Hum Reprod. 2020 Oct 1;35(10):2245-2252. doi: 10.1093/humrep/deaa176. PMID: 32910202; PMCID: PMC7518709. Frank-Herrmann P, Jacobs C, Jenetzky E, Gnoth C, Pyper C, Baur S, Freundl G, Goeckenjan M, Strowitzki T. Natural conception rates in subfertile couples following fertility awareness training. Arch Gynecol Obstet. 2017 Apr;295(4):1015-1024. doi: 10.1007/s00404-017-4294-z. Epub 2017 Feb 9. PMID: 28185073.	
II.2 OOCYTE/CORPUS LUTEUM QUALITY		
Pratip Chakraborty	Labelling the criteria of unexplained infertility (UI) as “unexplained” is heterogeneous. Hence, the treatment part is often empirical. More often, recurrent pregnancy loss is overlapped with UI. (Chakraborty P., Banerjee S, Saha P, Nandi SS, Sharma S, Goswami SK, Chakravarty BN, Kabir SN (2013) Aspirin and low-molecular weight heparin combination therapy effectively prevents recurrent miscarriage in hyperhomocysteinemic women PLoS One 8(9):e74155). Recently in a preprint version my lab documented oxidative stress and inflammatory-apoptosis cross talk in hyperhomocysteinemic rat/s. Being hyperhomocysteinemia a crucial factor for potentiating or activating different issues discussed in the guideline/s, like, oocyte quality, (Chakraborty P. Yasmin S, Chattopadhyay R, Goswami SK, Chakravarty BN. The effects of maternal hyperhomocysteinemia on embryo quality in mice. Hum.Reprod. 29, 177-178 (2014)) tubal factor, ovarian reserve (Kalapahar S, Sharma S, Chattopadhyay R, Parvin S, Behera S, Ghosh S, Chakraborty P, Chakravorty BN. Diabetic but not women with normal metabolic phenotype with unexplained infertility are in risk for decreased ovarian response: A prospesctive population based cohort study Hum.Reprod.35 i1419 (2020)) I request to include the hyperhomocysteinemia and diabetes	The suggested references seem interesting, however, animal studies and conference abstracts are not eligible to be included in the body of evidence of the guideline.

			entity in the issue/s of oocyte quality and ovarian reserve. This is particularly important since this guideline focus a global platform.	
Ulrich A. Knuth Michael Ludwig	17	342	The draft guideline on the approach to unexplained infertility (UI) by the Unexplained Infertility Guideline Group 2022 seems problematic because the definition of UI refers solely to the duration of unprotected intercourse with regular male findings. This is based on a normal cycle without defining the criteria for a normal cycle. While a cycle of 21 to 35 days is generally considered normal, no statement is made as to whether the individual cycle length must remain the same over multiple consecutive cycles. Clarification would be all the more important, as the guideline does not recommend further diagnostic confirmation by hormone tests in the case of regular cycles. Hormone measurements would thus only be indicated if cycle irregularities were present. Many years of clinical experience speak against such an approach, even if the evidence in the evaluated studies does not seem to be sufficient. Our comment: "Regular menstruation" is used synonymously for "eumenorrhea" respectively; we agree with the statements that one does not need luteal phase control by progesterone measurement, if there is Eumenorrhea. That means eumenorrhea is present if the cycle is (a) stable (without premenstrual spotting), (b) regular (with few fluctuations, ± 2 days), and (c) inconspicuous (within normal limits ≥ 24 days, ≤ 35 days), and (d) with normal bleeding pattern (maximum 7 days). If this is not the case, causes of the follicle maturation disorder need to be clarified.	The definition provided is one of a "normal" menstrual cycle. So the GDG does not think a modification is necessary.
Ulrich A. Knuth Michael Ludwig	17	342	The draft guideline on the approach to unexplained infertility (UI) by the Unexplained Infertility Guideline Group 2022 seems problematic because the definition of UI refers solely to the duration of unprotected intercourse with regular male findings. This is based on a normal cycle without defining the criteria for a normal cycle. While a cycle of 21 to 35 days is generally considered normal, no statement is made as to whether the individual cycle length must remain the same over multiple consecutive cycles. Clarification would be all the more important, as the guideline does not recommend further diagnostic confirmation by hormone tests in the case of regular cycles. Hormone measurements would thus only be indicated if cycle irregularities were present. Many years of clinical experience speak against such an approach, even if the evidence in the evaluated studies does not seem to be sufficient. Our comment: "Regular menstruation" is used synonymously for "eumenorrhea" respectively; we agree with the statements that one does not need luteal phase control by progesterone measurement, if there is Eumenorrhea. That means eumenorrhea is present if the cycle is	The definition provided is one of a "normal" menstrual cycle. So the GDG does not think a modification is necessary.

			(a) stable (without premenstrual spotting), (b) regular (with few fluctuations, ± 2 days), and (c) inconspicuous (within normal limits ≥ 24 days, ≤ 35 days), and (d) with normal bleeding pattern (maximum 7 days). If this is not the case, causes of the follicle maturation disorder need to be clarified.	
II.3 OVARIAN RESERVE				
Unexplained Infertility Guideline Australian Adaptation Committee	8	R6	While AMH may not predict fertility, it may set a warning that time is limited for the woman and more active treatment is needed. Women with a low AMH may ovulate early and embryo-uterine synchrony may be disturbed. This could be incorporated into a good clinical practice point or not recommend it at all here. The strong recommendation was queried, and the technical report needs to be examined	We see your point regarding women with low ovarian reserve may be candidates for active management sooner. Yet, the recommendation regards identification of the aetiology of infertility or predicting spontaneous pregnancy in women with regular cycles, i.e. ovulatory and unexplained infertility. While women with severely decreased ovarian reserve may ovulate earlier, this would show as shortened menstrual cycles. We are unaware of evidence suggesting embryo - endometrium asynchrony due to decreased ovarian reserve in natural cycles, and fail to see how to address this in an evidence based manner.
Mahmoud A Abdel-Aleem	21	410	The link between age and unexplained infertility particularly in the section of ovarian reserve" deserves discussion.	While ageing and quantitative ovarian reserve are related, quantitative ovarian reserve status is not related to fecundity in women with regular cycles (i.e., unexplained infertility), ageing is more relevant for decreased fecundity due to increased embryo aneuploidy rate which cannot be assessed with ovarian reserve tests.
Carlos Calhaz-Jorge	23	511	"… compared cycle 2-4 AFC in 148…". This section is about FSH and estradiol, not AFC. Can you check, please?	This was corrected in the text
Carlos Calhaz-Jorge	24	531	Units are missing. I guess "cubic centimeters" is needed	This was corrected in the text
Ulrich A. Knuth Michael Ludwig	24	539	We also agree that indeed an assessment of ovarian reserve (estradiol, FSH, cycle day 3-5) is not necessary to clarify the cause of subfertility. However, it is useful to check in case of an unfulfilled desire to conceive (> 1 year) in order to strategically decide whether to	Thank you for sharing your opinion. The current evidence consistently suggest that ovarian reserve status, by any assessment

			proceed rather earlier actively or not. The chances of conception are not different depending on the AMH level. Determination of AMH in primary diagnostics is insofar dispensable and should only be performed if the FSH tone is conspicuously high for the respective age of the infertile patient."	method, either FSH or AMH, is irrelevant for choosing the next step in management of unexplained infertility, which by definition involves a regularly ovulating woman.
Mario Sousa	24	539	In women with regular menstrual cycles, ovarian reserve testing is not required to identify the aetiology of infertility or to predict the probability of spontaneous conception over 6 to 12 months. In my general evaluation of the female, besides a full endocrinological screening including Tyrode and AMH, the endovaginal ultrasound is mandatory. In this analysis, the ovary is also fully evaluated regarding surface, structure, dimensions, volume, and number of antral follicles	Thank you for sharing your opinion. The current evidence consistently suggest that ovarian reserve status, by any assessment method, either FSH or AMH, is irrelevant for choosing the next step in management of unexplained infertility, which by definition involves a regularly ovulating woman.
II.4 TUBAL FACTOR				
Joel Bernstein	8	/	Points 7 to 13 Essentially the uterus and tubes both need to be tested so why not recommend a single test i.e.HyCoSy which usually includes tubal patency and 3 D Ultrasound? Ideally this could be preceded by a Chlamydia antibody screening to provide antibiotic cover if needed. Unfortunately today many specialists are only requesting a transvaginal ultrasound as a test of utero-tubal function.	Thank you for this comment, this was clarified in the justification.
Maruf Siddiqui	8	/	Laparoscopy has an advantsge over HyCoSy and HSG in suspected PID or in countries with high prevalence rate of PID. Adhesiolysis and correction of tubo ovarian relatioship leading to better outcome with OI/IUI following Laparoscopy in unexplained infertility.	If there is suspicion of adhesions this is not UI any longer.
Unexplained Infertility Guideline Australian Adaptation Committee	8	R 7 and 8	This should be merged with recommendation 47 where the therapeutic value of oil-based contrast media is noted. Why should a patient have a water-based test for diagnosis when an oil-based test would provide equivalent diagnosis with proven efficacy? The pros and cons of this approach should be discussed. It was felt the words “not recommended” should be replaced by “not essential” to indicate there may be clinical needs and patient desires to visualise the abdomen and pelvis.	The justification was amended to discuss the pros and cons of HSG and HyCoSy more extensively. Furthermore, a reference to the treatment part on tubal flushing was added.
Unexplained Infertility Guideline Australian Adaptation Committee	8	R 7-10	Clarification is sought on what visual demonstration means. Is this laparoscopy? Visual could also mean seeing an Xray or ultrasound.	The GDG did not want to restrict the recommendation as this depends on the history and other evaluations and available methods.
Marco Sbracia	8	R9	May be the chlamydia antibody testing considered an alternative non-invasive way to test tubal patency? This test is an no specific test and my not reveal most of cases with tubal obstruction, such as endometriosis or other infections. It should be considere as second	Please see the recommendation that is "conditional" also followed by the GPP below.

			line exams to determine the cause of tubal problems, and no screening test for it. This point should be amended or deleted (better!)	
Mahmoud A Abdel-Aleem	26	641	is there a role for tubal endoscopy techniques in detecting subtle tubal abnormalities? E.g. salpingoscope, fertiloscope..	These techniques were not part of the PICO question.
Petya Andreeva	27		1. Serology test (testing of Chlamydia antibody) has little, if any, value in testing for genital C. trachomatis infection. It should not be used for screening because previous chlamydial infection might or might not elicit a systemic antibody response. 2. Not only Chlamydia trachomatis, but infections with Ureaplasma spp, Mycoplasma spp., and anaerobic bacteria should be taken in mind as a reason for tubal factor.	The swab/PCR tests is for an active C. trachomatis infection (STD), not for tubal patency testing. The other bacteria mentioned haven't been shown to cause infertility (still under research) and therefore outside the scope of this question.
Kalmantis Konstantinos	27	644	Throughout the "tubal factor" category the committee has not mentioned HyFoSy as a diagnostic method for tubal patency. There are a number of studies that compare the diagnostic accuracy of this method, and is the only method whose results do not present a significant difference from the results of laparoscopy with dye. Therefore, HyFoSy is a beneficial method for both women and practitioners, especially with 3D-HDF-HyFoSy which provides high percentages for sensitivity, specificity, negative and positive predictive value.	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.
Panayotidis Costas	27	644	The committee needs in my opinion to review the difference of HYFOSY to HYCOSY or to make clear if HYFOSY can be considered a variation of HYCOSY or not. In my opinion it is different kind of screening test and a description should be added with a particular paragraph In the "tubal factor" category, on page 27, I notice that the HyFoSy method is not mentioned, and in general, Ultrasound Salpingography is poorly supported in the literature, primarily from Chinese studies using non-intended echogenic media. Since 2011, 49 studies have been published, mainly European, that mention the HyFoSy method. Of these studies, 4 have been published in Human Reproduction and the last one was published on May 3, 2022 with the title "Can hysterosalpingo-foam sonography replace hysterosalpingography as first-choice tubal patency test? A randomized non-inferiority trial." (Van Welie N et al, Hum Reprod. 2022 May 3;37(5):969-979). Of these 49 studies have been included for the writing of the guidelines only 2, one concerning pain levels on examination, and showing no difference in levels between HyFoSy and HyCoSy (Boned-López J et al, Arch Gynecol Obstet. 2021 Dec;304(6):1389-1398) and the other comparing the results of two-dimensional and three/four-dimensional ultrasound imaging using an echogenic medium, with three/four-dimensional imaging yielding better results (Alcázar JL et al, Hum Fertil (Camb). 2022 Feb;25(1):43-55).	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.

			Therefore, it is useful to consider that the HyFoSy method seems even superior in sensitivity and specificity to aqueous sodium chloride imaging and is equally valid as HSG (without the radiation, iodine exposure and with significantly lower pain levels). Or to reformulate and say that the HYFOSY is a new technique for the moment the evidence is …. Etc the results of the published articles show better results or as good as…. but further evidence is needed . But not to avoid entirely in the whole guideline to mention HYFOSY.	
Michael Grynberg	27	644	Authors completely bypass the use of HyFoSy with Foam. The only registered medium for tubal patency using sonography. The contrast media mentioned in chapter II.4 are either off label use (sonovue) of off the market (echovist)	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.
Michael Grynberg	28	Table 1	Literature mentioned is outdated. More recent, European studies discussing more accurated procedure (hyFoSy and approved medium (ExEm® Foam) are not mentioned.	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.
Michael Grynberg	28	Table 1	Chen 2019: off label use of Sonovue, with multiple side effects	A sentence has been added to the justification.
Michael Grynberg	28	Table 1	Cimen 1999: use of echovist, no longer available on the market.	A sentence has been added to the justification.
Michael Grynberg	28	Table 1	Liang 2019: off label use of Sonovue, with multiple side effects	A sentence has been added to the justification.
Michael Grynberg	28	Table 1	Radic 2005: use of echovist, no longer available on the market.	A sentence has been added to the justification.
Michael Grynberg	28	Table 1	Shahid 2005: use of echovist, no longer available on the market.	A sentence has been added to the justification.
Michael Grynberg	28	Table 1	Zhou 2012: off label use of Sonovue, with multiple side effects	A sentence has been added to the justification.
Exalto N. Emanuel MH.	32	642	The description of available tubal patency tests is incomplete. There are two worldwide used ultrasound techniques: HyFoSy (using foam) and HyCoSy (using saline). Both deserve to be mentioned.	This has been clarified in the justification.
Exalto N. Emanuel MH.	32	642	Tubal factor The subheading PICO Question: What is the accuracy of commonly used tests of tubal patency is promising. However, HyFoSy is not mentioned in the manuscript and Foam only once on page 32 in relation to pain perception during the procedure (Boned-Lopez et al., 2021). The PICO search terms are not mentioned. The formulation The GDG cannot formulate a recommendation on the use of contrast mediums or saline due to too little studies may be caused by only using HyCoSy as a surge term instead of using both HyCoSy and HyFoSy. The search term HyFoSy reveals 41 references in PubMed.	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.

In a comparison between HyCoSy and HyFoSy Piccioni (2017) concluded that HyFoSy allows a more accurate diagnosis of tubal patency compared with HyCoSy. In the same year Ludwin published in Human Reproduction on the accuracy of HyFoSy in comparison to HyCoSy and Laparoscopy. Exalto and Emanuel summarised all available literature on clinical aspects in 2019.
Grigovich (2021) summarized the advantages and disadvantages of HyCoSy and HyFoSy very precisely for radiologists in the USA. Ramos (2021) and Melcer (2021) are in favour of HyFoSy. Also, Engels (2021) and Zajicek (2022) published recently prospective studies on HyFoSy.
In a large multicentre study (Van Welie 2022) it was concluded that HyFoSy leads to similar pregnancy outcomes compared to HSG and is less painful.
Ramos (2022) stated that current evidence suggests that HyFoSy has emerged as a new option to make Fallopian tube assessment easier. HyFoSy should be the first-line diagnostic procedure to assess tubal patency.
Studies on pregnancy after HyFoSy, although small, showed promising results comparable to these after other tests. Bohîlțea (2022) recently reported a 37% pregnancy rate within 6 month after the procedure in a group of 672 infertile women undergoing HyFoSy
The above-mentioned literature is illustrating that HyFoSy is in all aspects, including the number of pregnancies after the procedure, comparable or even better than HyCoSy and in some (like pain and stability of the foam) superior to HyCoSy with saline. This technique therefore deserves to be mentioned in the guideline.
References
Bohîlțea RE, Mihai BM, Stănică CD, Gheorghe CM, Berceanu C, Dima V, Bohîlțea AT, Neagu S, Vlădăreanu R. Technical Tips and Tricks after 10 Years of HyFoSy for Tubal Patency Testing. J Clin Med. 2022; 11: 5946: 1-14
Engels V, Medina M, Antolín E, Ros C, Amaro A, De-Guirior C, Manzour N, Sotillo L, De la Cuesta R, Rodríguez R, San-Frutos L, Peralta S, Martin-Martínez A, Alcázar JL. Feasibility, tolerability, and safety of hysterosalpingo-foam sonography (hyfosy). multicenter, prospective Spanish study. J Gynecol Obstet Hum Reprod. 2021: 50; 102004; 1-6
Exalto N, Emanuel MH. Clinical aspects of HyFoSy as tubal patency test in subfertility workup. BioMed Res Int 2019; 4827376: 1-12
Grigovich M, Kacharia VS, Bharwani N, Hemingway A, Mijatovic V, Rodgers SK. Evaluating Fallopian Tube Patency: What the Radiologist Needs to Know. Radiographics. 2021; 41: 1876-18961
Ludwin I, Ludwin A, Wiechec M, Nocum A, Banas T, Basta P, Pitynski K. Accuracy of hysterosalpingo-foam sonography in comparison to hysterosalpingo-contrast sonography with air/saline and to laparoscopy with dye. Human Reprod 2017; 32:758-769
Piccioni MG, Riganelli L, Filippi V, Fuggetta E, Colagiovanni V, Imperiale L, Caccetta J, Panici PB, Porpora MG. Sonohysterography: comparison of Foam and Saline. J Clin Ultrasound. 2017; 45: 67-71
Ramos J, Pellicer N, Fernández-Sánchez M. Hysterosalpingography is obsolete: hysterosalpingo-contrast foam sonography should be the alternative. Reprod Biomed Online. 2022; 45: 839-842
Van Welie N et al. Can hysterosalpingo-foam sonography replace hysterosalpingography as first-choice tubal patency test? A randomized non-inferiority trial. Huam Reprod 2022: 37: 969-979

			Zajicek M, Kassif E, Weisz B, Berkovitz Shperling R, Lipitz S, Weissbach T, Barzilay E, Orvieto R, Haas J. "One-stop shop" for the evaluation of the infertile patient: hystero-salpingo foam sonography combined with two and three dimensional ultrasound and sonohysterography. J Obstet Gynaecol. 2022: 42; 670-674	
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	32	699	Add laparoscopy as comparator as shown in table 1+2 HSG and HyCoSy are comparable in diagnostic capacity "to laparoscopy with chromopertubation for tubal patency testing", thus selection of the technique used is up to the preference of the clinician and the patient.	The recommendation was adapted as suggested.
Michael Grynberg	32	699	"HyCoSy and HSG are comparable in diagnostic accuracy", however pain scores are lower (Serrano González 2022, van Welie 2022) for HyCoSy/HyFoSy and no radiation and iodine are used in those methodes,	This has been clarified in the justification.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	32	699	"HSG and HyCoSy are comparable in diagnostic capacity, thus selection of the technique used is up to the preference of the clinician and the patient." What about the therapeutic effect of tubal flushing? While tubal tests have a diagnostic and a therapeutic character, recommendations on its use should consider and integrate both dimensions.	Tubal flushing is covered in the treatment section. A reference to the treatment section has been added to the justification.
Christophe Blockeel	32	699	HyFoSy is not discussed. Literature in table I shows several studies with outdated contrast medium (no longer on market), does not show data of foam.	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.
Christophe Blockeel	32	699	"preference is up to clinician and patient" in ESHRE/ESGE guidelines of 2016 (Grimbizis 2016) a preference for ultrasound is given over HSG.	The ESHRE/ESGE guidelines from 2016 is on the diagnosis of congenital malformations, not tubal patency.

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	32	Line 707-17	Please mention HyFoSy To access the uterine cavity and fallopian tubes, Hysterosalpingo-Contrast Sonography (HyCoSy), using echogenic medium (commercial products or saline with air) or Hysterosalpingo-Foam Sonography (HyFoSy), using foam, can be performed.	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.
Panayotidis Costas	32	713	For the moment exists a " contrast" medium which is specifically destined for intrauterine injection for HYFOSY technique and holds a legal license to test tubal patency in Europe. Mentioning the technique does not mean that you promote a commercial product (and you can clarify this as well) but it is unfair to omit entirely the significance of the HYFOSY technique. Finally, it is the only ultrasound method whose results in 3D HDF mode are comparable to those of laparoscopy (Devine et al, Fertility and sterility 2022, Vol. 118, Issue 1, pp. 19–28.). Regarding the lack of evidence that GDG cannot formulate a recommendation on the use of contrast medium or saline due to too little studies I add here evidence - references in terms HYFOSY / HYCOSY with HSG or laparoscopy • Ludwin et al 2017, Accuracy of hysterosalpingo-foam sonography in comparison to hysterosalpingo-contrast sonography with air/saline ant to laparoscopy with dye., Hum Reprod. 2017 Apr1;32(4):758-769 • -Piccioni et al 2017, Sonohysterosalpingography: Comparison of foam and saline solution., J Clin Ultrasound. 2017 Feb;45(2):67-71 • -Riganelli et al 2018, Ultrasonography reappraisal of tubal patency in assisted reproduction technology patients: comparison between 2D and 3D-sonohysterosalpingography. A pilot study., Minerva Ginecol. 2018 Apr;70(2):123-128 • -Ludwin et al 2019, Inter-Rater Reliability of Air/Saline HyCoSy, HyFoSy and HyFoSy	Studies including the comparison HyFoSy and laparoscopy and dye were included in the guideline.

			Combined With Power Doppler for Screening Tubal Patency., Ultraschall Med. 2019 Feb;40(1):47-54	
Panayotidis Costas	32	714	Regarding patient tolerance and safety useful references: van Welie et al 2022, Can hysterosalpingo-foam sonography replace hysterosalpingography as first-choice tubal patency test? A randomized non-inferiority trial. , Hum Reprod. 2022 May 3;37(5):969-979 -Ramos et al 2022, Hysterosalpingography is obsolete: hysterosalpingo-contrast foam sonography should be the alternative., Reprod Biomed Online. 2022 Jun 1:S1472-6483(22)00399	The systematic review by Boned-Lopez included in the guideline includes 29 studies on patient tolerance.
Michael Grynberg	32	715	Although no recommendation can be given due to low numbers of studies, only off the market and off label contrast medium is mentioned in table 1 and none of the more recent papers studying Foam (Ludwin 2017, Piccioni 2017, Riganelli 2018) are mentioned	Ludwin 2017 and Piccioni 2017 are included in the SR by Alcazar et al., 2020. Riganelli 2018 did not provide raw data to calculate sensitivity/specificity/PPV and NPV.
Christophe Blockeel	32	715	There are no references for the use of foam, please add to table 1 van Schoubroeck 2013, Ludwin 2017, Piccioni 2017, Riganelli 2018, Situmorang 2020	Ludwin 2017, van Schoubroek 2013 and Piccioni 2017 are included in the SR by Alcazar et al., 2020. Riganelli 2018 did not provide raw data to calculate sensitivity/specificity/PPV and NPV. Situmorang 2020 was published in a journal not indexed for Pubmed.
Christophe Blockeel	32	718	HSG: disadvantages of HSG are not mentioned. This is described in the guidelines of ESHRE/ESGE guidelines of 2016 (Grimbizis 2016): “Its disadvantages include painful, risk of infection and irradiation of the patient. It is more invasive than ultrasound, not always easy and needing radiological unit.”	This was clarified in the justification.
Ulrich A. Knuth Michael Ludwig			We do not agree with the comments on tubal factor and uterine factor and would always favour a diagnostic LSK and HSK with chromopertubation over other methods in a fertility patient to be sure that no pathology is missed. This is especially true in older age of a fertility patient, in order not to deprive her of a therapeutically valuable IVF	Thank you. If there is high suspicion (symptoms or other history) that there is a need for laparoscopy, the guideline does not prevent this. However, routine examinations do not have evidence to recommend them.
Michael Grynberg	33	729	Not all advantages are mentioned HyCoSy/ HyFoSy can also be used for “all-in-one”infertility checkup (Zajicek 2021, Levaillant 2019) and have lower pain scores. They use no radiation and Iodine.	This has been clarified in the justification.

Petya Andreeva	33	735	- Urogenital infection with C. trachomatis in women should be diagnosed by vaginal or cervical swabs. - The performance of nucleic acid amplification tests (NAATs) with respect to sensitivity and specificity, is better than any of the other tests available for the diagnosis of chlamydial infection. NAATs are the most sensitive tests for these specimens and are the recommended test for detecting C. trachomatis infection. - Serology test (testing of Chlamydia antibody) has little, if any, value in testing for genital C. trachomatis infection. It should not be used for screening because previous chlamydial infection might or might not elicit a systemic antibody response. References: 1. Recommendations for the Laboratory-Based Detection of Chlamydia trachomatis and Neisseria gonorrhoeae — 2014. Recommendations and Reports. March 14, 2014 / 63(RR02);1-19 https://www.cdc.gov/mmwr/preview/mmwrhtml/rr6302a1.htm#:~:text=Recommendations%20for%20the,RR02)%3B1%2D19 2. Zhou, Ying et al.Performance of point-of-care tests for the detection of chlamydia trachomatis infections: A systematic review and meta-analysis. eClinicalMedicine, 2021, Volume 37, 100961; DOI: https://doi.org/10.1016/j.eclinm.2021.100961 3. US Preventive Services Task Force, Davidson KW, Barry MJ, et al. Screening for Chlamydia and Gonorrhea: US Preventive Services Task Force Recommendation Statement. JAMA. 2021;326(10):949-956. doi:10.1001/jama.2021.14081	The swab/PCR tests is for an active C. trachomatis infection (STD), not tubal patency testing.
Carlos Calhaz-Jorge	33	736	I suggest to remove the first word “Serum”	This was adapted as suggested by the reviewer.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol	33	756	Chlamydia antibody testing for tubal patency could be considered as a non-invasive alternative. While we agree that in women without a history of STD, Pelvic surgery or Appendicitis, Chlamydia antibody testing could be an alternative for immediate HSG and HYCOSY, we think sensitivity of Chlamydia antibody testing is not good enough to allow it as a replacement for visual tubal testing with HSG/HYCOSY. So Chlamydia antibody testing can have a place in the work-up, but the formal diagnosis ‘unexplained infertility’ requires visual tubal testing with HSG/HYCOSY or laparoscopy. Also, consider the therapeutic effect of tubal flushing?	Please see the evidence that is "conditional" also followed by the GPP below.

Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				
Mario Sousa	33	756	Chlamydia antibody testing for tubal patency could be considered as a non-invasive alternative. In my general evaluation I perform urethral swab in males and cervix swab in females. I have cases cured of infertility just because patients were treated with the adequate antibiotics after this diagnosis. These patients do not have tubal obstruction but silent endometritis.	Thank you for this comment. Endometritis is outside the scope of this guideline.
Aboubakr Mohamed Elnashar	33	756	Chlamydia antibody testing is a good negative test & not an alternative to tubal patency tests	Please see the evidence that is "conditional" also followed by the GPP below.
Aboubakr Mohamed Elnashar	33	757	Laparoscopy to replace visual demonstration of tubal patency	The GDG did not want to restrict the recommendation as this depends on the history and other evaluations and available methods.
Carlos Calhaz-Jorge	34	761-762	I suggest to add the text highlighted: "… systematic review, showed CAT is a clinically usable non-invasive tubal…". That CAT is a non-invasive tubal patency test was already known. Its reliability is the concern.	This was adapted as suggested by the reviewer.
Carlos Calhaz-Jorge	34	772-773	The Hubacher paper used "past pelvic inflammatory disease symptoms, previous history of a lower genital tract infection, previous vaginal discharge, and antibodies to Chlamydia trachomatis" in the definition of "high-risk medical history". I suggest to make it clear in the text.	This information was added to the text.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	35	Table 3	Is that the relevant question? Chlamydia can cause functional disorders that can also explain infertility in open fallopian tubes. The comparison with laparoscopy therefore misses the point. The question should be "can fertility be predicted with a chlamydia test" in unexplained infertility. This examination would not be an alternative to tubal patency testing but a supplement, especially in the case of open fallopian tubes.	Thank you for this suggestion, we will keep it in mind for the update of the guideline.
II.5 UTERINE FACTOR				
Monica Varma			Does a diagnosis of chronic endometritis by endometrial biopsy change the management? Is it required to be ruled out	Thank you for this. If there is chronic endometritis, that is no longer considered UI.
Unexplained Infertility Guideline Australian	8-9	Recommendation 13	Some of the group disagreed with this because ultrasound quality is very variable even in high-class healthcare settings. Concern was expressed that chronic endometritis will be	We agree that some cases of chronic endometritis may be missed. But at the moment there is no evidence that routine

Adaptation Committee			missed. Others also felt the recommendation was too prescriptive in the light of the subsequent recommendation to not do a hysteroscopy.	hysteroscopy would improve pregnancy rate. When the evidence is there we are happy to change the recommendation.
Mahmoud A Abdel-Aleem	39	927	The value of HSG in the diagnosis of uterine factor (Asherman syndrome or mullerian anomalies) deserve mentioning	HSG is already included in the text.
Carlos Calhaz-Jorge	39	946	It should be: "…98% sensitivity…, 100% specificity…"	This was corrected in the text.
Maria Elisabetta Coccia	39	948	Considering the study could be written I suggest : Ultrasound preferably 3D, Is recommended to exclude uterine anomales and confirmed also in unexplained infertility. generally is considered the gold standard techniques from recurrent pregnancy loss to infertility	Thank you. The recommendation is always on UI. We did not look for evidence outside UI.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	39	948	"Ultrasound, preferably 3D, is recommended to exclude uterine anomalies in women with unexplained infertility." should be "To establish a diagnosis unexplained infertility, uterine anomalies should be excluded with Ultrasound, preferably 3D". Please cite the evidence that 3D is superior to 2D ultrasound for this indication. Is this addition necessary considered in the light of low income countries?	As indicated in the introduction, the first part of the diagnosis section concerns "patients under investigation for infertility", therefore there is no need to revise the recommendation. The evidence to support the recommendation can be found in the paragraph above the recommendation. The GDG formulated the recommendation to take into consideration that not every fertility clinic has access to an ultrasound machine with 3D- modality.
Carlos Calhaz-Jorge	41	994	The Almog paper refers to saline instillation sonography (SIS), not hystero-contrast sonography (HyCoSY), a technique in which a specific type of contrasts is used. It would be better to use only SIS in this paragraph.	This was corrected in the text.
Carlos Calhaz-Jorge	41	994-995	The sentence is not clear. Could you please, revise it?	This was amended in the text.
Ziller V., Goeckenjahn M., Köhn F.-M..,	41	1001	"At present, there is no high-quality evidence to support the routine use of hysteroscopy as a screening tool in the general population of subfertile women with a normal uterine cavity on ultrasound or hysterosalpingogram in the basic fertility work-up to improve	Thank you for this comment. Given that there is no strong evidence to support further evaluation, we also need to restrict

Hancke K. , Sonntag B.			reproductive success rates. In women undergoing in vitro fertilisation (IVF), low-quality evidence from all studies reporting these outcomes, suggests that performing a screening hysteroscopy before IVF may increase live birth and clinical pregnancy rates." How do GDP get to the conclusion not to recommend hysteroscopy based on this reference?? Low level of evidence should not be mistaken for a strong recommendation. How about adhesions and chronic endometritis – things that would not be detected. As the population of unexplained is not the general population we recommend to for less restrictive suggestions – as did e.g. Kamath et al.	unnecessary workup that are expensive and time consuming for the women. We would need to build up the evidence to recommend further evaluation. Also, it has to be remembered that the recommendation is for UI. If there is a reason to suspect Asherman, that is not UI any longer and of course further investigations has to be carried out.
II.6 LAPAROSCOPY				
Gustavo Botti	/	/	I think is useful to roule out endometiosis or adhesions	There is no justification to perform a diagnostic laparoscopy in all infertile women. The GDG has suggested in the justification to reserve this test for specific patients.
Gustavo Botti	/	/	I do not believe that any treatment should be indicated until the diagnostic stage is completed with a laparoscopy if necessary.	There is insufficient evidence to suggest that clinically relevant diagnoses will be missed by omitting a laparoscopy in patients at low risk for tubal pathology.
Nusrat Mahmud			I am working in a country where MRI and Ultrasound facilities are present but can't give the minute details. Considering laparoscopy in cases of UI gives additional information, like mild endometriosis/ pelvic adhesions and can be managing simultaneously.	Evidence to advise every infertile women because this is clinically relevant is missing and therefore routine diagnostic laparoscopy in women at low risk for tubal pathology is not advised.
Nusrat Mahmud			I would suggest routine laparoscopy and hysteroscopy should be considered in UI if the facilities permits and depending on surgeons skill rather than not mention as NOT RECOMENDED	The GDG does not agree. The evidence for the usefulness and clinical relevance of a routine laparoscopy is not present. It is therefore not recommended
Ulrich A. Knuth Michael Ludwig			Although the recommendations speak of routine laparoscopic diagnostics in UI, there is obviously a circular argument here, since the diagnosis can probably only be made with an inconspicuous laparoscopy. If the guideline is adopted in this way, private insurance companies in Germany may take the view that it is not indicated in relevant cases and will refuse to cover the costs. On the other hand, coverage of costs for an IVF measure would be refused with the argument that UI exists if no pathological constellation of physiology or anatomy have been proven and the patient is actually healthy.	The indications for considering a diagnostic laparoscopy are mentioned in the justification.

Unexplained Infertility Guideline Australian Adaptation Committee	9	R14	See above for concerns about not doing a laparoscopy	The GDG has discussed this suggestion, however, has decided not to adapt the recommendation. The recommendation is about "routine" laparoscopy, not laparoscopy on indication.
Joel Bernstein	9	R14	Should possibly change this to routine hysteroscopy and laparoscopy?	Thank you for the comment. This section was specifically concerning laparoscopy. Hysteroscopy is discussed in the previous chapter.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	43	1071	Same as above, recommendation should be less restrictive. The aim should be to explain as many unexplained as possible as this would change further management. As mentioned in the cited references many findings as mild endometriosis would switch patients from UI to other diagnosis. or known endometriosis – should be “suspected”	The GDG has discussed this suggestion, however, has decided not to adapt the recommendation. The recommendation is about "routine" laparoscopy, not laparoscopy on indication.
Mario Sousa	43	1071	Laparoscopy can be useful in cases with a clinical suspicion of endometriosis	Thank you for your comment, this has been added to the text.
Maria Elisabetta Coccia	43	1071	I suggest to add “in infertile women at low risk for tubal pathology”	The GDG has discussed this suggestion, however, has decided not to adapt the recommendation. The recommendation is about "routine" laparoscopy, not laparoscopy on indication.
Gustavo Botti	44	1084	A patient cannot be labeled as UI without ruling out pelvic pathology with a laparoscopy. HSG does not have sufficient sensitivity or specificity to diagnose endometriosis or pelvic adhesions	The GDG agrees that HSG will miss mild endometriosis and subtle lesions. However evidence is lacking to justify routine laparoscopy for every patient as is elaborated upon in the justification.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	44	1091	add history of PID, previous ectopic pregnancy or “clinically suspected and” known endometriosis	Thank you for your comment, this has been added to the text.
II.7 CERVICAL/VAGINAL FACTOR				
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau	45	1106	Important PCT references are lacking ; Steures https://pubmed.ncbi.nlm.nih.gov/17561002/ https://pubmed.ncbi.nlm.nih.gov/17482611/ https://pubmed.ncbi.nlm.nih.gov/16997935/	The study by Steures et al. compared OS+IUI with IUI in a natural cycle, not PCT in two different groups. Van der Steeg et al.

Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				formulated a prediction model, where PCT was just one of the parameters.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	45	1115	Oei et al 1998 included women with all types of infertility, including a substantial number of women with PCOS.	The GDG checked the included population in the study of Oei et al., 1998. No PCOS patients were included.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	45	1116	In a retrospective cohort study, including 2476 patients with UI, the long-term overall pregnancy rates were compared after a positive or a negative PCT ~~were compared~~.	This was corrected in the text.
Bulent Tandogan	46	1140	Although mycoplasma hominis is mentioned about vaginal microbiota, Ureaplasma Urealyticum, which can cause serious infection and infertility, is not mentioned. I think it would be good to include this cause of infection in the guide.	The GDG did not make a selection in this section, all relevant evidence according to the PICO question on vaginal microbiota in

				patients with unexplained infertility was included.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	46	1172	Vaginal microbiota testing could be considered in couples with unexplained infertility only in a research setting. Why this statement; everything is allowed in a research setting.	Vaginal microbiota is an emerging research topic in infertility. Current literature shows that there are differences between fertile and infertile patients, and the GDG would like to encourage further research on this topic.
II.8 MALE GENITO-URINARY ANATOMY				
Liliana Ramos	/	/	II.8 and II.9: many of the referred papers so not specify males had a status of normospermia. Especially when DNA damage is high, motility is very much affected. I do support the final recommendation nr. 19 For recommendation nr. 20 it should also be “not recommended when WHO semen analysis is normal” (delete “probably”)	The recommendation was adapted as suggested
Lars Björndahl			“WHO semen analysis is normal” – this is ambiguous. The reference limits for semen examination provided by WHO have been rightfully critized for giving the impression that there are exact and distinct limits between fertility and male infertility. It is therefore a huge problem that these guidelines refer so uncritically refer to these reference limits. Furthermore, the guideline is even more ambiguous – “when WHO semen analysis is normal”- exactly which characteristics are supposed to be “normal”. A longstanding misinterpretation is the use of sperm concentration in spite of the fact that the WHO recommendation is to primarily use total sperm number (millions per ejaculate) due to that sperm concentration is to a high degree dependent on dilution of spermatozoa with secretions from other sources than the testicles and epididymides. It appears that the guidelines uncritically sustains the general lack of understanding of interpretation of human semen examination results.	A sentence was added to the definition chapter specifying when further investigations are necessary.

Unexplained Infertility Guideline Australian Adaptation Committee	9	R 17	A qualification is sought to say the word “consistently” should be added ie one normal WHO result is not enough	A sentence was added to the introduction specifying that one semen analysis from a lab with external quality control is enough for diagnostics.
Sabine Kliesch	48	1221	The chapter refers to the results of the EAA ultrasonography study which sets up reference values for genitourinary ultrasound in fertile men with proven spontaneous pregnancy induction and shows the association between ultrasound findings and semen analysis. The study is not powered to give information on an added value for sonography in men with unexplained infertility. Thus, the evidence suggested here and the recommendation are not valid.	The GDG found no direct evidence to support the sentence, only indirect evidence from fertile healthy men.
Lars Björndahl	48	1221-1233	Have the references been scrutinized according to e.g. Standards in semen examination: publishing reproducible and reliable data based on high-quality methodology. Hum Reprod, 2022.DOI: 10.1093/humrep/deac189? Is for instance a MAR test of ≥1% relevant? How was vas size measured?	GDG acknowledges the long-standing issue with the quality of provided information on used methodology in studies based on results from basic semen examination (Björndahl et al., 2022; Vasconcelos et al., 2022). GDG agrees with the need to develop strategies to address standardisation in reporting the results of a semen analysis for publication.
Liliana Ramos	48	1231	I think the value of >=1% is not correct. Do you mean 51%?	The value of ≥1% is as reported in the study.
Mario Sousa	48	1234	Testicular imaging is not recommended when WHO semen analysis is normal. In cases of infertility without a clinical diagnosis, I perform testicular imaging. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari	48	1234	Add to recommendation “and having undergone physical examination without abnormalities”	The GDG considered your comment but decided that there is no need for a normal physical examination when the results of the semen analysis according to WHO criteria is normal.

Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				
Lars Björndahl	48	1234	The Recommendation is formulated different from the PICO question that is much wider that just “Testicular imaging”	The reviewer has a point. The formulation of the key question sounds broader than what is actually addressed, however, the PICO terms were ultrasound in addition to physical examination.
Carlos Calhaz-Jorge	48	1237-1238	No evidence is presented that supports this sentence. Of course, I agree with it but it would be good to have a reference.	The GDG found no direct evidence to support the sentence, only indirect evidence from fertile healthy men.
II.9 MALE ADDITIONAL TESTS				
Dimitrios G. Goulis Giovanni Corona			The establishment of an etiological diagnosis is central in any medical approach. The diagnosis of an “unexplained” or “idiopathic” disease is, by definition, a diagnosis of exclusion. The present guideline seems to adopt an approach of “the diagnosis of unexplained infertility requires normal sperm count” and “no andrological examination or intervention is required as long as the sperm quality is normal”; therefore, the contribution of male factors in the couple with “unexplained” infertility seems to be almost null. We cannot agree with this oversimplistic approach. If no attempt to establish an etiological diagnosis is made, many potentially dangerous conditions will be missed (indicatively, but not exclusively: balanced translocations, sexual dysfunctions, male accessory gland infections, sperm dysfunctions, testicular cancer, metabolic diseases) with obvious consequences for the reproductive and general health of the man. Therefore, a basic diagnostic approach (history, clinical examination, hormonal and metabolic profile evaluation, series of spermiograms, testicular imaging) is justified and evidence-based. Additional tests can be ordered according to the findings of the basic approach.	A paragraph has been added to the definition section to discuss the need for a semen analysis according to WHO criteria in a laboratory with external quality control and clinical history taking.
Dimitrios G. Goulis Giovanni Corona			Many statements in the guideline are based on evidence derived from studies of other populations than men with “unexplained” infertility. A short list would include: • DNA fragmentation test (men with infertility)	For chapter II.8, only indirect evidence from fertile healthy men was identified with the literature search. The studies included in the ASA and DNA fragmentation section

	• Anti-sperm antibodies (men with infertility and positive antibodies) • EAA ultrasonography study (normal men)	have been checked, and one study has been excluded in both sections.
Dimitrios G. Goulis Giovanni Corona	On the contrary, there is some evidence derived from populations of men with “unexplained” infertility, which is not discussed in the guideline.	To the best of our knowledge, we have included all evidence from studies complying with the definition of unexplained infertility by ICMART
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	The definition of unexplained infertility includes "normal sperm quality". On the one hand, this is understandable, but it reduces the concept of "normality" to the WHO definition of sperm quality and thus mainly to the number, motility and morphology. In fact, the recommendations of the guidelines can be summarized as follows: as long as the sperm quality is "normal", there is no need for any further andrological examination. This is in contrast to some statements of the 6th edition of the WHO laboratory manual for the examination and processing of human semen. At the beginning of chapter 4 (advanced examinations; seminal oxidative stress and reactive oxygen, assessment of the acrosome reaction, assessment of sperm chromatin, transmembrane ion flux and transport in sperm, computer-aided sperm analysis (casa)) WHO clearly indicates that “normal sperm parameters” do not exclude infertility: “However, in many patients, the infertility is rather due to a dysfunction of the spermatozoa, where ejaculate examination yields parameters that appear to be completely normal.” In addition, another comment can be found in chapter 3 “extended examinations” (multiple sperm defects, sperm DNA fragmentation, genetic tests, immunological methods, marker of male genital tract inflammation, antibody coating of spermatozoa, biochemical assays for accessory sex gland function). The statement of WHO (“The tests described in this chapter are not necessary for routine semen analysis but may be useful in certain circumstances for diagnostic or research purposes.”) indicates, that WHO is aware, that more tests than standard semen analysis may be helpful to examine male fertility/infertility. In order to explain the discussions and ambiguities surrounding the WHO manual in more detail, it must be clear that there is fundamentally a decisive difference in the question, at which value a pregnancy can still occur and, on the other hand, how likely the occurrence of a pregnancy will be in the future. In the WHO "norm curve", all participants had achieved a pregnancy. From this point of view, it can only be deduced to what extent the sperm values are distributed in a sample that is capable of conceiving. A statement about the probability of achieving a pregnancy in the future or how long the time to pregnancy might be cannot be derived from this "norm curve" design.	A sentence was added to the definition chapter specifying when further investigations are necessary.

	Reducing male factor diagnosis to regular sperm analysis, as no further exams will follow, this will leave our patients “unexplained” even knowing that within WHO ranges subfertility is present. Looking at the studies listed, some of them show that, despite normal sperm quality, individual functional parameters are not within the reference ranges in couples with unexplained infertility. Concerning the male factor, this guideline is not helpful or even senseless, as it does not provide any active instructions for action. It would have been more helpful to recommend extended or advanced sperm or semen examinations at least in the context of research (as it is recommended for measurement of oxidative stress). Our opinion is that extended sperm function diagnostics can of course explain situations that were previously unexplained, even in the case of "normal sperm quality". Due to the rigidity of the guideline, no further development in this area can be expected in the future.	
Ulrich A. Knuth Michael Ludwig	On the male side, it is not taken into account that the current WHO manual in the 6th edition does not specify any standard values and only describes percentiles in a group of males achieving a pregnancy within a year after first semen analysis. A clear statement is missing defining the ranges of pathological findings in semen analysis. As in the previous WHO manual 5, the following values could be classified as normal: 1. Sperm count: there should be at least 15 million sperm per millilitre of ejaculate. 2. Motility: at least 40% of the spermatozoa should be able to move progressively. 3. Shape: At least 4% of the sperm should have a normal shape. 4. Volume: the volume of the ejaculate should be at least 1.5 millilitres. 5. pH: The pH of the ejaculate should be between 7.2 and 7.8. In addition, it must be required that these values are not undercut in any examination during the period of infertility treatment. Since the current draft makes many further examinations (DNA fragmentation, hormone examination, microbiological clarification) dependent on the "normality" of the spermtest, a different procedure would result from a different definition of normal ejaculate.	A paragraph has been added to the definition section to clarify this.
Maria Schubert	There is a wide variety of male factors that may contribute to impaired fertility status. The guideline summarizes andrological aspects of unexplained infertility in an unfortunate fashion, telling the reader that no andrological examination or intervention is required as long as the sperm quality is normal. There are many studies that show, that	The GDG employed the definition of unexplained infertility as defined by ICMART: "apparently normal testicular function, genito-urinary anatomy and a

			normozoospermia itself is not enough to evaluate fertility status. It is rather sperm function that needs to be addressed. Normozoospermia is not where andrologic workup ends, but rather where it starts, searching for etiologic factors (for example Single Nucleotide Polymorphisms (SNPs)). Further, the definition of unexplained infertility in this guideline mentions normal sperm count as prerequisite. In male infertility, the lack of identified etiologic male (and female) factors may lead to the diagnose of unexplained - (no pathologic findings) or idiopathic (pathologic findings in basic semen analysis) male infertility. The common aspect being the lack of identified etiologic factors. If the guideline group considers normozoospermic men to be included only, this would neglect a large proportion of infertile men, and would thus be not representative. The share of clinical andrological contribution by experts in the field is very unfortunate to the share of experts in female infertility. This is also reflected in the disproportion of chapters dealing with female vs. male diagnostics when this is known to affect the couple similiarily. To the future reader this leads to the assumption that the major focus in couple infertility is to be laid on the female side. This neglects that the identification of etiologic factors on the male side, by further and thorough diagnostics, beyond basic semen analysis, may reduce the therapeutic burden on the female side.	normal ejaculate". The composition of the GDG was multidisciplinary, including experts in the field of reproductive endocrinology, reproductive surgery, andrology, safety and quality in ART, a nurse and a patient representative. The GDG found no evidence that in men with unexplained infertility and normal semen analysis, FSH levels are influenced, having repercussions on reproductive outcomes. However, a sentence was added to the justification about the emerging evidence with regards to polymorphisms in the FSH-B-gene. The GDG has included a paragraph in Annex 3 highlighting the urgent necessity of further research to address gaps in the understanding of male infertility, such as identifying new aetiological causes.
Unexplained Infertility Guideline Australian Adaptation Committee	9	R 22	Some endocrinologists felt where the very low but normal range WHO semen analysis was present, hormones should be measured. Occasional pituitary disease can manifest as low normal semen analysis. Maybe added “consistently” to the statement	The GDG found no evidence that in men with unexplained infertility and normal semen analysis, reproductive hormones levels are influenced, having repercussions on reproductive outcomes.
Unexplained Infertility Guideline Australian Adaptation Committee	9	R 18-24	The semen analysis section has a lot of “not recommended” and could be reordered into one more encompassing guideline point.	The guideline recommendations are ordered in the order of the clinical questions that were addressed.
Hunida Elmegrab	50		Its better to include normal sperm function test with normal semen analysis to diagnose unexplained infertility in male	The GDG understands this comment as a suggestion to include tests to assess normal sperm function in men with UI, but such tests are not clinically and soundly validated. The GDG hopes that ongoing

				research in male infertility diagnosis will provide such tests in the future.
Maria Schubert	50	1253	The cited studies with relevant patient samples clearly show a higher proportion of INFERTILE men with POSITIVE anti-sperm antibodies compared to fertile men. This study strengthens the relevance of anti-sperm antibodies in case of unexplained infertility. The cohort studies including methods of assisted reproduction do not show significant differences between anti-sperm antibody positive or negative men. The conclusion, not to test for anti-sperm antibodies in unexplained infertile men, is not given by the studies selected. The studies merely give rise to the assumption, that assisted reproductive techniques may overcome the negative effect of anti-sperm antibodies.	The overall quality of the evidence was low and the heterogeneity within studies was high, which does not allow recommending ASA testing. High heterogeneity in the studies is caused by confounding factors, study designs and quality of methodology, outcomes analysed, study size. Therefore, the true effect of testing for ASA cannot be conclusive and GDG cannot formulate a recommendation in favour of ASA testing.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	50	1253	Clarify semen anti-sperm antibody detection versus II.10 Page 58 Line 1546	Anti-sperm antibodies in section II.9 are in semen and in section II.10 in serum as also specified in the text.
Sabine Kliesch	50	1254	The cited studies with relevant patient samples clearly show a higher proportion of INFERTILE men with POSITIVE anti-sperm antibodies compared to fertile men. This study strengthens the relevance of anti-sperm antibodies in case of unexplained infertility. The cohort studies including methods of assisted reproduction do not show significant differences between anti-sperm antibody positive or negative men. The conclusion, not to test for anti-sperm antibodies in unexplained infertile men, is not given by the studies selected. The studies merely give rise to the assumption, that assisted reproductive techniques may overcome the negative effect of anti-sperm antibodies.	The overall quality of the evidence was low and the heterogeneity within studies was high, which does not allow recommending ASA testing. High heterogeneity in the studies is caused by confounding factors, study designs and quality of methodology, outcomes analysed, study size. Therefore, the true effect of testing for ASA cannot be conclusive and GDG cannot formulate a recommendation in favour of ASA testing.
Carlos Calhaz-Jorge	50	1258	I suggest to add the text highlighted in yellow: "(MAR)≥50% in 15.6% (166/1060) vs 1.9% (2/107)) and…"	This was adapted in the text as suggested.
Carlos Calhaz-Jorge	50	1288	"was not significant" in this line should be removed	This was corrected in the text.
Sabine Kliesch	51		DNA fragmentation test show differences in the outcome of MAR. The studies quoted do not reflect the population of men with unexplained infertility. Thus the recommendation is not valid.	Due to the low quality of the evidence and the significant heterogeneity of the available studies following systematic literature search, the GDG cannot

				recommend in favour of routine DNA fragmentation testing.
Jackson Kirkman-Brown Meurig Gallagher	51-52	1312-1335	We would have liked to see a clearer statement – as is made in the WHO manual – that sperm DNA damage tests are in fact a heterogenous group of totally different tests that must not be read as the same or interchangeable and thereby evidence for one cannot be read as evidence supporting use of another. This helps ensure that if one test claims new evidence patients are not exploited by a different, not evidence-based, test being claimed to have parity. We believe this is very important to avoid patient exploitation and change current practice, the way these are approached as one test does not help.	This has been clarified in the justification.
Verena Nordhoff	52	1335	We agree, that SDF tests have limited capacity to discriminate between couples who would benefit from the test for allocation to a specific MAR technique. However it might be relevant in individual cases and could draw a complete picture of the DNA-status of a men´s sperm.	This is considered outside the scope of this guideline.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	52	1335	"Testing for sperm DNA fragmentation is not recommended when WHO semen analysis is normal." Testing for sperm DNA fragmentation is not needed to establish the diagnosis UI". Some of us state that they are unaware of any indication for sperm DNA fragmentation. In fact, by mentioning that tests like sperm DNA fragmentation are not recommended for unexplained infertility, one suggests that they might be of value elsewhere. Please provide the references for that statement.	As specified in the introduction, the patient population in this section is couples with unexplained infertility specifically, hence the recommendation that this test is not recommended in couples with unexplained infertility.
Hunida Elmegrab	52	1335	From our observational study even with WHO normal semen analysis still we have male with positive DNA fragmentation test which after treatment pregnancy occurs spontaneously .	Current literature is controversial on the association between conventional semen parameters and DNA fragmentation. Several meta-analyses have shown that different SDF assays have different predictive accuracy for pregnancy and each assay had a different predictive value for

				IVF and ICSI (Cissen et al., 2016, Zhao et al., 2014). Individual experience and practice, while valuable, is not supported by the literature in this instance.
Mario Sousa	52	1335	Testing for sperm DNA fragmentation is not recommended when WHO semen analysis is normal. In cases of infertility without a clinical diagnosis, I perform sperm TUNEL analysis. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	53	1367	Sperm chromatin condensation test is probably not recommended when WHO semen analysis is normal. No evidence change to is “not recommended”	The recommendation has been adapted as suggested.
Mario Sousa	53	1367	Sperm chromatin condensation test is probably not recommended when WHO semen analysis is normal. In cases of infertility without a clinical diagnosis, I perform sperm Aniline blue (histones) analysis. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Liliana Ramos	53	1367	As no studies support any evidence for the use of Sperm chromatin condensation test, and having this parameter a very strong correlation with DNA fragmentation tests, the “probably” should be taken from the recommendation and just conclude that is not recommended.	The recommendation has been adapted as suggested.
Sabine Kliesch	54		There have been several studies been published recently with focus on unexplained and idiopathic male infertility and FSH. None of these studies is cited, none of the relevant findings with low FSH in case of variants in the FSH-B-Gene is reflected. Of course, there is	The GDG found no evidence that in men with unexplained infertility and normal semen analysis, FSH levels are influenced,

			good evidence to analyse hormones, especially FSH and testosterone in men with normal semen analysis, as normal sperm count does not necessarily reflect normal sperm function that may be influenced by FSH and / or testosterone. Thus the recommendation is not valid.	having repercussions on reproductive outcomes.
Jean Calleja-Agius	54	1399	Remove 'male infertility' to read as follows: Azoospermia is the aetiological category....	This was corrected in the text.
Maria Schubert	54	1412	There have been several studies been published recently with focus on unexplained and idiopathic male infertility and FSH. None of these studies is cited, none of the relevant findings with low FSH in case of variants in the FSH-B-Gene is reflected. Of course, there is good evidence to analyse hormones, especially FSH and testosterone in men with normal semen analysis, as normal sperm count does not necessarily reflect normal sperm function that may be influenced by FSH and / or testosterone. Thus the recommendation is not valid.	The GDG found no evidence that in men with unexplained infertility and normal semen analysis, FSH levels are influenced, having repercussions on reproductive outcomes. However, a sentence was added to the justification about the emerging evidence with regards to polymorphisms in the FSH-B-gene.
Sabine Kliesch	55		There is poor evidence for microbiology testing in infertile couples including those with unexplained infertility. However, with clinical signs of leucocytospermia microbiology testing is recommended to explore the potential risk for sperm dysfunction, sperm transportation and infectious signs during pregnancy. The passage does not reflect studies in this field and thus the recommendation is not valid.	The need to test for leucocytospermia was discussed by the GDG, and it was decided that this is reflective of a sperm abnormality and therefore is not unexplained infertility/this is a transient condition, unlikely to "cause" UI.
Maria Schubert	55	1445	There is poor evidence for microbiology testing in infertile couples including those with unexplained infertility. However, with clinical signs of leucocytospermia microbiology testing is recommended to explore the potential risk for sperm dysfunction, sperm transportation and infectious signs during pregnancy. The passage does not reflect studies in this field and thus the recommendation is not valid.	The need to test for leucocytospermia was discussed by the GDG, and it was decided that this is reflective of a sperm abnormality and therefore is not unexplained infertility/this is a transient condition, unlikely to "cause" UI.
Mario Sousa	65	1811	Sperm aneuploidy screening is not recommended when WHO semen analysis is normal. In cases of infertility without a clinical diagnosis, I perform sperm aneuploidy screening. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
II.10 ADDITIONAL TESTS FOR SYSTEMIC CONDITIONS				
Mario Sousa	9	25	Anti-sperm antibodies should be performed. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Unexplained Infertility Guideline Australian	10	R 26	The word "should" might be better than "can" -suggest check technical report on Grade recommendation	The GDG formulated a conditional recommendation, and the wording "can be considered" is in line with conditional

Adaptation Committee				recommendations according to the GRADE methodology.
Marco Sbracia	9	R27	In patients with suspected unexplained infertility testing for antithyroid antibodies should be performed since in several countries autoimmune thyroid diseases are frequent also with normal levels of TSH and FT4 and FT3, such as reported in the last guideline edition of European and British Thyroid associations. Consequently at least one test for these autoantibodies should be suggested, considering the test for coeliac disease in these guidelines is reported to be considered.	The aim of this guideline is to compare diagnostic approaches in unexplained and explained infertility rather than plan for treatment approaches. This does not prevent the clinician doing other tests depending on their circumstances and interests but the GDG does not encourage this as a diagnostic process.
Mario Sousa	9	R27	I always perform a basal immunological analysis, as the reason is to full evaluate non visible causes, as for me there is always a cause: Coombs (direct and indirect) Circulating immune complexes C3, C4, CH50 Rheumatoid Factor Lupus antibody ANA ENA ANCA Anti-Thyroglobulin Anti-Thyroid Antibodies Anti-Peroxidase Anti-Thyroid Antibodies Anti-phospholipid antibodies (IgG, IgM) Anti-cardiolipin antibodies (IgG, IgM) Anti-beta2-glycoprotein I antibodies (IgG, IgM)	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Mario Sousa	9	R28	I perform a basal thrombophilia analysis, as the reason is to full evaluate non visible causes, as for me there is always a cause: Functional antithrombin III Prothrombin gene mutations (Factor II) Methylene-Tetra-Hydro-Folate Reductase (MTHFR) gene mutations Factor V Leiden gene mutations Plasminogen activation inhibitor (PAI) gene mutations FATHER 1 Resistance to activated protein C (Factor V Leiden) (APCR ratio) Fibrinogen (D dimers) Fibrinogen	Individual experience and practice, while valuable, is not supported by the literature in this instance.

			Plasminogen Functional protein C Functional protein S Free protein S	
Mario Sousa	10	R31	I perform as routine a Karyotype analysis. Depending on the results of IVF, I also study the genetic background in cases of failed fertilization, failed embryo development, failed implantation. In cases where chronic respiratory symptoms are present, I study PCD gene mutations	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 31	It was felt this was dependent on the overall definition of unexplained infertility. If there had been miscarriages before, karyotype/genetic tests might be indicated.	The guideline covers unexplained infertility. RPL is considered outside the scope of this guideline.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 32	The vitamin D issue is controversial and may vary from country to country. Perhaps some clarification is needed in the wording.	The recommendation was adapted to "Testing for vitamin D deficiency in females is not recommended for diagnosis of unexplained infertility"
Marco Sbracia	10	R33	The same observation previously done, TSH in the normal range does not mean no thyroid problems (see last edition of British and European thyroid association). Furthermore, anti-thyroid antibodies may negatively impact reproduction especially in case of IVF.	The aim of this guideline is to compare diagnostic approaches in unexplained and explained infertility rather than plan for treatment approaches. We were unable to find differences in anti-bodies, except for coeliac disease.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 33 and 34	The order of these might be better if 34 came before 33. 27 could then follow these or be incorporated into the others.	The GDG agreed to change the order of the recommendations.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 35	It depends on the menstrual cycle regularity and definition of “normal”. If the cycle is variable, prolactin was felt to be important.	According to the ICMART definition of unexplained infertility, the menstrual cycle is regular.

Mario Sousa	10	35	PRL belongs to the routine endocrinological approach	There is no evidence that prolactin levels are higher in unexplained infertility, while accepting that prolactin secreting tumours may rarely have normal cycles.
Carlos Calhaz-Jorge	59	1583	I suggest to add the text highlighted in yellow: "…with medical therapy and anti-thyroid antibodies (n=15)."	This was added to the text.
Carlos Calhaz-Jorge	59	1590	Just a proposal: "…, parous women) ~~investigated~~ thyroid disfunction and auto-immunity in infertility were investigated."	This was adjusted in the text.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	59	1601	Testing for thyroid antibody and other autoimmune conditions (apart from coeliac disease) in women with unexplained infertility is probably not recommended. Why probably?	The recommendation was adapted as suggested.
Carlos Calhaz-Jorge	59	1601 - Recommendation	Does this recommendation imply that coeliac disease should be routinely screened? Evidence in lines 1574-77 shows a very minimal clinical relevance.	The recommendation states "it can be considered". Lines 1571-1573 cover a systematic review which reported stronger data.
Carlos Calhaz-Jorge	60	1642	I suggest to add the text highlighted: "…case-control study, 230 women with UI and 240 fertile…"	This was added to the text.
Mahmoud A Abdel-Aleem	61	1001	Definition of normal uterus should be specified including a definition of normal endometrium in relation to different phase of menstrual cycle and thickness of the myometrium and appearance of the cervix and uterine serosa.	The GDG chose not to do this given the variation between clinics, sonographic skills, equipment and clinical experience.

Jackson Kirkman-Brown Meurig Gallagher	62-63	1681-1725	This analysis is complex and includes things that are read as crossclaims to the sperm DNA damage testing. If there can be a claim DNA is 'not recommended' we presume this of 'research only' is a level beneath that – perhaps this could be clearer? We also wonder about the increasing prevalence of patients paying to be in research as highlighted by Joyce Harper – could this encourage this practice further by providing a concept that research means it has hope?	Research recommendations are formulated in situations where more research is advisable, such as for newer tests and treatments or in areas where current evidence points towards an effect however heterogeneity in study methods and population prevents firm conclusions or recommendations for its clinical application.
Carlos Calhaz-Jorge	62	1684	I suggest some rearrangement/specification: "In a prospective cohort study the role of oxidative stress in sperm DNA integrity (DNA fragmentation and ROS generation) and the peroxidation status of seminal plasma (malondialdehyde…"	The effect on sperm parameters was not within scope, however, "the role" was changed into "the levels"
Carlos Calhaz-Jorge	62	1689	"(8.6% vs 5.2%..." should be "(8.6 nmol/mL vs 5.2 nmol/mL…"	This was corrected in the text.
Carlos Calhaz-Jorge	62	1690	"0.78% vs 0.46%" should be "0.78 nmol/ mg of protein vs 0.46 nmol/ mg of protein"; "234% vs 148%" should be "234 nmol/L vs 148 nmol/L"	This was corrected in the text.
Carlos Calhaz-Jorge	62	1704	I suggest "22 fertile men" instead of "22 healthy men"	This was corrected in the text.
Carlos Calhaz-Jorge	62	1705	I could not find the meaning of "NO" in any part of the text. Sorry if it's my mistake	This was added to the text.
Carlos Calhaz-Jorge	63	1731	I could not find the meaning of "SOD" in any part of the text. Sorry if it's my mistake	This was added to the text.
Carlos Calhaz-Jorge	63	1735	I suggest "infertility, the authors investigated…"	This was adjusted in the text.
Carlos Calhaz-Jorge	63	1743	I suggest "infertility, the authors investigated…"	This was adjusted in the text.
Carlos Calhaz-Jorge	63	1747	I could not find the meaning of "FRAP" in any part of the text. Sorry if it's my mistake	The abbreviation was already explained in the text, but was not listed in the abbreviations list. It has now been added.
Mitranovici Melinda Ildiko	63	1750	The authors are interested only in follicular fluid oxidative stress biomarkers but not from endometrial level where interferes with implantation.	The GDG was unable to find information on endometrial oxidative stress in unexplained versus explained infertility.
Sabine Kliesch	64		In patients with UI a high proportion of chromosomal anomalies could be demonstrated in the literature cited. The conclusion, that this is not worth testing, is a contradiction in itself, especially a balanced translocation may go along with increased abortion rates in couples with UI. The analysis of the FSHB Gene variances is also not reflected adequately and studies on males with unexplained infertility are not considered. Thus the recommendation is not valid.	While genetic abnormalities were found in a proportion of patients with unexplained infertility, there was no consistent pattern as to prevalence and type of disorders comparing unexplained and explained infertility
Aboubakr Mohamed Elnashar	64	1759	Male unexplained infertility what is definition? Is there difference with male idiopathic infertility? Is sperm DNA fragmentation to be considered in male unexplained infertility?	The ICMART definition of unexplained infertility that was adopted in this guideline states for the male partner "apparently normal testicular function, genito-urinary

				anatomy and a normal ejaculate". We took male unexplained infertility to be the same as male idiopathic infertility if the WHO semen analysis was normal.
Carlos Calhaz-Jorge	64	1787	"… at the studied polymorphisms"	This was corrected in the text.
Mario Sousa	65	1811	Sperm aneuploidy screening is not recommended when WHO semen analysis is normal. In cases of infertility without a clinical diagnosis, I perform sperm aneuploidy screening. The reason is to full evaluate non visible causes, as for me there is always a cause.	Individual experience and practice, while valuable, is not supported by the literature in this instance.
Michael Morris	65	1811	In my opinion, this strong recommendation would be highly controversial, in disagreement with existing practice and even guidelines in some countries, and not clearly supported by the literature. For example: In the ESHRE draft - The cited study by Ertosun et al on karyotyping (line 1777-1780) suggests possible utility, admittedly without statistical analysis - The draft document also quotes "a significantly higher prevalence of chromosome abnormalities was observed in women with secondary infertility" (Papanikolaou et al, line 1805-1806) Other - The German AWMF guidelines (summarized by Wyrwoll et al 2021, https://doi.org/10.1515/medgen-2021-2051) clearly state "In couples with unexplained infertility, meaning that clinical diagnostics did not detect a reason for this condition, karyotyping should be offered to both partners". - Ventimiglia et al 2021 (doi: 10.3389/fendo.2021.801125) underlined the importance of genetic testing and remark that many men with identified possible causes "would have been recognised as having idiopathic infertility, with standard diagnostic exams." Furthermore, the phrasing of the recommendation is very broad and could be misunderstood: I assume the recommendation refers to diagnostic tests and does not preclude for example carrier testing which may be desired before ART. My suggestion: - Change the recommendation to address specifically diagnostic testing - Adjust the recommendation to "Conditional" to take into account the unclear literature and to respect existing guidelines (eg Germany).	The issue is whether genetic or genomic testing produces different results in unexplained versus explained infertility. We could not find evidence in the literature that this made a diagnostic difference. While respecting the German guideline, our evidence was not as positive towards genetic testing. There is no reason currently to remove the "strong" recommendation.

Liliana Ramos	65	1811	Genetic tests should be done only in the context of research, with the increasing information from NGS, it might be important in the future. Therefore, while now it is not enough evidence, it might be of importance to do more research	The recommendation was rephrased to "Genetic or genomic tests are currently not recommended in couples with unexplained infertility"
Liliana Ramos	65	1821	Remove from … and expensive intervention via IVF and PGT-A, depending on the particular genetic condition. As there is no recommendation for genetic testing, why suggest to use PGT-A "depending ofn the genetic condition'?	Adapted as suggested by the reviewer.
Adam Balen	66	1825	"Testing for vitamin D deficiency in females is not recommended". I think that this is too strong a recommendation. In the UK the lower limit of "normal" for vitamin D has recently been lowered from 75 nmol/l to 50 nmol/l, with less than 25 nmol/l being considered deficient, 25-50 insufficient and more than 50 nmol/l "sufficient". However, there is evidence that it is important that vitamin D levels are replete rather than just within the normal range and that this translates into improved outcomes with assisted conception treatments (1, 2). The vast majority of patients that we test are deficient in vitamin D and there are large ethnic variations, with those with darker skin usually having very low levels. There will inevitably be huge variations across Europe in this regard relating to skin colour, diet and sunlight exposure. Vitamin D deficiency has been associated with a number of poor reproductive outcomes in women and men (3, 4, 5) and also associated with miscarriage (6). Therefore at the very least I believe that it should be a GPP to measure vitamin D in couples attending with UI. This harmonises with points 36 "BMI evaluation in the female is considered good practice in pre-conception care" and 54 "A healthy diet and regular exercise, supported by behavioural therapy when necessary, are recommended". GPP 1. Chu J, Gallos I, Tobias A, Tan B, Eapen A, Coomarasamy A. Vitamin D and assisted reproductive treatment outcome: a systematic review and meta-analysis. Hum Reprod. 2018 Jan 1;33(1):65-80. doi: 10.1093/humrep/dex326. PMID: 29149263. 2. Zhao J, Huang X, Xu B, Yan Y, Zhang Q, Li Y. Whether vitamin D was associated with clinical outcome after IVF/ICSI: a systematic review and meta-analysis. Reprod Biol Endocrinol. 2018 Feb 9;16(1):13. doi: 10.1186/s12958-018-0324-3. PMID: 29426322; PMCID: PMC5807754. 3. Pilz S, Zittermann A, Obeid R, Hahn A, Pludowski P, Trummer C, Lerchbaum E, Pérez-López FR, Karras SN, März W. The Role of Vitamin D in Fertility and during Pregnancy and Lactation: A Review of Clinical Data. Int J Environ Res Public Health. 2018 Oct 12;15(10):2241. doi: 10.3390/ijerph15102241. PMID: 30322097; PMCID: PMC6210343. 4. Várbíró S, Takács I, Tűű L, Nas K, Sziva RE, Hetthéssy JR, Török M. Effects of Vitamin D on	While accepting that vitamin D deficiency is common, the evidence studied was not able to show a difference between unexplained and explained infertility. There may be value in measuring vitamin D when treatment is planned, but this is a diagnostic question here. Still, the recommendation was adapted to "Testing for vitamin D deficiency in females is not recommended for diagnosis of unexplained infertility"

			Fertility, Pregnancy and Polycystic Ovary Syndrome-A Review. Nutrients. 2022 Apr 15;14(8):1649. doi: 10.3390/nu14081649. PMID: 35458211; PMCID: PMC9029121. 5. de Angelis C, Galdiero M, Pivonello C, Garifalos F, Menafra D, Cariati F, Salzano C, Galdiero G, Piscopo M, Vece A, Colao A, Pivonello R. The role of vitamin D in male fertility: A focus on the testis. Rev Endocr Metab Disord. 2017 Sep;18(3):285-305. doi: 10.1007/s11154-017-9425-0. PMID: 28667465. 6. Tamblyn JA, Pilarski NSP, Markland AD, Marson EJ, Devall A, Hewison M, Morris RK, Coomarasamy A. Vitamin D and miscarriage: a systematic review and meta-analysis. Fertil Steril. 2022 Jul;118(1):111-122. doi: 10.1016/j.fertnstert.2022.04.017. Epub 2022 May 28. PMID: 35637024.	
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	67	R 34	TSH measurement is considered good practice in preconception care. But there is little evidence that without overt symptoms these women will have clinical hypothyroidism, thus the number of TSH measurements does not result in a significant benefit, thus why state this as good practice.	Hypothyroidism can occur in the absence of clinical symptoms and is confirmed by TSH and fT4 measurement. This is a GPP and not a formal recommendation for preconception care.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	67	1883	What is the normal range of TSH? Endocrine Society with upper limit of 2.5 mU/l pre-conception, despite intense discussion of this limit?	Individual studies have different recommendations for cut-off levels. The GDG leaves it to individual clinics and laboratories to decide the relevant levels for their patients.
Mario Sousa	67	1883	No additional thyroid evaluation in the female is recommended if TSH is within the normal range. In the general evaluation I always request Thyroid autoantibodies, as there are cases with euthyroidism.	While individual clinical experience is valuable, the literature reviewed did not provide evidence to support this approach.

J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	69	1943	"Reproductive outcomes are known to be impaired in men with low and high BMI" based on which reference?	This is not a formal recommendation, however, the sentence was adapted and a reference was added.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	69	1943	"Reproductive outcomes are known to be impaired in men with low and high BMI" based on which reference? One of our group members:" AGREE get rid of this old wive's tale"	This is not a formal recommendation, however, the sentence was adapted and a reference was added.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	69	1947	While healthy lifestyle intervention may improve spontaneous conception. Could a reference be added? Do you mean obese couples? Same with reproductive outcomes in men.	This is not a formal recommendation. The literature on lifestyle advice and intervention in infertility with regard to conception is contested with little evidence to show benefit with regards to fertility other than in anovulatory women. The sentence was however adapted and a reference was added.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang	69	1947	While healthy lifestyle intervention may improve spontaneous conception,…….. based on what evidence???	This is not a formal recommendation. The literature on lifestyle advice and intervention in infertility with regard to conception is contested with little evidence to show benefit with regards to fertility other than in anovulatory women. The

Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				sentence was however adapted and a reference was added.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	70	1939	References obesity. Where is Mutsaerts NEJM? https://pubmed.ncbi.nlm.nih.gov/27192672/ and Legro https://pubmed.ncbi.nlm.nih.gov/35041662/ https://onlinelibrary.wiley.com/doi/10.1111/obr.13325	These are all intervention studies, therefore not relevant in the diagnostic section. Furthermore, the population under study is not specifically UI.
III. TREATMENT				
Marco Sbracia			In this topic the role of woman's age is completely forgotten, whereas it is the major factor determining the outcome in infertile patients.	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely,			We miss the individualized approach i.e. starting treatment according to the patients'' profile and prognosis for natural conception. Prognosis-based management for example calculation the Hunault score is not mentioned. We miss the duration of subfertility. We miss accounting for the age of the women.	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.

Femke Mol, Mariette Goddijn		
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	The guideline is missing a section on prognosis for natural conception. After establishment of a diagnosis unexplained infertility (UI), the management should be guided by prognosis for natural conception: in couples with good prospects for natural conception treatment may be delayed, whereas in case of poor prognosis immediate treatment is warranted. We recommend to formulate separate PICO-question(s) on this topic and a new literature search.	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	Before treatment advice can be made there need to be recommendations on prognosis for natural conception.	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Christina Bergh Jan Bosteels	Recommendations on when to start treatment are prognosis dependent. It is remarkable that references https://pubmed.ncbi.nlm.nih.gov/36331493/ ,	The RCTs by Farquhar, Bensdorp and Steures are included in the body of

Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	https://pubmed.ncbi.nlm.nih.gov/16844491/ , https://pubmed.ncbi.nlm.nih.gov/29174128/ and https://pubmed.ncbi.nlm.nih.gov/25576320/ are not mentioned in the guideline. These high quality RCTs all consider prognosis for natural conception as inclusion criteria and therefore should be instrumental to inform the guideline. The Cochrane review from Veltman-Verhulst SM does not cover this. In short, • In couples with a prognosis for natural conception >30%, treatment with IUI does not add over expectant management (Steures). • In couples with a prognosis for natural conception <30%, IUI is better than expectant management (Mol F, Farquhar C). • As a first line treatment, IUI is equally effective as IVF, but cheaper (Bensdorp).	evidence of the treatment section. The RCT by Wessel was published after the final literature update.
Priya Bhide	To define the value of an intervention in order to make a recommendation, the authors should use comparisons where a single intervention is compared, with all other variables constant. E.g. clomiphene + timed intercourse vs natural cycle + IUI – it has two comparisons: CC vs natural cycle and timed intercourse vs IUI – how would this define the contribution of a single intervention?	In the treatment section, all forms of active treatment were compared with each other.
III.1 EXPECTANT MANAGEMENT		
George Lainas	In the expectant management section, no recommendation on expectant management is provided. Only recommendation comparing expectant management to treatment is provided. Therefore, recommendation on expectant Mx only, could be provided (i.e. Hanault model) or this paragraph could be part of active treatment.	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
George Lainas	GDG should define what difference in chances was selected to provide a reccomendation of a treatment vs another or vs/ expectant management	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	We miss the comparison timed intercourse without medication (LH test) compared to expectant management. Different reviews including two Cochrane reviews mention this comparison.	The GDG had a discussion regarding this topic. The consensus of the discussion was that the GDG considers timed intercourse without medication a form of expectant management and not an active treatment.

Mira Töyli			I would like to add one clinically relevant question to the guideline that I would, as a clinician the GDG to add to the guideline. When should treatment move from expectant management to active treatments. And what is the evidence supporting the recommendation. Is it one year (when the couple is officially suffering from infertility, 1,5 years or 2 years.)	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 39	This statement is probably not true for good prognosis patients but may be so for poor prognosis. This reinforces the need for stratification by prognosis. The cited evidence from the Cochrane review clearly differentiated prognoses but the recommendation did not do so.	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The reason for this is pragmatic: while in some countries, such as the Netherlands, the use of prognosis models is standard procedure, in most countries within Europe it is not because of the lack of validated, dynamic prognostic models. The GDG therefore decided to make a general recommendation instead of a specified one based on prognosis.
Marco Sbracia	10	R 40	In this point the committee suggest to use low dose regimen in gonadotropin treatment in order to avoid multiple pregnancy or OHSS, but without a test for ovarian reserve how is possible to schedule a performing treatment? These guidelines are for general practitioner or for specialist in Reproductive Medicine? These patients should or more appropriately "must" undergo ovarian reserve tests (AMH or AFC).	The GDG pointed out that ORT is not necessary to identify the aetiology of infertility or to predict the probability of pregnancy. However, as specified in the ESHRE guideline on Ovarian Stimulation, for predicting high and poor response to ovarian stimulation, AFC or AMH determination is recommended.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 40	A statement needs to acknowledge the need for adequate monitoring, whether hormonal and/or ultrasound	A sentence was added to the GPP.
Jean Calleja-Agius	10	R 41	Remove double fullstop	This was corrected in the text.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 41-44	Some of these conclusions conflict with the NICE guidelines where IUI is apparently not recommended. This may be due to a desire to reduce multiple pregnancy, cost-effectiveness, access to therapy etc and may be country and patient specific. IUI guidelines appear to differ from some aspects of the Cochrane recommendations as well as NICE, so this deserves review.	The GDG disagrees with the NICE Guidelines advice not to recommend IUI. There were several (Cochrane) meta-analysis published since 2019 and the GDG has evaluated them all.

			Recommendation 41 seems to be conflicted with 39 and 44. If IVF is not recommended over IUI-OI and IUI-OS is recommended over expectant management, IVF should not be recommended over expectant management. The most relevant Cochrane systematic review and network meta-analysis on this topic was not identified as the underlying evidence here.	
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	74	2138	We think that rephrasing the title to expectant management vs active treatment is more clear.	This comparison is specified in the PICO question underneath the title.
Priya Bhide	74	2153	Can the authors explain how the quality of evidence leading to this recommendation is strong with the low event rate and suboptimal information size – the SoF table is not available	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a strong recommendation. The reasoning of the GDG is explained in the justification.
Jackson Kirkman-Brown Meurig Gallagher	75	2174-2186	This RCT is contentious and differs from results seen in practice in national registries – the fact it saw no benefit of IVF – which few believe is often ignored. As such less weight must be given to this evidence as there was not clarity that all the requirements for UEI that you describe here were met.	This one RCT will probably never be repeated. The reasoning behind this recommendation is explained in full in the justification.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg	75	R. 38	IUI in a natural cycle is not recommended over expectant management in couples with unexplained infertility. Please provide references	The evidence for this recommendation can be found in section III.1 Expectant management, subsection "IUI in a natural cycle vs expectant management".

Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	75	2176	One more RCT found similar superiority of IUI in stimulated cycles compared to expectant management in women with poor natural conception chances (Wessel et al, HumRep, 2022) This study suggests in a subgroup analysis IUI might not increase live birth rate in older women compared to natural conception at home. Though very low certainty of evidence it points towards a knowledge gap: how to treat women with unexplained subfertility aged above 38 years.	The GDG is aware of the RCT by Wessel et al., however, it was published after the final update of the literature search.
George Lainas	75	2176	"INTRA-UTERINE INSEMINATION (IUI) IN A NATURAL CYCLE VS. EXPECTANT MANAGEMENT" One RCT on 332 patients is it enough to formulate a strong statement?	One well-executed RCT is considered high quality evidence to justify a strong recommendation.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	75	2188	Recently in November 2022 we published a RCT comparing IUI-OS and expectant management in couples with a poor prognosis for natural conception. We recommend to extend your search to include this second RCT (Wessel et al. nov 2022) in your Evidence. DOI: 10.1093/humrep/deac236	This RCT was published after the final literature search for the guideline. Furthermore, the conclusion of the study is in line with the evidence presented in the guideline, and would not change the recommendation.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	75	2194	This analysis includes both poor prognosis and moderate prognosis	That is correct. Both studies report no significant difference between groups and the direction of the effect is the same in both studies, therefore the pooled data were included in the guideline.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari	76	R 43	IUI with ovarian stimulation is recommended over expectant management in couples with unexplained infertility. Add in couples with a poor prognosis based on the studies: Steures 2006 et al, Farquhar 2018 et al DOI: 10.1016/S0140-6736(17)32406-6, Wessel et al 2022 DOI: 10.1093/humrep/deac236	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The reason for this is pragmatic: while in some countries, such as the Netherlands, the use of prognosis models is standard procedure, in most countries within Europe it is not because of the lack of validated, dynamic prognostic models. The GDG therefore decided to

Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				make a general recommendation instead of a specified one based on prognosis.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	76	2195	A distinction should be made b/n poor prognosis and moderate prognosis (see below).	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The reason for this is pragmatic: while in some countries, such as the Netherlands, the use of prognosis models is standard procedure, in most countries within Europe it is not because of the lack of validated, dynamic prognostic models. The GDG therefore decided to make a general recommendation instead of a specified one based on prognosis.
Priya Bhide	76	2195	Can the authors explain how the quality of evidence leading to this recommendation is strong with the low event rate and suboptimal information size – the SoF table is not available. Can a single underpowered RCT be classified as a SR and MA?	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a strong recommendation. The reasoning of the GDG is explained in the justification.
Gustavo Botti	76	2195	I do not think that an IUI should be performed without first ruling out pelvic pathology (endometriosis or adhesions) with a laparoscopy if necessary	There is insufficient evidence to suggest that clinically relevant diagnoses will be missed by omitting a laparoscopy in patients at low risk for tubal pathology.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	76	2199	"The weight of evidence strongly suggests that IUI with ovarian stimulation is recommended in preference to expectant management, particularly for couples with poor prognosis". The statement formulated as such is incorrect. According to the available evidence effect is seen only in the poor prognosis group (OR 4.48, 95% CI 2.00 to 10.01; 1 RCT; 201 women), but not in the moderate prognosis (OR 0.82, 95% CI 0.45 to 1.49; 1 RCT, 253 women)	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The reason for this is pragmatic: while in some countries, such as the Netherlands, the use of prognosis models is standard procedure, in most

				countries within Europe it is not because of the lack of validated, dynamic prognostic models. The GDG therefore decided to make a general recommendation instead of a specified one based on prognosis.
Jean Calleja-Agius	77	2222	Please stress that the evidence level here is very poor – maybe even remove it from being a recommendation at all	The GDG decided to restructure the section and the recommendation was removed.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	77	2223	Cochrane review does not take into account individual prognosis https://doi.org/10.1002/14651858.cd001838.pub6 Based on Carosso 2022 the recommendation should be rephrased since waiting for 1 year is cost-effective “the GDG opinion is that the decision to use IVF should be based on patient characteristics, costs and patient preferences”	The Cochrane review cannot take into account individual prognosis when the included RCTs were not designed this way. Carosso et al. did also not take individual prognosis into account.
III.2 ACTIVE TREATMENT				
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	/	/	We think that rephrasing the title to IUI versus IVF is more clear. We recommend comparison to include a separate comparison about IUI versus IVF in older women (38 years or older). This is an important knowledge gap which need to be resolved.	The GDG discussed your suggestion, however, decided not to formulate a separate recommendation. The GDG has set the age-limit for UI to 40 years of age, therefore, creating a separate recommendation for ages 38-40 seemed inappropriate.
Mario Sousa	10	42	After 2-3 IUI, I perform IVF if the ovarian age is less than 35y, IVF immediately if the ovarian age is ≥ 35y	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.

Unexplained Infertility Guideline Australian Adaptation Committee	10	R 43-45	Should there be a recommendation on what type of stimulation is superior? There are different stimulation options that should be discussed.	The reader is referred to the ESHRE guideline on Ovarian Stimulation for the review of different stimulation options.
Unexplained Infertility Guideline Australian Adaptation Committee	10	R 43 and 44	It was felt that patient perception and distress were high in a couple with this diagnosis, and this may promote practice towards IVF to maximize a per cycle higher success rate of IVF over IUI. Has cost-effectiveness and patient choice been considered in high and low cost healthcare systems? Grade evaluation will assess this and may vary from country to country.	Patient perspective was taken into account when formulating the recommendations for the guideline. Cost effectiveness is context based (drugs are expensive in low income settings while services such as monitoring and nurse/doctor time are comparatively more expensive in high income settings)
Carlos Calhaz-Jorge	10	R 44	I suggest to add “as the first line treatment”	The GDG thought this was redundant since the previous recommendation indicated IUI is the first-line treatment.
Marco Sbracia	10	R44	This point contradicts the other point 4 and 42. Furthermore, it has not mentioned in any of these points the age of woman with unexplained infertility that instead is the most relevant condition for the diagnosis and treatment of these patients. Woman's age should be always considered before starting diagnostic and treatment procedures.	The GDG understands where confusion may arise and has therefore decided to restructure the expectant management section.
Jackson Kirkman-Brown Meurig Gallagher	75	2188-2207 2324-2341	We welcome the clarity of answers	The GDG understands where confusion may arise and has therefore decided to restructure the expectant management section.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	76 81		Factors for individual decision towards IVF are age and AMH/AFC as discussed in the diagnostic section. Refer to this section. Recommendation p. 81: "IVF is probably not recommended over IUI with ovarian stimulation in couples with unexplained infertility in the female age group < 38 years." - We suggest specifying this recommendation based on the available evidence: as sensitvity analysis has demonstrated higher LBR of IVF vs IUI in women > 38 years.	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The GDG has set the age-limit for UI to 40 years of age, therefore, creating a separate recommendation for ages 38-40 seemed inappropriate.
Priya Bhide	79	2262	Could the authors summarise the output from this comparison?	This information is included in the annexes of the guideline, which will be published on the ESHRE website together with the guideline.
Priya Bhide	80	2309	OS + timed intercourse vs OS with IUI – there is uncertainty of benefit – the intervention tested here is IUI – indicating no benefit for IUI	It is a bit risky to extrapolate in this situation and while algebraically this may

			Natural cycle IUI vs OS + IUI – the intervention tested here is OS – indicating benefit of OS Hence could the authors explain why OS + timed intercourse is not the recommended treatment since this intervention (OS) is shown to be effective rather than the actual IUI process In the comparison of stimulated IUI vs expectant management, the beneficial effect may be due to the OS rather than the IUI	seem to be correct, we are talking about complex interventions where 1 part cannot be dissociated from the second.
Carlos Calhaz-Jorge	80	2294	"…timed intercourse with gonadotropins to ovarian…"	This was corrected in the text.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	81	2339	We miss the safety aspects of IUI-OS, for example withholding insemination when more than 3 dominant follicles developed. When using strict cancel criteria multiple pregnancies can be prevented. We recommend to extend your search to include the RCT of Danhof N. et al. DOI: 10.1093/humrep/dey268 We miss the network comparison FSH vs Clomid vs Letrozole as ovarian stimulation for IUI. Was Bensdorp et al 2015, BMJ included?	The GDG discussed the need to specify cancellation criteria in the GPP. However, it was implied with the recommendation to use a low-dose gonadotropin regime. Different ovarian stimulation regimes were not part of the PICO. The reader is referred to the ESHRE guideline on ovarian stimulation for information on the comparison of ovarian stimulation protocols for IUI. Bensdorp 2015 is included in the systematic review by Nandi 2022 and therefore not mentioned separately.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	81	2339	There is no strong evidence to give a recommendation that IUI with ovarian stimulation over natural IUI is recommended as a first-line treatment for couples with unexplained infertility. Please provide the references	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a strong recommendation. The reasoning of the GDG is explained in the justification.

Carlos Calhaz-Jorge	81	2340	I suggest the GDG to considered the inclusion of a cautious GPP stating that other parameters must be considered when deciding IUI vs IVF, namely the female age	A GPP has been added to clarify this.
Mario Sousa	81	2340	IVF is probably not recommended over IUI with ovarian stimulation in couples with unexplained infertility. No. IVF is the first choice if the ovarian age is ≥35y	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Maria Elisabetta Coccia	81	2340	I suggest to add: ≤ 38 years. In women ≥38 years, live birth rate was significantly higher after IVF treatment	The GDG discussed your suggestion, however, decided not to adapt the recommendation. The GDG has set the age-limit for UI to 40 years of age, therefore, creating a separate recommendation for ages 38-40 seemed inappropriate.
Mario Sousa	82	2365	ICSI is not recommended over conventional IVF in couples with unexplained infertility. But should be used in cases of failed fertilization or poor embryo development, or after 2 IVF failures	Failed fertilisation was not within the scope of the key question for which the recommendation was formulated. When formulating the PICO to address the key question, the definition of unexplained infertility in accordance with ICMART 2017 as was stated.
III.3 MECHANICAL-SURGICAL PROCEDURES				
Liliana Ramos	11	/	Recommendation number for “If incidentally minimal to mild endometriosis”…. Is missing	This is not a recommendation, but a conclusion, which is why it does not have a number.
George Lainas	11	/	Number of recommendation is missing, as well as level of evidence	This is not a recommendation, but a conclusion, which is why it does not have a number.
Unexplained Infertility Guideline Australian Adaptation Committee	11	R 47	Should cross reference to recommendations on tubal testing previously	In the section of tubal patency testing, a reference was added to the treatment section on tubal flushing.
Carlos Calhaz-Jorge	11	R 48	Maybe better “Endometrial scratch should not be offered in unexplained infertility”	The recommendation was adapted as suggested by the reviewer.
Mario Sousa	11	48	I offer Scratching if RIF or RPL occurs	RIF and RPL are considered outside the scope of this guideline.

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	8, 11,	R 7-10, R 47	Recommendation 47 (tubal flushing) should be integrated with recommendation 7 to 10.	Tubal flushing is an intervention, recommendations 7 to 10 are on diagnosis.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	84	2419	Seyam et al – needs to be checked for trustworthiness (same author under investigation for plagiarism (RCTs in PCOS) In general, there are more studies that we think are problematic.	All of the RCTs in this guideline have been checked and none have an editorial note that they are either under investigation or withdrawn.
Carlos Calhaz-Jorge	84	2424	I suggest to add the text highlighted in yellow: "10/100 in women not submitted to microhysteroscopy (RR 4.30…"	Thank you for your comment. This has been clarified.
Carlos Calhaz-Jorge	84	2426-2431	Casini et al (2006) included 181 women with infertility; 92 of them were operated because of fibroids; one inclusion criterion was fibroid <4.0cm; they found a significant improvement in the pregnancy rate after surgery in SM fibroids. My questions: Are those patients diagnosed as UI? Is this Casini paper the one included in this "Evidence"subsection?	This trial included women with normal findings after the usual diagnostic tests except for the presence of uterine fibroids. It is not well known to what extent fibroids influence fertility and for this reason the GDG included this single published RCT on treatment. We elaborated on this in more detail in the text.
Carlos Calhaz-Jorge	84	2432	Nothing in the Evidence text refers to "abnormalities not seen at routine imaging". Additionally, as the recommendation uses "screening hysteroscopy", my suggestion is to remove either "screening" or "not seen at routine imaging" because they repeat the same concept.	We have removed the word 'screening' before hysteroscopy.

Mario Sousa	84	2432	Screening hysteroscopy for the detection and possible correction of intrauterine abnormalities not seen at routine imaging is not recommended The text is confusing. Hysteroscopy is a routine mandatory part of female evaluation. If defects are found is another thing.	We have removed the word 'screening' before hysteroscopy.
Carlos Calhaz-Jorge	85	2466-2470	The population of the van Welie study is described. However, no results are presented	Thank you for your comment. This has now been included.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	85	2471	In stead of :"HSG (i.e., tubal flushing) with an oil-soluble contrast medium is preferable over a water-soluble contrast medium." …. Risks and benefits of tubal flushing with oil-based contrast should be discussed with all couples with unexplained infertility. Suggested ref for risks: Safety of HSG with oil-based contrast medium: a Systematic review. Published in Reproductive BioMedicine Online 2021 doi: 10.1016/j.rbmo.2021.03.014 Reference https://pubmed.ncbi.nlm.nih.gov/35399811/ is missing	The recommendation was adjusted. The SR by Roest et al. was already cited in the justification.
Carlos Calhaz-Jorge	85	2474-2475	Being the largest trial it would be good to have the Dreyer study described in the "Evidence" text	The study by Dreyer is included in the systematic review by Wang et al., 2020 and therefore not described separately in the evidence section.
Carlos Calhaz-Jorge	86	2495-2496	Is "further information" needed in the present context? I suggest to delete this lines, maybe here by default.	The further information is referring to the details of the studies in the evidence tables, and the summary of evidence tables.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	86	2498	There is a recent Cochrane review on endometrial scratching (Bui et al 2022; https://www.cochranelibrary.com/cdsr/doi/10.1002/14651858.CD011424.pub4/full though the certainty of the evidence was very low for most outcomes, and many studies present with potential integrity issues (ie Maged et al; see below)	The systematic review of Bui et al., 2022 is not specific for unexplained infertility, and also does not include sub-analyses by infertility diagnosis.

J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	87	2534	Maged et al – this author has multiple retracted studies and expressions of concern	There are 5 RCT's cited in the evidence section for "the use of endometrial scratching in IUI". Excluding the study by Maged et al would not change the recommendation.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	87	2539	Recommendation 48 cannot be strong because it is at least partly based (partly) on potentially fabricated data. In fact, none of the studies that you cite shared data when we (BWM, MvW) requested that.	Even if the data is potentially fabricated, the (lack of) plausibility of the underlying biological mechanism and the heterogeneity in methodology of the studies warrant a strong recommendation.
Priya Bhide	87	2539	Could the authors provide a pooled estimate of all studies examining this intervention to support the recommendation?	The GDG relies on published meta-analyses and does not make their own. The GDG made an exception for the tubal patency testing.
III.4 ALTERNATIVE THERAPEUTIC APPROACHES				
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	90	2625	Based on the available evidence, shouldn't this recommendation be "strong"?	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	90	2625	Adjunct oral antioxidant therapy to females undergoing fertility treatment is ~~probably~~ not recommended	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	90	2626	Adjunct oral antioxidant therapy to males undergoing fertility treatment is ~~probably~~ not recommended.	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Carlos Calhaz-Jorge	90	2625 and 2626	In the absence of reliable information why not to say "It is not recommended" (as it was decided in previous subsections) instead of "probably not recommended"?	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients'

				perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	91	2643	Acupuncture in women is probably not recommended should be Acupuncture in women is ~~probably~~ not recommended	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	91	2660	Inositol supplementation in women is probably not recommended, should be .. not recommended (based on one underpowered RCT this should not be recommended)	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients' perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.

Carlos Calhaz-Jorge	91	2660	In the absence of reliable information why not to say “It is not recommended” (as it was decided in previous subsections) instead of “probably not recommended”?	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients’ perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Jackson Kirkman-Brown Meurig Gallagher	92	2693-2702	We wonder whether this important finding should come first in the section before the don’t recommend supplements messages	The guideline recommendations stick to the order of the clinical questions that were addressed.
Mario Sousa	11	R 50	Antioxidants for males is beneficial to sperm	This clinical question looked at outcomes of LBR and multiple pregnancy rates. Surrogate outcomes such as effect on semen parameters were not considered.
Mario Sousa	11	R 51	I offer Acupuncture in cases of RIF and RPL	This guideline only looked at unexplained infertility patient population. RIF and RPL patients are considered outside the scope of this guideline.
Unexplained Infertility Guideline Australian Adaptation Committee	11	R 51	Some felt the recommendation should be stronger in using “should not” be recommended. The Grade assessment framework does not reflect the recommendation strength and should be revisited.	When formulating recommendations, not only the quality of the available evidence needs to be taken into account, but also benefits versus harms, patients’ perspective, health system perspective and the resource use. After considering all these factors, the GDG decided on a weak recommendation.
Mario Sousa	11	R 53	Psychological guidance is introduced since the first consultation	The GDG agrees with the reviewer. This comment is reflected in the GPP.
IV. QoL				
Unexplained Infertility Guideline Australian Adaptation Committee	11	R 55	Some counsellors were surprised by these statements although little data is available. The wording may be too strong for the evidence available. There is little discussion on stress in the guidelines. Stress is often perceived as a cause of infertility and yet there is little evidence for a biological causal link between infertility and stress.. Would be valuable for patients and clinicians to be given information on this so that clinicians do not	The GDG has reviewed the recommendation and has decided not to change it.

			inadvertently support the notion of this causal link. Maybe this could be covered in the update. Maybe we need to use the words “we don’t know “ more often!	
Carlos Calhaz-Jorge	94	2741-2748	This paragraph seems a little confusing. In line 2743 “childless males from couples with unexplained infertility undergoing fertility workup”. If they are undergoing fertility workup how can we consider the males having unexplained infertility? Then it was stated that men in the previous condition had higher FertiQoL scores compared to those belonging to couples with several infertility factors “before diagnostic disclosure and in the follow-up 2 to 3 months…”. Sorry, can you please make the text more clear?	The males were diagnosed with unexplained infertility after the fertility work up but the first intervention in the study was performed before the diagnosis was given. The four study occasions are now clarified in the text.
Carlos Calhaz-Jorge	94	2759	Is this wording format acceptable as a recommendations?	The GDG has reviewed the recommendation and has decided not to change it.
Carlos Calhaz-Jorge	94	2759	Second half of the recommendation (related to men): As it refers to a male partner of a couple where the female suffers from PCOS, I find strange to use “men with unexplained infertility” (last line). Men can have normal sperm but the infertility is not unexplained. Can be clarified, please?	If we understand the comment correctly, it is advised to not write "men with unexplained infertility". Therefore, this term is changed to "men from a couple with UI infertility".
ANNEX 1				
Dimitrios G. Goulis Giovanni Corona			The issues named above can be partially explained by the fact that very few scientists and clinicians working primarily in the field of Andrology have been invited to participate in the writing group. Therefore, the guideline is much more oriented towards the diagnostic and therapeutic approach of the female unexplained infertility than the couple unexplained infertility, which was its initial task.	As described in the Manual for ESHRE guideline development, the GDG was composed of content experts, including an andrologist, and non-expert clinicians, a nurse and a patient representative, and a balance was achieved in geographical location, gender and expertise. Furthermore, the ESHRE Special Interest Group Andrology provided feedback both during the formulation of the key questions and during stakeholder review.
Maria Schubert			In my work as Clinician Scientist I focus on idiopathic and unexplained male infertility. One of the overarching aims is the identification of putative etiologic factors contributing to impaired fertility. I very much support the emerge of a “couple” guideline on unexplained infertility. However, in the current status of the guideline the male part is hardly considered, or even	As explained in the definition section of the guideline, the GDG is adhering to the ICMART definition of unexplained infertility, which states for the male "apparently

			rejected from further diagnostic procedures when normozoospermia is present. I herewith apply to contribute more substantially to the male part of this guideline. Please consider this offer seriously, as I believe that Clinicians working in the field of infertility should be informed evidence-based on both sides of the couple.	normal testicular function, genito-urinary anatomy and a normal ejaculate".
Sabine Kliesch	97	2787	ii) The share of clinical andrological contribution by experts in the field is very unfortunate to the share of experts in female infertility. This is also reflected in the disproportion of chapters dealing with female vs. male diagnostics when this is known to affect the couple similiarily. To the future reader this leads to the assumption that the major focus in couple infertility is to be laid on the female side. This neglects that the identification of etiologic factors on the male side, by further and thorough diagnostics, beyond basic semen analysis, may reduce the therapeutic burden on the female side. The guideline, in its current version, focusses very much on which analyses in the male NOT to perform, instead of suggesting or discussing examinations, that increase phenotyping and may therewith identify etiologic causative factors.	As described in the Manual for ESHRE guideline development, the GDG was composed of content experts, including an andrologist, and non-expert clinicians, a nurse and a patient representative, and a balance was achieved in geographical location, gender and expertise. Furthermore, the ESHRE Special Interest Group Andrology provided feedback both during the formulation of the key questions and during stakeholder review.
Mol BW	98	2792	many of the members of the guideline development group work or have worked as IVF doctor; either public or private. This is a serious conflict of interest and should be declared clearly; i.e. doctor X has worked from then till ……. in private/public clinic X that provides infertility treatments (…… IUI cycles per year; …… IVF cycles per year)	All GDG members have declared their affiliation and their conflict of interest. Furthermore, as described in the Manual for ESHRE guideline development, the GDG was composed of content experts and non-expert clinicians, a nurse and a patient representative, and a balance was achieved in geographical location, gender and expertise. Therefore conflict of interest from working as an IVF doctor is highly unlikely to have had an influence in this document.
ANNEX 3				
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn			A top ten prioritized research agenda was developed by an international team (Duffy et al. Hum Reprod. 2020;35(12):2715-2724. These remain important knowledge gaps	Thank you for your suggestion, the GDG will take it into consideration when deciding on the top priorities for research in the field of unexplained infertility.

Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	101		Recommendations for research: Add: The role of lifestyle intervention in unexplained infertility	Thank you for your suggestion, the GDG will take it into consideration when deciding on the top priorities for research in the field of unexplained infertility.
Mitranovici Melinda Ildiko	101	2802-2810	Future research: the role of oxidative stress biomarkers in endometrial implantation, are they important or not?	Thank you for your suggestion, the GDG will take it into consideration when deciding on the top priorities for research in the field of unexplained infertility.
Mitranovici Melinda Ildiko	101	2802-2810	Future research: the role of different endometrial biomarkers, are important or not, we are not talking about repeated implantation failure	Thank you for your suggestion, the GDG will take it into consideration when deciding on the top priorities for research in the field of unexplained infertility.
GENERAL COMMENTS				
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol			There are a lot of recommendations on what not to do, but the guideline would benefit from an initial list of tests that should be done to establish a diagnosis of unexplained infertility.	A flow chart will be produced and will be published together with the final version of the guideline.

Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely				
Monica Varma			Does the age of patient make any difference in the diagnosis /management in the list of recommendations	The GDG has set the threshold for unexplained infertility to 40 years of age for the woman. For management, age refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Unexplained Infertility Guideline Australian Adaptation Committee		R 14-24	Some members felt these recommendations were unstructured and reflected the fact that questions were not well organized from the start in terms of prognosis	The GDG had several online meetings to discuss the PICO questions that are the backbone of this guideline. The recommendations are structured according to the PICO questions.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn		20; 27; 49-52	Is “Probably” standard wording for conditional recommendations in guidelines, we hope not.	"Probably" is standard wording for conditional recommendations, according to the GRADE manual for guideline development.
Carlos Calhaz-Jorge	/	/	A great thanks to all the authors for their hard work and congratulations for the way they cope with a not at all easy topic.	Thank you for your kind words.
Liliana Ramos	/	/	Sometimes in the text the authors use UI and sometimes unexplained infertility. I should be consistent throughout the document	This has been adapted for consistency.
Adam Balen	/	/	This is an excellent document and a very balanced and appropriately written guideline. I congratulate the authors on a great piece of work.	Thank you for your kind words.
George Lainas	/	/	Congratulations to the GDG for providing a thorough and well constructed document.	Thank you for your kind words.
George Lainas	/	/	Counseling prediction tools are absent, regarding treatment comparison of interventions for couples with UI	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.
Maria Elisabetta Coccia	/	/	VERY WELL DONE – COMPLETE – FROM DIAGNOSIS TO TREATMENT	Thank you for your kind words.

Mol BW	/	/	I want to express our appreciation towards the GDG for their time on effort on this important topic.	Thank you for your kind words.
Mitranovici Melinda Ildiko	/	/	Thank you for your kind invitation to review such an amazing guideline.	Thank you for your kind words.
Sabine Kliesch	/	/	It is about time that clinicians working in the field of reproductive medicine are provided with evidence-based information on the optimal diagnostic and therapeutic work-up on couples with infertility of unknown origin. We therefore appreciate the effort of the ESHRE to set up a respective guideline. However, we, as the German Societies of Andrology and of Urology, have major concerns i) on the andrological content of the guideline, and ii) on the content of the guideline group.	Thank you for your kind words. As described in the Manual for ESHRE guideline development, the GDG was composed of content experts and non-expert clinicians, a nurse and a patient representative, and a balance was achieved in geographical location, gender and expertise.
Panayotidis Costas	/	/	Dear respectful colleagues and ESHRE committee for this guideline, first I would like to congratulate for your hard work to summarise and bring practical recommendations regarding the Unexplained Hypofertility diagnosis and management.	Thank you for your kind words.
Jackson Kirkman-Brown Meurig Gallagher	/	/	We wish to recognize the effort that has clearly gone into this comprehensive and excellent document.	Thank you for your kind words.
J.A. Wessel, Elena Kostova, Monique Mochtar, Madelon van Wely, Femke Mol, Mariette Goddijn	/	/	We noted several problematic studies were included. Please be aware that not everything that is published is valid. At least for the update of the guideline make sure a team tests all studies with a checklist.	All studies included in the guideline have been checked for editorial notes of concern or withdrawal. At the time of stakeholder review, this checklist had not been published. The GDG has discussed the issue of trustworthiness of research papers and has decided not to exclude studies unless proven.
Dimitrios G. Goulis Giovanni Corona	/	/	The European Academy of Andrology, based on the expertise of its members, will comment only on the andrological aspects of unexplained infertility.	Thank you. The GDG welcomes all feedback.
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	The guideline is potentially flawed by not taking a prognostic approach to investigation and treatments. Obviously significant factors such as tubal, ovulation and semen analysis need exclusion to lead to a diagnosis of unexplained infertility. The remaining couples have a good, intermediate or poor prognosis which demands different approaches. No attempt is made to introduce prognosis models or approaches, some of which are currently available but disputed. Some members felt you should first identify those with low chances of	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.

			natural conception due to identifiable causes and the remainder should have prognostic assessment prior to treatment.	
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	The guideline is not consumer (patient) orientated in many of its approaches. For instance, while AMH does not predict subsequent fertility, it does indicate likely egg numbers if IVF is needed. It may indicate the chance of a woman having one child only when she may desire more and so is prognostic of her lifetime chance of success in achieving her family size aims. Family size aspirations should be a strong consideration in terms of investigations and treatment. In terms of unexplained infertility, having a child (children) is important but for many women, finding a potential cause to explain her lack of fertility may be important. She and her partner may wish to undergo more testing than recommended here to understand the problems they face. At least the choice should be considered. A more consumer-oriented perspective on presentation of the guideline would be appreciated to address their aspirations and questions.	A patient leaflet and flowcharts will be published with the guideline as tools for the practical application of the guideline.
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	It was felt a flow chart of investigations and treatment options would be valuable for patients and health care providers. There are many negative statements about what should not be done for investigation and treatment but few positive ones. These deficiencies could be remedied by a more visual depiction of the process advocated by the guideline.	A flow chart will be produced and will be published together with the final version of the guideline.
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	None of the papers discussed have gone through any form of integrity testing and some have been retracted or are under review for implausible data. While it may be too late to remedy this, there should be a clear discussion of the problem and the next update should include rigorous checking.	All included RCTs were checked, none are retracted. Furthermore, none have currently and editorial message that the publication is under investigation.
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	The problem of unexplained infertility is it is a diagnosis of exclusion. The guideline covers diagnostic investigations and rejects some investigations that would move the diagnosis to explained infertility eg laparoscopy might make a diagnosis of endometriosis, luteal progesterone monitoring may lead to diagnosis of luteal phase defect or anovulation. The group understood this was a circular argument ie what comes first – all diagnostic attempts or minimal testing as advocated in these guidelines. Many clinicians may not want to reject previously standard tests such as laparoscopy, hormone testing, DNA sperm testing etc. it was also pointed out that many groups internationally did not have good access to high quality ultrasound for instance. This definition problem will probably be raised by many groups. The group was also concerned re age issues and cutoff at 40 years was queried. Some felt the guideline could deal with a younger age group rather than those where egg quality is	The GDG has reviewed all available evidence on the benefit of further testing to establish the diagnosis of UI and found none. Therefore, further testing such as laparoscopy, DNA fragmentation, hormone testing are not recommended. The GDG has set the age cut-off on 40 years, based on a review of 237 studies on UI, as specified in the definition section.

			deteriorated. ICMART definition does not appear to involve age and ESHRE needs to decide on an age range to which this implies.	
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	Can unexplained infertility be experienced in single women or same sex couples ie is this a heteronormative definition? Is a woman who has used donor sperm and not got pregnant, have unexplained infertility? This could be covered in the introduction and should be reviewed in recommendations 1-3. It was felt single people and same sex couples should also be acknowledged in the introduction.	The reviewer has a point. A statement has been added to the introduction.
Unexplained Infertility Guideline Australian Adaptation Committee	/	/	No technical data was available to see if the Grade recommendations were appropriately worded.	The evidence tables and summary of evidence tables will be published together with the final version of the guideline.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	/	/	We appreciate the effort and would like to thank the guideline development group for their hard work on this important topic.	Thank you for your kind words.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	/	/	Clearly structured guideline with 2 parts, with the main aims to standardize the diagnosis of UI for daily medical practice and for studies and treatment. The guideline focuses on clearly defined medical outcome parameter (critical live full-term singleton birth etc.) and economic and psychosocial aspects. The structure with summary of recommendations and structured narrative questions with concice answers is precise. In comparison to the ASRM guideline this guideline also includes recommendation for pre-conception counseling, diagnosis and alternative treatments.	Thank you.
Ziller V., Goeckenjahn M., Köhn F.-M.., Hancke K. , Sonntag B.	/	/	Even if the GDG, with its frequently negative recommendations on interventions in the absence of evidence, presumably wants to prevent the misuse of recommendations in routine practice outside of corresponding studies, this seems to be in contrast to the research questions then formulated on p. 101. The absence of evidence should not always be the basis for not recommending an intervention, especially not with grading “strong” - this is an issue in many aspects of the guideline - if there is no supportive evidence, this should be mentioned, but not necessarily be the basis for not doing it - in many aspects the recommendation should rather be "do it under research conditions". In the current context the guideline will solidify the diagnosis of “unexplained infertility” as only very few diagnostic procedures are recommended. As “unexplained infertility” is found in only about 10 % of our patients we would expect to	The GDG disagrees with the reviewer. The strong recommendations against investigations are not solely based on the absence of evidence, but also on biological plausibility, methodological heterogeneity, the benefit of testing in the absence of treatment options, acceptability to stakeholders, costs etc.

			detect numerous rare conditions. Rare is rare but in unexplained sterility we will often find the rare. This will make it difficult to create evidence, as rare findings are difficult to study, but in many aspects of this guideline, in diagnostic as well as in therapeutic approaches, the absence of evidence is interpreted as “strong” evidence not to recommend certain measures. This should be re-evaluated thoroughly as we feel the level of evidence, even as provided in the text, does not always support strong recommendations and is also missing the option to focus on further research.	
Åsa Magnusson	/	/	I would just like to express my admiration for the great work and effort made by the Unexplained Infertility Guideline group. Well elaborated and very useful in clinical practice. I have nothing further to add	Thank you for your kind words.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	/	/	Please find our comments, suggestions and questions below. First, we want to express our appreciation towards the members of the GDG and the ESHRE support staff for their time on effort on this important topic. We are pleased to see that there will be an ESHRE guideline on unexplained infertility. We look forward to see your responses.	Thank you for your kind words.
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven	/	/	Suggested structure: A diagnosis of unexplained infertility is established if a couple is trying to conceive for 12 months and the following series of tests show no abnormalities • Normal semen-analysis as confirmed by one/two semen analysis with the Following criteria……. • Normal ovulation as confirmed by: urinary LH measurements, ultrasound monitoring or mid-luteal progesterone measurement. Or maybe	A flow chart will be produced and will be published together with the final version of the guideline.

Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely			recommendation on page 17 (no ovulatory test needed), which seems in contrast with the above. • Tubal patency as confirmed by HyCoSy or HSG (Is one-sided patency enough to diagnose unexplained infertility) • Normal cavum uteri as confirmed by ultrasound. As part of good practice, we recommend the following • TSH measurement • Evaluation of body mass index The following tests are not recommended as a routine . • Hysteroscopy • Laparoscopy • Vaginal microbiota testing • Post Coital test • ……..	
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	/	/	The references should be checked for trustworthiness. A checklist that could be used is here: https://assets.researchsquare.com/files/rs-2119486/v1/97359e33-7686-4b8c-98e4-20f2fdce2d41.pdf?c=1673464062 Below are the problematic papers that we identified in the guideline. • Hamed HO, Shahin AY, Elsamman AM. Hysterosalpingo-contrast sonography versus radiographic hysterosalpingography in the evaluation of tubal patency. International journal of gynaecology and obstetrics: the official organ of the International Federation of Gynaecology and Obstetrics 2009;105: 842 215-217. • Rezk M, Shawky M. The safety and acceptability of saline infusion sonography versus hysterosalpingography for evaluation of tubal patency in infertile women. Middle East Fertility Society Journal 2015;20:108-113. • Malek-Mellouli M, Gharbi H, Reziga H. The value of sonohysterography in the diagnosis of tubal patency among infertile patients. La Tunisie medicale 2013;91: 387-390. • Ibrahim MI, Moustafa RA, Abdel-Azeem AA. Letrozole versus clomiphene citrate for superovulation in Egyptian women with unexplained infertility: a randomized controlled trial. Archives of gynecology and obstetrics 2012;286: 1581-1587. • Maged AM, Al-Inany H, Salama KM, Souidan, II, Abo Ragab HM, Elnassery N. Endometrial Scratch Injury Induces Higher Pregnancy Rate for Women With Unexplained Infertility	All studies included in the guideline have been checked for editorial notes of concern or withdrawal. At the time of stakeholder review, this checklist was in pre-print. The GDG has discussed the issue of trustworthiness of research papers and has decided not to exclude studies unless proven.

			Undergoing With Ovarian Stimulation: A Randomized Controlled Trial. Reproductive sciences (Thousand Oaks, Calif)n2016;23: 239-243 • Parsanezhad ME, Dadras N, Maharlouei N, Neghahban L, Keramati P, Amini M. Pregnancy rate after endometrial injury in couples with unexplained infertility: A randomized clinical trial. Iranian journal of reproductive medicine 2013;11: 869-874. • Seyam EM, Hassan MM, Mohamed Sayed Gad MT, Mahmoud HS, Ibrahim MG. Pregnancy Outcome after Office Microhysteroscopy in Women with Unexplained Infertility. International journal of fertility 2606 & sterility 2015;9: 168-175. • Jafarabadi MN, Bagheri M, Ebrahimi Z, Shariat M, Haghollahi F. Endometrial scratching effect on pregnancy rate in intrauterine insemination cycles: a randomized controlled trial. International journal of women's health and reproduction sciences 2020;8: 85-89. From the studies on endometrial scratching most were not willing to share data in an IPD (Senocak GC and others). We would suggest to label these studies as awaiting further classification (https://documentation.cochrane.org/display/EPPR/Policy+for+managing+potentially+problematic+studies%3A+implementation+guidance) section 2.3. and not use them for this guideline.	
Christina Bergh Jan Bosteels Frank Broekmans Astrid Cantineau Arri Coomarasamy Vinh Dang Annemieke Hoek Joop Laven Rong Li Abha Maheswari Ben W. Mol Anja Pinborg Annika Strandell Chris Venetis Lan Vuong Madelon van Wely	/	/	PICO questions on prognosis should be incorporated in the guideline For example “In couples with the diagnosis Unexplained Infertility, how can the prognosis for a spontaneous pregnancy be reliably estimated?” “In couples with UI, how can it be decided on when to start infertility treatment?”	This comment refers to a prognosis-based approach. A section was added in the treatment chapter of the guideline: III.0 When to start treatment.

Priya Bhide	/	/	Annex 6,7,8 are not available for review – it is hence impossible to evaluate/understand the GRADE/evaluation. The presented evidence and its correlation to the quality of evidence is not possible	The evidence tables and summary of evidence tables will be published together with the final version of the guideline.