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Review

Fertility Preservation: An Integrated Review of Male and Female Strategies, Techniques, Outcomes, and Future Directions

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Abstract

Background/Objectives: Fertility preservation (FP) has become an integral component of multidisciplinary care for patients at risk of treatment- or disease-related infertility. Although several established and emerging fertility preservation strategies are currently available, important challenges remain regarding patient selection, timing of intervention, long-term reproductive outcomes, and the translation of experimental approaches into routine clinical practice. This review critically appraises the current evidence on female and male fertility preservation, highlighting established techniques, emerging technologies, unresolved clinical challenges, and future research priorities. Oocyte vitrification remains the preferred strategy for postpubertal women when ovarian stimulation is feasible, whereas ovarian tissue cryopreservation has become an established option for selected patients requiring urgent treatment or for prepubertal girls. In men, sperm cryopreservation remains the standard of care, while fertility preservation in prepubertal boys is still investigational. Despite excellent laboratory outcomes, the real-world utilization of cryopreserved reproductive material remains unexpectedly low, highlighting persistent barriers in patient referral, counselling, and long-term follow-up. **Methods:** A structured narrative review was conducted using PubMed/MEDLINE and Embase to identify international guidelines, systematic reviews, meta-analyses, multicentre cohorts, registry-based studies, and original clinical investigations published between January 2015 and June 2025. Evidence was synthesized according to sex, age, fertility preservation indication, and clinical applicability. Interventions reviewed include oocyte and embryo cryopreservation, ovarian tissue cryopreservation (OTC), GnRH agonist co-treatment, ovarian transposition, in vitro maturation (IVM), sperm banking, surgical sperm retrieval (SSR), testicular tissue cryopreservation (TTC), and spermatogonial stem-cell (SSC) approaches. **Results:** Oocyte vitrification remains the preferred strategy for postpubertal women when ovarian stimulation is feasible, whereas ovarian tissue cryopreservation has become an established option for selected patients requiring urgent treatment or for prepubertal girls. In men, sperm

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cryopreservation prior to therapy is standard; SSR provides sperm for selected azoospermic cases. Paediatric FP strategies are asymmetric: OTC is clinically established, while TTC remains investigational. Random-start COS and aromatase inhibitor or tamoxifen protocols minimise treatment delay and hormonal exposure, and GnRHa co-treatment can serve as an adjunctive ovarian-protective measure. Despite high laboratory success, utilisation of banked gametes and tissue is modest, underscoring the need for structured referral pathways, patient counselling, and longitudinal follow-up. **Conclusions:** FP should be systematically integrated into both oncological and non-oncologic care pathways, with timely referral, age- and sex-specific counselling on live-birth probabilities, and long-term follow-up. Future priorities include harmonization of clinical protocols, standardized outcome reporting, registry-based long-term follow-up, and validation of emerging technologies to improve both reproductive outcomes and equitable access to fertility preservation.

Keywords: fertility preservation; oocyte vitrification; embryo cryopreservation; ovarian tissue cryopreservation; GnRH agonists; in vitro maturation; sperm cryopreservation; surgical sperm retrieval; testicular tissue cryopreservation; spermatogonial stem cells

1. Introduction

Fertility preservation (FP) is now recognized as an essential component of patient-centred care for individuals at risk of gonadotoxicity. Cancer therapies may compromise reproductive potential through direct damage to germ cells and the gonadal microenvironment, with risk influenced by patient age, therapeutic modality, cumulative dose, and radiation exposure [1–6]. Fertility counselling should balance oncological urgency with reproductive goals and convey realistic, evidence-based probabilities of future live birth [1,7,8]. The increasing survival of young cancer patients and individuals affected by chronic diseases has shifted attention from survival alone to long-term quality of life, including the preservation of reproductive potential.

Beyond oncology, FP is relevant for patients with benign haematological and metabolic disorders (e.g., thalassemias, sickle-cell disease, galactosemia), autoimmune conditions necessitating gonadotoxic agents, genetic risks of premature ovarian insufficiency (e.g., Turner syndrome, BRCA carriers considering risk-reducing salpingo-oophorectomy), severe andrological disorders (e.g., non-obstructive azoospermia), and elective deferral of childbearing. Individualized assessment of timing, modality, and eligibility within multidisciplinary pathways is critical to optimize reproductive outcomes [1,9,10].

Despite remarkable advances in fertility preservation over the past decade, several clinically relevant questions remain unresolved. The optimal selection of fertility preservation strategies according to patient age, diagnosis, treatment urgency, and reproductive goals continues to be debated. Furthermore, important uncertainties persist regarding the long-term effectiveness of ovarian and testicular tissue cryopreservation, the management of pediatric patients, the safety of emerging technologies, and the relatively low real-world utilization of cryopreserved reproductive material despite high technical success rates. These unresolved issues highlight the need for a critical appraisal of current evidence rather than a purely descriptive overview. Therefore, the aim of this review is not only to summarize currently available fertility preservation strategies but also to critically compare established and emerging approaches, discuss their clinical applicability, analyze existing controversies, identify knowledge gaps, and outline future research priorities that may improve both reproductive outcomes and equitable access to fertility preservation.

2. Methods (Narrative Review Design)

We conducted a structured narrative review to provide a comprehensive and critical synthesis of the current evidence regarding fertility preservation in female and male patients. Electronic literature searches were performed in PubMed/MEDLINE and Embase to identify relevant studies published between January 2015 and June 2025. The search strategy combined terms related to fertility preservation, including: oocyte vitrification, embryo cryopreservation, ovarian tissue cryopreservation, testicular tissue cryopreservation, sperm banking, surgical sperm retrieval, in vitro maturation, and spermatogonial stem cells, as well as broader terms capturing both oncological and non-oncological indications.

We prioritized international guidelines (ASCO, ESHRE), systematic reviews, meta-analyses, and large multicenter or registry-based studies reporting clinical outcomes. Exclusion criteria were non-human studies (except mechanistic translational work), single-case reports, and publications lacking outcome data. All included studies were critically appraised for relevance, methodological quality, and applicability to clinical decision-making. This approach enabled the review to integrate both technical and practical considerations, while highlighting gaps, ongoing debates, and areas for future research.

3. Female Fertility Preservation Strategies

3.1. Embryo Cryopreservation

Embryo cryopreservation is a well-established fertility preservation strategy for women with access to a male partner or donor sperm and remains the option with the most robust reproductive outcomes. Advances in cryobiology have led to the widespread adoption of vitrification, which minimizes ice crystal formation and has largely replaced conventional slow-freezing techniques. Consequently, vitrified blastocysts achieve post-thaw survival rates exceeding 95%, with implantation and live-birth rates comparable to those obtained after fresh embryo transfer [7–10].

The procedure requires controlled ovarian stimulation (COS), which may raise concerns in hormone-sensitive malignancies. However, modified stimulation protocols, including the use of aromatase inhibitors (letrozole) or selective estrogen receptor modulators (tamoxifen), as well as random-start COS, allow timely oocyte retrieval while limiting supraphysiologic estrogen exposure, even in patients with estrogen receptor-positive breast cancer [7–10]. Although embryo cryopreservation remains associated with the highest cumulative live-birth rates, its applicability is limited by the need for sperm availability and may raise ethical, legal, or personal concerns. Therefore, careful multidisciplinary counselling is essential to ensure that this option is aligned with the patient's reproductive goals and personal preferences.

3.2. Oocyte Cryopreservation

Oocyte cryopreservation via vitrification is currently considered the first-line fertility preservation option for postpubertal women who do not have access to sperm or prefer not to create embryos. The transition from slow freezing to vitrification has markedly improved oocyte survival, fertilization rates, and clinical outcomes, bringing them closer to those observed with fresh oocytes [7,8,10].

Nevertheless, real-world effectiveness is strongly influenced by patient age at cryopreservation and the number of mature oocytes stored, with diminishing returns in women over 35 years. Importantly, observational studies indicate that a substantial proportion of patients never return to use their cryopreserved oocytes (often <10%). These findings suggest that successful oocyte cryopreservation should not automatically be equated with successful fertility preservation, emphasizing the importance of individualized counselling

regarding realistic reproductive expectations, future family planning, and the potential underutilization of cryopreserved gametes [7,8,10].

3.3. Ovarian Tissue Cryopreservation (OTC)

Ovarian tissue cryopreservation is indicated for patients in whom controlled ovarian stimulation is contraindicated or cannot be performed, including prepubertal girls and women requiring urgent gonadotoxic therapy [2,9,10]. The technique involves surgical retrieval of ovarian cortical tissue, cryopreservation (most commonly by slow freezing), and subsequent autologous transplantation after cancer treatment [9,10].

Numerous studies have reported the restoration of endocrine function and successful spontaneous pregnancies after orthotopic transplantation [9,11]. Nevertheless, several challenges continue to limit the widespread application of this approach. Significant ischemia–reperfusion injury following transplantation may compromise follicular survival and reduce graft longevity. Furthermore, in patients with certain malignancies—particularly hematologic cancers—the risk of reintroducing malignant cells remains a major concern [2,8,10].

Reported rates of endocrine function recovery after transplantation vary widely across studies, generally ranging from approximately 60% to 90%, depending on patient age, tissue volume, cryopreservation technique, and transplantation site [4–6,8–19]. Given this heterogeneity, these figures should be interpreted cautiously, and outcomes discussed within the context of center-specific expertise and patient selection. Although more than 130 live births have been reported worldwide, OTC should still be regarded as a specialized and partially experimental strategy requiring careful risk–benefit assessment [2,8–10]. Current evidence suggests that ovarian tissue cryopreservation should be considered complementary rather than an alternative to oocyte cryopreservation. While OTC uniquely enables fertility preservation without delaying cancer treatment and offers the additional benefit of restoring endocrine function after transplantation, reproductive outcomes remain more heterogeneous and are influenced by patient selection, ovarian reserve, graft survival, surgical expertise, and center experience. Therefore, the choice between OTC and oocyte cryopreservation should be individualized within a multidisciplinary fertility preservation program.

3.4. Gonadotropin-Releasing Hormone Agonist (GnRHa) Co-Treatment

Temporary ovarian suppression with GnRH agonists during chemotherapy has been investigated as a strategy to reduce treatment-related ovarian insufficiency [2,10,15]. Evidence supports a protective effect on ovarian function, particularly in women with breast cancer [2,15]. However, evidence regarding long-term fertility and live-birth outcomes remains less robust and varies according to cancer type and treatment [2,10,15]. Therefore, GnRHa co-treatment should be considered an adjunctive strategy and should not replace established fertility preservation methods such as oocyte, embryo, or ovarian tissue cryopreservation [2,10].

3.5. Ovarian Transposition and In Vitro Maturation

Ovarian transposition may reduce radiation-induced ovarian damage in patients undergoing pelvic radiotherapy, although success depends on radiation fields and scatter dose. In vitro maturation (IVM) of oocytes represents an alternative for patients with limited time for stimulation and may be combined with OTC. However, clinical experience and live-birth data remain limited, and these approaches should be offered selectively in experienced centers [7,8,12]. An overview of the multidisciplinary decision-making pathway for fertility preservation is provided in Figure 1.

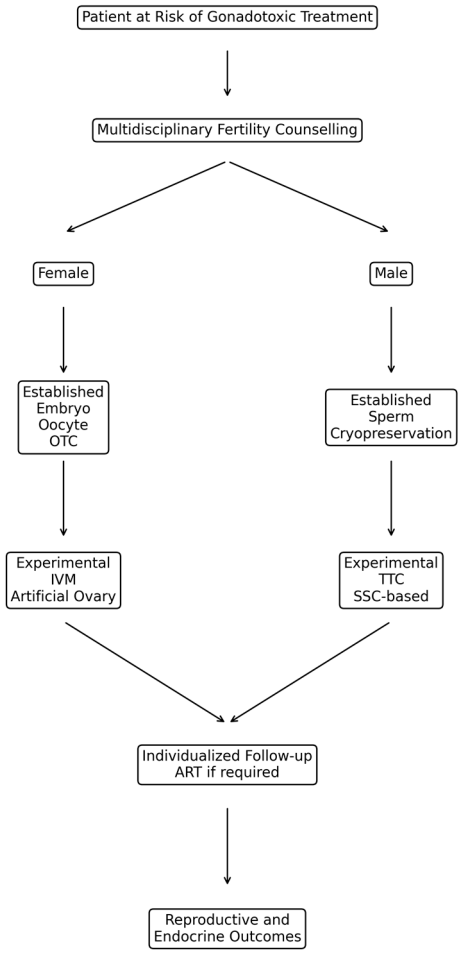


Figure 1. Schematic overview of the multidisciplinary decision-making pathway for fertility preservation in patients at risk of treatment-related infertility. The figure summarizes the established and emerging fertility preservation strategies currently available for female and male patients according to clinical setting and reproductive status.

4. Male Fertility Preservation Strategies

4.1. Sperm Cryopreservation

Sperm cryopreservation represents the reference standard for fertility preservation in postpubertal males facing gonadotoxic treatments. The technique is well established, widely available, and relies on semen collection followed by cryopreservation, most commonly using conventional slow-freezing protocols with cryoprotectants to preserve sperm viability.

Despite its technical simplicity and safety, the actual utilization rate of cryopreserved samples remains modest, with observational studies reporting usage in approximately 9% of patients. When thawed samples are used, reported pregnancy rates are around 25–30%, largely dependent on female partner factors and the assisted reproductive technique employed. Due to reduced sperm motility and viability after thawing, assisted reproduction—particularly intracytoplasmic sperm injection (ICSI)—is frequently required to achieve fertilization [1]. Collectively, these findings emphasize the importance of comprehensive pre-treatment counselling regarding both the effectiveness and realistic likelihood of future use.

4.2. Surgical Sperm Retrieval

Surgical sperm retrieval techniques, including testicular sperm extraction (TESE) and microdissection TESE (micro-TESE), may be considered in selected patients when

ejaculation is not feasible (e.g., due to obstruction or severe illness) or in cases of azoospermia. Micro-TESE, in particular, allows targeted sampling of seminiferous tubules under magnification and may improve sperm retrieval rates while minimizing tissue loss [20–22].

Therefore, these procedures require specialized surgical expertise and are associated with procedural risks, such as bleeding, infection, and potential testicular damage. Moreover, retrieved sperm typically necessitate the use of ICSI for successful fertilization. Consequently, surgical sperm retrieval should be offered selectively and preferably in experienced centers, after careful evaluation of risks, benefits, and alternative options [1].

4.3. Paediatric and Experimental Strategies

In prepubertal boys, conventional sperm cryopreservation is not possible due to the absence of mature spermatozoa. In this setting, testicular tissue cryopreservation (TTC) has emerged as an experimental strategy aimed at preserving spermatogonial stem cells prior to gonadotoxic therapy. Feasibility and short-term safety of testicular biopsy and cryopreservation have been demonstrated in pediatric and adolescent programs, with acceptable surgical morbidity and tissue preservation outcomes [4,10]. Nevertheless, no peer-reviewed publication has yet reported a successful live birth following human testicular tissue cryopreservation. However, very recent preliminary findings have suggested that the first successful fertility restoration may have been achieved, although these data are currently available only as a preprint and therefore require independent validation before they can be considered established clinical evidence [5]. Proposed future applications include autologous transplantation of testicular tissue or in vitro maturation of spermatogonial stem cells, both of which remain experimental. Significant concerns persist regarding the risk of malignant cell reintroduction, particularly in hematologic malignancies, as well as uncertainties surrounding long-term safety and efficacy. As such, TTC should currently be considered an investigational approach, offered only within ethically approved research protocols and with comprehensive counseling of patients and families [4,10].

5. Overview of Clinical Outcomes and Real-World Utilization

Across both female and male fertility preservation strategies, a substantial gap remains between technical feasibility, clinical success, and real-world use. Observational studies report that fewer than 10% of patients ultimately return to use cryopreserved reproductive material, underscoring that successful preservation does not automatically translate into parenthood. Utilization is influenced by age at preservation, survival outcomes, personal circumstances, relationship status, and access to assisted reproductive technologies.

Oocyte vitrification is associated with high post-thaw survival rates; however, cumulative live-birth probability remains strongly age-dependent, with diminishing returns in women undergoing preservation at more advanced reproductive ages. Embryo cryopreservation provides the most predictable reproductive outcomes, yet remains limited by the need for sperm availability and ovarian stimulation.

In ovarian tissue cryopreservation, restoration of endocrine function after autologous transplantation is frequently reported, but outcomes vary widely across centers and patient populations. Importantly, endocrine recovery should not be equated with reproductive success, as follicle loss due to ischemia–reperfusion injury and reduced ovarian reserve often result in assisted reproduction outcomes comparable to those observed in women with diminished ovarian reserve.

In males, sperm cryopreservation enables assisted reproduction with acceptable success rates, primarily through ICSI; however, long-term follow-up of cancer survivors is essential to assess how fertility preservation interventions are ultimately translated into reproductive decision-making and parenthood.

6. Clinical Pathways, Ethics, and Health-System Implementation

The growing complexity and diversity of fertility preservation strategies require well-structured clinical pathways, including rapid referral systems, multidisciplinary counseling, and timely coordination between oncology, reproductive medicine, and surgery. Early referral is critical to expand the range of feasible options and to allow individualized risk–benefit assessment.

Ethical considerations remain central to fertility preservation practice, particularly in pediatric patients, where issues of informed consent, assent, and future autonomy must be carefully considered. Additional ethical challenges include posthumous reproduction, long-term storage of reproductive tissues, and safety concerns related to the potential reintroduction of malignant cells following tissue transplantation.

From a healthcare system perspective, equitable access to fertility preservation is strongly influenced by cost coverage, geographic availability, and the concentration of specialized expertise. Centralization of complex and experimental procedures—such as ovarian or testicular tissue cryopreservation and surgical sperm retrieval—may improve quality, safety, and outcome monitoring, but requires coordinated referral networks and shared registries. Participation in national and international registries is essential to capture long-term oncological and reproductive outcomes, refine patient selection, and inform evidence-based guidelines [1,7–9]. The main female fertility preservation techniques, including their indications, timelines, strengths, and limitations, are summarized in Table 1.

Table 1. Female preservation techniques—indications, timelines, pros/cons.

Technique	Indications	Typical Timeline	Strengths	Limitations/Notes
Oocyte vitrification (COS)	Postpubertal; time for stimulation; many cancers incl. ER+ with letrozole	10–14 days (random-start; DuoStim optional)	Large amount of evidence; scalable; patient autonomy	Delay needed; hormone exposure; age-dependent yield [4–6,10]
Embryo vitrification	Partner/donor sperm available	As above	High survival and outcomes	Legal/ethical consent; disposition issues [4–6,10]
OTC + transplant	Prepubertal/time-critical; contraindication to COS	48–72 h (surgery + cryo); months to re-implant	Restores endocrine function; natural conception	Contamination risk; variable duration; specialised centre [5,10]
IVM ± vitrification	Very limited time; COS contra-indicated	24–72 h	Minimal delay; hormone-sparing	Lower evidence; lab-dependent outcomes [4,10]
GnRHa during chemo	Adjunct across cancer types	Concurrent with chemotherapy	Protects menses/ovarian function	Not a replacement for cryopreservation [6,10]
Ovarian transposition	Planned pelvic RT	Pre-RT (surgery)	Reduces ovarian dose	Surgical risks; may combine with cryo [10]

7. Male Fertility Preservation: Techniques and Practical Considerations

Fertility preservation in male patients is generally less time-consuming than in females but encompasses a wide spectrum of clinical scenarios, ranging from straightforward sperm cryopreservation to complex surgical and experimental approaches. Effective implementation requires awareness of procedural indications, technical limitations, and realistic reproductive outcomes, particularly in pediatric and high-risk oncological populations.

This section outlines established and emerging male fertility preservation strategies, followed by a discussion of their practical and clinical implications.

7.1. Sperm Cryopreservation: Practical and Technical Considerations

Sperm cryopreservation is the standard of care for fertility preservation in postpubertal males undergoing potentially gonadotoxic treatments. Whenever feasible, collection of two to three ejaculates is recommended prior to therapy, with 2–5 days of sexual abstinence for each sample, prioritizing preservation before the initiation of cancer treatment.

Laboratory processing typically includes semen analysis, dilution with cryoprotective media (commonly glycerol-based), controlled-rate freezing or vapor-phase cooling, and long-term storage in liquid nitrogen. Post-thaw sperm motility and viability vary considerably, and although fertilization is achievable, assisted reproductive technologies—most notably intracytoplasmic sperm injection (ICSI)—are frequently required to achieve pregnancy [1].

7.2. Surgical Sperm Retrieval and Cryopreservation

In patients unable to ejaculate or in those with azoospermia, surgical sperm retrieval (SSR) techniques may be considered prior to gonadotoxic therapy. Available approaches include testicular sperm extraction (TESE), microdissection TESE (micro-TESE), and percutaneous epididymal or testicular sperm aspiration (PESA/TESA), depending on the underlying etiology.

Micro-TESE has demonstrated improved sperm retrieval rates in non-obstructive azoospermia by enabling targeted sampling of seminiferous tubules. Retrieved sperm may be cryopreserved for future use or utilized immediately in conjunction with ICSI. These procedures carry inherent risks, including testicular injury and anesthetic complications, and should therefore be performed in experienced, high-volume centers [1].

7.3. Electroejaculation and Special Clinical Circumstances

In selected patients with neurogenic conditions or severe anejaculation, electroejaculation under anesthesia may enable sperm collection for cryopreservation. In cases of suspected retrograde ejaculation, sperm retrieval from post-ejaculatory urine following appropriate alkalinization may also be considered. These approaches remain niche but can be valuable alternatives when standard ejaculation is not possible.

7.4. Testicular Tissue Cryopreservation in Prepubertal Boys

In prepubertal boys, the absence of mature spermatozoa precludes conventional sperm cryopreservation. Testicular tissue cryopreservation (TTC) has therefore emerged as an experimental strategy aimed at preserving spermatogonial stem cells prior to gonadotoxic exposure. The procedure involves testicular biopsy, tissue fragmentation, exposure to cryoprotectants, and cryopreservation using slow-freezing or vitrification protocols, followed by storage in liquid nitrogen.

Potential future applications include autologous tissue re-implantation, spermatogonial stem cell transplantation, or in vitro spermatogenesis. However, clinical fertility restoration following TTC has not yet been achieved in humans, and no live births have been reported. Safety concerns remain substantial, particularly regarding the risk of malignant cell re-introduction and the long-term genetic and epigenetic integrity of derived gametes [4–6,10].

7.5. Experimental Restoration Strategies

Several experimental strategies aimed at restoring fertility from cryopreserved testicular tissue are under active investigation. Autotransplantation of thawed tissue has resulted

in successful spermatogenesis in animal models, but translation to human clinical practice remains cautious. Isolation, expansion, and transplantation of spermatogonial stem cells represent promising yet technically demanding approaches, requiring reliable methods for oncological decontamination. In vitro spermatogenesis and tissue-engineering or organoid-based platforms are advancing rapidly but are not yet ready for clinical application [4–6,10].

Male fertility preservation encompasses both highly effective standard-of-care interventions and emerging experimental strategies with significant translational challenges. While sperm cryopreservation offers a reliable and widely accessible option for most post-pubertal patients, surgical and tissue-based approaches are reserved for selected clinical scenarios and require specialized expertise. In prepubertal boys, fertility preservation remains investigational, underscoring the need for long-term follow-up, registry-based outcome reporting, and continued ethical oversight. Together, these considerations highlight the importance of individualized counseling and centralized care pathways to ensure safe and meaningful fertility preservation for male patients. The main male fertility preservation techniques, including their indications, timelines, strengths, and limitations, are summarized in Table 2.

Table 2. Male preservation techniques—indications, timelines, pros/cons.

Technique	Indications	Typical Timeline	Strengths	Limitations/Notes
Sperm cryopreservation	Postpubertal; before gonadotoxic therapy	Same-day; repeated over 2–5 days	Established; widely available	Quality may be poor at baseline; low utilisation (~9%) [1]
SSR (TESE/micro-TESE)	Azoospermia/ non-ejaculation	Day-case surgery	Enables ICSI; option when ejaculation fails	Invasive; centre-dependent outcomes [1]
Electroejaculation	Neurogenic/ anejaculation	Same-day under anaesthesia	Allows collection when other methods fail	Resource-intensive; requires anaesthesia
TTC (prepubertal)	Boys prior to high-risk therapy	48–72 h (biopsy + cryo)	Captures SSCs for future options	Experimental; no human live births to date [7–9]
SSC-based/IVS (experimental)	Research settings	N/A	Potential definitive restoration	Oncological safety unproven; regulatory barriers [7–9]

8. Implementation of Fertility Preservation: Referral Pathways, Ethical Considerations, and Quality Assurance

The effective delivery of fertility preservation requires more than technical availability of cryopreservation strategies. Given the time-sensitive nature of oncological care, the heterogeneity of patient populations, and the variable efficacy of available interventions, fertility preservation must be embedded within structured clinical pathways that integrate rapid referral, ethical governance, and systematic outcome monitoring. The following sections outline key procedural components and conclude with considerations for implementation at the health-system level.

8.1. Triage and Rapid Referral Pathways

Timely access to fertility preservation is critically dependent on early identification and rapid referral from oncology to reproductive medicine. Same-day or expedited referral protocols should be established to minimize delays and expand the range of feasible preservation options. Counseling should be comprehensive and individualized, addressing

treatment-specific gonadotoxic risks, age- and sex-appropriate preservation strategies, procedural timelines, and realistic probabilities of future live birth rather than procedural success alone.

Standardized triage tools and checklists may facilitate documentation of eligibility, informed consent, and logistical requirements, including laboratory capacity and surgical scheduling for procedures such as ovarian or testicular tissue cryopreservation. Such structured approaches help ensure consistency, transparency, and equitable access across patient populations.

8.2. Ethics, Consent, and Legal Considerations

Ethical and legal issues are integral to fertility preservation, particularly in pediatric and adolescent patients. In minors, decision-making requires a balance between assent from the patient and consent from parents or legal guardians, with careful consideration of future autonomy. Additional ethical challenges include ownership of gametes and tissues, posthumous reproduction, duration of storage, and cross-border transfer of cryopreserved material.

Patients should receive clear counseling regarding disposition options (future use, donation, or disposal), the current limitations of long-term safety data for offspring, and disease-specific risks, including the potential reintroduction of malignant cells following tissue re-implantation. These discussions should be documented and revisited as clinical circumstances evolve.

8.3. Data Collection, Quality Assurance, and Registries

To move beyond proof-of-concept toward evidence-based practice, fertility preservation programs should participate in national and international registries and systematically report standardized outcome measures. Key performance indicators include collection yield, post-thaw survival, endocrine recovery after tissue transplantation, pregnancy rates, and live-birth outcomes.

Long-term survivorship follow-up is essential to assess not only biological success but also real-world effectiveness, including utilization rates and reproductive decision-making over time. Registry-based data are critical to refining patient selection, benchmarking outcomes across centers, and informing clinical guidelines [1–10,12].

8.4. Cost, Access, and Equity

Access to fertility preservation remains highly variable and is strongly influenced by cost coverage, geographic availability, and the concentration of specialized expertise. Strategies to improve equity include early referral, streamlined scheduling, travel support, and public or insurance-based funding where available.

Centralization of complex or experimental procedures—such as ovarian and testicular tissue cryopreservation—may enhance quality, safety, and outcome monitoring. However, this approach requires robust referral networks and coordinated care pathways to avoid exacerbating disparities.

Incorporating fertility preservation into routine oncological care demands coordinated clinical pathways, ethical oversight, and system-level commitment to data collection and equitable access. Structured referral processes and centralized expertise can improve quality and safety, while registry participation is essential to close existing knowledge gaps regarding long-term effectiveness. Together, these elements are critical to ensuring that advances in fertility preservation translate into meaningful reproductive outcomes for cancer survivors.

9. Indications for Fertility Preservation: Clinical Context and Ongoing Debates

Fertility preservation encompasses a clinically and etiologically heterogeneous spectrum of indications, reflecting the expanding populations exposed to actual or anticipated gonadotoxic risk. In oncology, fertility preservation is primarily intended to mitigate treatment-related gonadal damage associated with chemotherapy, radiotherapy, targeted therapies, and immunotherapies, while respecting the urgency and priorities of cancer care. However, the degree of gonadotoxicity varies widely according to treatment regimen, cumulative dose, and patient-specific susceptibility, making risk stratification and timing of intervention central—and often debated—elements of clinical decision-making.

Beyond cancer, fertility preservation has assumed growing relevance in patients with benign hematologic, metabolic, autoimmune, and genetic conditions. In these settings, gonadal impairment may arise from chronic disease burden, repeated cytotoxic exposure, or prophylactic surgical interventions, as in individuals with BRCA pathogenic variants undergoing risk-reducing salpingo-oophorectomy. Elective fertility preservation for age-related fertility decline further broadens the indication spectrum, raising important questions regarding cost-effectiveness, realistic outcome expectations, and appropriate patient selection. Across all indications, the principal clinical challenge lies not in the availability of techniques, but in matching the right modality to the right patient at the right time while balancing reproductive benefit, medical risk, and ethical considerations [1–19,23,24].

In women, oocyte vitrification is generally considered the preferred option when sufficient time for ovarian stimulation is available and when hormonal exposure is deemed oncologically safe. Ovarian tissue cryopreservation is reserved for situations in which stimulation is contraindicated, impractical, or impossible, including prepubertal patients, but remains the subject of ongoing debate due to variability in reproductive outcomes and safety concerns related to tissue reimplantation. In men, sperm cryopreservation prior to gonadotoxic therapy represents a well-established standard of care, whereas surgical sperm retrieval is limited to selected clinical scenarios and requires specialized expertise. In prepubertal boys, testicular tissue cryopreservation remains investigational and is currently justified only within ethically approved protocols, reflecting the absence of proven fertility restoration in humans to date.

Elective fertility preservation represents a distinct and particularly debated indication, as it is driven not by imminent medical risk but by anticipated age-related fertility decline. This scenario necessitates transparent counseling regarding age-dependent success rates, likelihood of utilization, and financial implications to ensure that patient expectations align with achievable outcomes. Taken together, these diverse indications underscore the need for individualized, context-aware fertility preservation counseling that integrates medical evidence, ethical responsibility, and long-term reproductive goals [7–9,11,12,18,19,23,25].

10. Technical Principles—From Gametes to Gonadal Tissue

Cryopreservation represents the technological backbone of fertility preservation, yet its successful application depends on a delicate balance between biophysical principles, cellular complexity, and clinical objectives. Across all reproductive materials, the primary challenge is to prevent intracellular ice formation and osmotic injury during cooling and warming, processes that can irreversibly compromise cellular integrity. However, the optimal cryopreservation strategy varies substantially between gametes, embryos, and gonadal tissue, reflecting differences in size, water content, membrane permeability, and organizational complexity.

Two principal cryopreservation approaches are currently employed: slow freezing and vitrification. Slow freezing relies on controlled-rate cooling combined with gradual

exposure to cryoprotectants, allowing cellular dehydration and reducing ice crystal formation. Vitrification, by contrast, achieves ultra-rapid cooling in the presence of higher concentrations of permeating and non-permeating cryoprotectants, inducing a glass-like solid state that effectively eliminates ice formation. In oocyte and embryo cryopreservation, vitrification has become the dominant method due to consistently superior post-warming survival, fertilization potential, and clinical outcomes. Nevertheless, vitrification is technically demanding and raises ongoing debates regarding cryoprotectant toxicity, operator dependency, and the use of open versus closed systems [7,9,11,12,23].

In ovarian tissue cryopreservation, slow freezing remains the most widely validated approach, supported by the majority of reported clinical pregnancies and live births following tissue transplantation. The preservation of tissue architecture, stromal components, and follicular niches poses challenges that differ fundamentally from those encountered in single-cell cryopreservation. Although vitrification of ovarian tissue is under active investigation and shows promising experimental results, comparative clinical data remain limited, and no consensus has yet emerged regarding its equivalence or superiority to slow freezing. Testicular tissue cryopreservation protocols are largely adapted from ovarian models but require additional optimization, given differences in tissue composition, stem cell distribution, and biopsy handling, particularly in prepubertal patients [7,9,11,12,23].

Beyond the cryopreservation technique itself, laboratory quality management systems are critical determinants of safety and reproducibility. Comprehensive documentation should include cryoprotectant lot traceability, device and protocol validation (including the choice between open and closed vitrification systems), liquid nitrogen management, and a robust chain-of-custody from informed consent through long-term storage and potential future use. Traceability assumes particular importance in tissue banking, where multiple tissue fragments may be stored per patient, thawed sequentially, and re-implanted over extended periods, often spanning many years. These requirements highlight the need for standardized laboratory governance frameworks to support both clinical reliability and regulatory compliance [9,11,23,26,27].

11. Female Procedures—Protocol Details and Practical Considerations

Fertility preservation procedures in women are characterized by a high degree of technical sophistication and require careful alignment between oncological timelines, reproductive potential, and procedural risk. Unlike male fertility preservation, most female strategies are time- and hormone-dependent, making protocol selection and counseling central to clinical decision-making. This section outlines current procedural approaches while highlighting areas of ongoing debate and practical complexity.

Controlled ovarian stimulation (COS) represents the cornerstone of oocyte and embryo cryopreservation and is most commonly performed using gonadotropin-releasing hormone (GnRH) antagonist protocols, which allow flexibility and facilitate the use of random-start initiation. Random-start COS has become standard practice in the fertility preservation setting, as it minimizes treatment delays without compromising oocyte yield. In patients for whom estrogen exposure is a concern—most notably those with hormone-sensitive breast cancer—stimulation protocols incorporating aromatase inhibitors (letrozole) or, less commonly, selective estrogen receptor modulators (tamoxifen) are widely adopted. Although these regimens effectively limit peak estradiol levels, debate persists regarding their impact on oocyte competence and long-term oncological safety, underscoring the need for individualized risk–benefit assessment [7–9,11,14,16,18,19,23–25].

Final oocyte maturation is typically induced using a GnRH agonist trigger, alone or in combination with low-dose human chorionic gonadotropin, to reduce the risk of ovarian hyperstimulation syndrome. Following transvaginal oocyte retrieval, cumulus–oocyte

complexes are denuded, metaphase II oocytes are identified, and vitrification is performed. When a sperm source is available and embryo creation is acceptable to the patient, embryo cryopreservation—most often at the blastocyst stage—is frequently pursued, reflecting its more predictable reproductive outcomes. However, this option introduces additional ethical, legal, and relational considerations that must be addressed during counseling.

For patients with extremely limited time or contraindications to conventional stimulation, in vitro maturation (IVM) offers an alternative approach, allowing retrieval of immature oocytes with subsequent laboratory maturation and vitrification. While IVM reduces treatment delay and hormonal exposure, its clinical effectiveness remains lower than that of conventional COS, and its use is largely confined to selected scenarios and experienced centers.

Ovarian tissue cryopreservation (OTC) involves laparoscopic harvesting of cortical ovarian tissue, immediate exposure to cryoprotectants, and cryopreservation—most commonly via slow-freezing protocols. Prior to any autologous transplantation, careful screening for malignant infiltration is essential, and re-implantation is typically deferred until oncological remission is achieved and reproductive goals are clearly defined. Although restoration of endocrine function and spontaneous pregnancies have been reported, reproductive outcomes remain variable, and the procedure continues to raise concerns related to ischemic follicle loss and oncologic safety [9,11,12,23,24,26,27].

Comprehensive counseling is therefore integral to female fertility preservation. Patients should be informed not only about procedural steps and technical success rates, but also about age-dependent probabilities of future live birth, the distinction between gamete or tissue banking and reproductive outcomes, and the potential need for assisted reproduction even after successful tissue re-implantation. Explicit documentation of consent regarding long-term storage and disposition of cryopreserved material (use, donation, or disposal) is essential. Together, these considerations emphasize that fertility preservation in women is not a single intervention, but a longitudinal clinical process requiring anticipatory guidance, ethical clarity, and ongoing follow-up.

12. Male Procedures—Protocol Details and Practical Considerations

Fertility preservation in male patients ranges from straightforward sperm cryopreservation in postpubertal men to complex, investigational interventions in prepubertal boys, each requiring careful alignment of procedural timing, patient characteristics, and long-term reproductive goals. Clinical success depends not only on laboratory technique but also on individualized counseling, ethical oversight, and structured follow-up.

Sperm cryopreservation remains the standard of care for postpubertal males facing gonadotoxic therapies. Optimal collection involves two to three ejaculates, spaced 2–5 days apart, prior to initiation of treatment whenever feasible. Semen analysis establishes baseline parameters, followed by addition of cryoprotectants (typically glycerol-based) and controlled-rate freezing or vapor-phase cryopreservation, with long-term storage in liquid nitrogen. Post-thaw sperm motility and viability vary considerably, often necessitating assisted reproduction, particularly intracytoplasmic sperm injection (ICSI), to achieve fertilization [1].

In patients unable to ejaculate or in those with azoospermia, surgical sperm retrieval (SSR) techniques—including testicular sperm extraction (TESE), micro-TESE, and percutaneous epididymal/testicular aspiration (PESA/TESA)—can provide sperm for cryostorage or immediate use in assisted reproduction. Micro-TESE offers higher retrieval rates in non-obstructive azoospermia but requires surgical expertise and carries procedural risks. Both ejaculatory and surgical collections must be integrated into a quality-controlled laboratory workflow, with attention to traceability, cryoprotectant handling, and post-thaw evaluation.

For prepubertal boys, conventional sperm banking is not possible, and fertility preservation relies on testicular tissue cryopreservation (TTC). This procedure involves testicular biopsy, tissue fragmentation, cryoprotectant equilibration, and controlled-rate freezing. While preclinical models demonstrate potential for autologous tissue transplantation, spermatogonial stem cell (SSC) transplantation, and in vitro spermatogenesis, human fertility restoration remains investigational. To date, no peer-reviewed publication has reported a successful live birth following human testicular tissue cryopreservation. However, preliminary data suggesting the first successful fertility restoration have recently been reported as a preprint and await independent peer review before clinical confirmation [4–6,10,28,29]. Clinical implementation therefore requires enrollment in ethically approved research protocols, meticulous tissue handling to minimize malignant contamination, and long-term follow-up to assess safety and efficacy.

Ethical and logistical considerations are central to male fertility preservation. In minors, structured frameworks must address consent and assent, tissue ownership, and long-term storage. Programs must implement safeguards against malignant contamination during tissue re-implantation and provide transparent counseling on the experimental nature of investigational interventions. Centralization of services enhances procedural expertise, laboratory quality, and the capacity for longitudinal monitoring, all of which are essential for safe and effective male fertility preservation [3–6,10,28,29].

13. Outcomes and Real-World Effectiveness

The success of fertility preservation should be evaluated using clinically meaningful outcomes rather than laboratory survival metrics alone, with live birth representing the most relevant endpoint. These outcomes are influenced by multiple interrelated factors, including patient age at cryopreservation, quantity and quality of gametes, embryos, or tissue preserved, and future reproductive context, such as partner availability and use of assisted reproductive technologies (ART).

Oocyte vitrification demonstrates consistently high post-warming survival and fertilization potential; however, cumulative live-birth probability remains strongly age-dependent, and observational studies indicate that a substantial proportion of patients never return to use stored oocytes. Ovarian tissue cryopreservation (OTC) frequently restores endocrine function, enabling spontaneous conception in some cases and assisted reproduction in others. However, reproductive outcomes remain highly variable, reflecting differences in ischemia-related follicular loss, previous chemotherapy exposure, patient characteristics, and tissue processing protocols [30].

In men, sperm cryopreservation provides a reliable option for future assisted reproduction, but uptake is similarly modest. Post-thaw fertilization frequently requires ICSI, and long-term reproductive success depends on patient engagement, oncological recovery, and partner availability. Across sexes, real-world utilization of banked material rarely exceeds 10%, reflecting the interplay of oncological timing, personal circumstances, and complex decision-making regarding family planning [1,7–9,11,12,19,23–27,31–36].

Pediatric fertility preservation warrants particular caution. In selected girls, OTC has established clinical pathways, with reports of endocrine recovery and spontaneous pregnancies; however, live-birth outcomes remain limited and dependent on long-term follow-up [37]. In boys, testicular tissue cryopreservation (TTC) remains investigational, with no confirmed human live births to date. These pediatric interventions emphasize the need for ethically governed protocols, centralized expertise, and comprehensive registry participation to monitor long-term outcomes, assess offspring safety, and refine counseling strategies [3–6,9,10,12,23,26,27,32–34,38–40].

Taken together, these observations underscore that laboratory success does not automatically translate into reproductive success. Fertility preservation should therefore be framed as a probabilistic intervention, with counseling grounded in evidence-based expectations, individualized patient factors, and transparent discussion of uncertainties. Registries and long-term follow-up remain essential for improving outcome prediction, guiding clinical decision-making, and ensuring the safe translation of fertility preservation into meaningful reproductive outcomes.

14. Service Models, Quality, and Equity

The delivery of effective fertility preservation (FP) services requires more than technical capability; it depends on the organization of care, integration within oncology pathways, and equitable access across diverse patient populations. Timely referral from diagnosing teams to reproductive specialists is critical to ensure that patients can access appropriate interventions without delaying oncological treatment. Integrated multidisciplinary counselling ensures that decisions are informed by individualized risk assessment, procedural feasibility, and realistic reproductive expectations.

High-quality FP programs should define, monitor, and regularly evaluate key performance indicators (KPIs), including time from referral to intervention, collection yield, cryosurvival rates, endocrine recovery following tissue transplantation, and reproductive outcomes such as pregnancy and live birth rates. Participation in national and international registries is essential to standardize reporting, facilitate benchmarking, and support continuous improvement, while also enabling long-term surveillance of patient and offspring outcomes [1–10,24,26,27,31,38].

Equity considerations remain central to FP service delivery. Coverage of procedural and storage costs, geographic accessibility, and support for vulnerable populations—including adolescents, patients facing urgent or high-risk treatment, and socioeconomically disadvantaged groups—are critical to ensure fair access. Centralization of specialized procedures, such as ovarian or testicular tissue cryopreservation, can improve quality, safety, and outcome monitoring, but must be balanced against potential barriers to care, including travel burden and regional disparities. Designing fertility preservation services that are both efficient and equitable is therefore essential to maximize clinical effectiveness while ensuring ethical and patient-centred care. A comprehensive summary of the current fertility preservation strategies and their clinical status, advantages, and limitations is provided in Table 3.

Table 3. Summary of current fertility preservation strategies.

Current Clinical Status	Main Limitations	Main Advantages	Target Population	Strategy
Established	Requires ovarian stimulation and sperm availability; may delay cancer treatment	Highest cumulative live-birth rates; well-established technique	Postpubertal women with a partner or donor sperm	Embryo cryopreservation
Established	Requires ovarian stimulation; lower utilization rates than expected	Standard first-line option; no need for partner; excellent survival after vitrification	Postpubertal women	Oocyte cryopreservation

Table 3. *Cont.*

Current Clinical Status	Main Limitations	Main Advantages	Target Population	Strategy
Established in selected indications	Surgical procedure; heterogeneous outcomes; potential risk of malignant cell reintroduction	No treatment delay; restores fertility and endocrine function	Prepubertal girls and women requiring urgent treatment	Ovarian tissue cryopreservation (OTC)
Adjunctive strategy	Does not replace cryopreservation techniques; uncertain effect on live birth	May reduce risk of premature ovarian insufficiency	Premenopausal women receiving chemotherapy	Gonadotropin-releasing hormone agonists (GnRHa)
Experimental/ Emerging	Limited evidence; available only in specialized centers	No treatment delay; avoids ovarian stimulation	Selected women unable to undergo ovarian stimulation	In vitro maturation (IVM)
Established	Requires semen collection; utilization rates remain relatively low	Simple, inexpensive, highly effective	Postpubertal males	Sperm cryopreservation
Established	Surgical procedure; limited to selected patients	Allows sperm retrieval when ejaculation is not possible	Men with azoospermia	Surgical sperm retrieval (TESE/micro-TESE)
Experimental	Still experimental; no peer-reviewed live birth reported to date	Only available fertility preservation option	Prepubertal boys	Testicular tissue cryopreservation (TTC)

15. Discussion

An integrated, sex-specific approach is essential for effective fertility preservation, yet achieving this in clinical practice requires careful coordination, evidence-based decision-making, and acknowledgment of remaining uncertainties. In women, oocyte vitrification remains the first-line strategy when ovarian stimulation is feasible and oncologically safe, offering high post-thaw survival and reproducible reproductive outcomes. Ovarian tissue cryopreservation (OTC) provides an alternative when stimulation is contraindicated, time-limited, or impossible, with the potential to restore endocrine function and enable spontaneous or assisted conception. However, outcomes after OTC are heterogeneous, influenced by ischemic follicle loss, patient age, prior therapy, and center-specific protocols, highlighting the need for standardized approaches and transparent counseling regarding probabilistic success.

In men, sperm cryopreservation is technically straightforward, highly effective, and widely available, yet utilization remains modest. Despite these advantages, real-world utilization remains surprisingly low. This discrepancy highlights the importance of proactive counselling, timely referral, and structured survivorship pathways to maximize the likelihood that cryopreserved sperm will ultimately contribute to successful reproductive outcomes. Surgical sperm retrieval provides critical options for azoospermic or non-ejaculatory patients, while prepubertal testicular tissue cryopreservation remains investigational, emphasizing the need for ethically governed research programs and long-term monitoring of safety and efficacy.

Pediatric fertility preservation illustrates a broader asymmetry in the field: OTC pathways are increasingly established and supported by clinical experience, whereas

testicular tissue preservation is still experimental. This gap highlights both a need for continued technological development and the ethical responsibility to balance potential benefits with uncertain long-term outcomes.

Across sexes and age groups, priority gaps remain. These include the standardization of stimulation, retrieval, and cryopreservation protocols, the systematic collection of registry-based real-world data, and long-term follow-up of offspring to ensure both reproductive success and safety. Integrating these elements within a centralized, multidisciplinary service model—which accounts for ethical, social, and equity considerations—will be crucial to translating technical advances into improved, predictable reproductive outcomes for all patient populations [1–10].

16. Conclusions

Fertility preservation is a time-sensitive, integral component of comprehensive cancer and high-risk care, requiring early identification, multidisciplinary coordination, and individualized counseling. In women, oocyte vitrification and ovarian tissue cryopreservation (OTC) represent complementary strategies, with selection guided by patient age, ovarian reserve, timing relative to therapy, and oncological considerations. While oocyte vitrification offers predictable outcomes when stimulation is feasible, OTC extends fertility and endocrine preservation to patients with limited time or prepubertal status, albeit with variable reproductive success and ongoing safety considerations.

In men, sperm cryopreservation remains highly effective and technically straightforward, yet underutilization highlights the need for proactive referral, structured survivorship pathways, and patient engagement. Surgical retrieval methods expand options for azoospermic or non-ejaculatory patients, while testicular tissue cryopreservation (TTC) in prepubertal boys remains investigational, emphasizing the ethical imperative to balance potential benefits with uncertain long-term outcomes.

Across sexes and age groups, the field continues to confront critical gaps. Standardization of stimulation, retrieval, and cryopreservation protocols, participation in registry-based outcome monitoring, and long-term follow-up of offspring are essential to translate laboratory potential into meaningful real-world reproductive outcomes. Moreover, ensuring equitable access, ethical governance, and patient-centered counseling are fundamental to maximizing the clinical impact of fertility preservation strategies.

Ultimately, fertility preservation should be regarded not simply as a laboratory procedure but as an integral component of multidisciplinary patient care. Continued technological innovation, together with standardized clinical pathways, long-term outcome monitoring, and coordinated healthcare delivery, will be essential to narrow the gap between technical success and meaningful reproductive outcomes for patients facing treatments that threaten their future fertility [1–10].

17. Research Agenda and Future Directions

Despite substantial advances in fertility preservation, several key research priorities remain. Standardization of ovarian and testicular tissue cryopreservation (OTC/TTC) protocols is essential to reduce outcome variability and facilitate cross-center comparisons. Optimizing pediatric indications, including defining eligibility, timing, and ethical safeguards, remains a critical challenge, particularly given the investigational nature of testicular tissue preservation and the long horizon to potential reproductive use.

Technological innovation is also a major focus. Oncological decontamination strategies for gonadal tissue, advancement of spermatogonial stem cell (SSC) culture, and development of in vitro spermatogenesis platforms could expand reproductive options for prepubertal patients and those with high-risk malignancies. Parallel efforts are needed to generate

robust long-term safety data, including systematic follow-up of offspring conceived after fertility preservation, to ensure both reproductive success and ethical accountability.

From a clinical implementation perspective, research should address decision-support tools and realistic, age-calibrated prognostic models to improve individualized counseling, patient understanding of probabilistic outcomes, and appropriate utilization of fertility preservation resources. Integration of these tools could help bridge the gap between technical feasibility and meaningful reproductive success, fostering informed, patient-centered care [3–7,9–12,23–29,32–36].

18. Limitations of the Evidence and of This Review

The evidence base for fertility preservation is heterogeneous, largely observational, and rapidly evolving, which poses several limitations. Selection biases, differences in patient populations, and variation in laboratory protocols complicate cross-study comparisons. Outcome reporting is inconsistent across centers and registries, and definitions of success—such as live-birth probability versus laboratory survival—are not harmonized.

Furthermore, the rapid pace of technological innovation and the emergence of experimental strategies (e.g., testicular tissue cryopreservation, in vitro spermatogenesis) necessitate continuous evidence updates. While this review synthesizes high-quality sources and clinical guidelines, it remains constrained by the underlying limitations of the primary literature. Moving forward, the field would benefit from harmonized definitions, core outcome sets, prospective data capture, and registry-based real-world monitoring, which would sharpen clinical inferences, enable benchmarking, and support evidence-based counseling and policy decisions [1–12,18,23–27,29,32–36].

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