Coping with Sociopsychological Challenges in MRKH: Progress in Uterine Transplantation and Uterus Tissue Engineering Approaches

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1.0 Abstract

Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome is a disorder of sex development affecting 1 in 5000 cisgender females with a typical female karyotype (46, XX). MRKH is characterized by the absence or underdevelopment of the uterus and upper portion of the vagina, and it poses significant challenges to affected individuals seeking to conceive and lead fulfilling lives. This literature review first navigates the multifaceted dimensions of MRKH research, addressing diagnostic intricacies, genetic and molecular insights, clinical implications, available treatments, and the profound psychosocial impacts on quality of life. Furthermore, the review critically analyses the heterogeneity in psychological results observed in MRKH studies, discussing the varied impacts on mental well-being. The review also incorporates qualitative data derived from interviews conducted with individuals who have personal or professional connections to the MRKH condition. Then, the review delves into the complexities surrounding vaginal reconstruction procedures, ethically evaluating Uterine Transplantation (UTx) and uterine Tissue Engineering (TE) studies, emphasizing the need for a comprehensive understanding of the psychological implications of these interventions. Hence, through a holistic examination of the clinical, psychological and ethical aspects, this review aims to contribute to a more comprehensive understanding of MRKH syndrome and paves the way for improved interventions and support for those navigating its challenges.

1.1 Layman Summary

Worldwide, 1 in 5000 cisgender females are affected by the Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome, a genetic condition that prevents the normal development of the uterus and upper vagina, making it challenging to conceive or bear children. In this literature review, various aspects of MRKH are explored from the diagnostic challenges to the genetic and molecular insights, clinical impacts, available treatments, and the significant psychological toll on quality of life. Qualitative data obtained from interviewing people related personally or medically by the condition, is added to enhance the urgency of certain syndrome-related aspects. Furthermore, this review critically analyzes the different results from psychological MRKH studies by further emphasizing the emotional challenges faced by the affected individuals. Additionally, it discusses the complexities of vaginal reconstruction procedures and ethical considerations around interventions like Uterine Transplantation (UTx) and Tissue engineering (TE).

By bringing together existing research, this review emphasizes the urgent need for more investigation into the MRKH syndrome. It aims to increase awareness of the profound consequences for those affected and contribute to a better clinical, psychological and societal understanding of this condition. Through this exploration, we hope to pave the way for improved support and interventions for individuals navigating the complexities of MRKH syndrome.

2.0 Relevance of the Study

Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome is a genetic condition that presents as the absence or underdevelopment of the uterus and upper portion of the vagina, resulting in the inability to conceive or bear children (Chen et al., 2022). This rare syndrome is estimated to affect 1 in 5000 cisgender females with a typical female karyotype (46, XX) (Tsarna et al., 2022). Studies have highlighted the profound psychological, quality of life, and sexual function challenges faced by individuals living with MRKH syndrome (Tsarna et al., 2022). In light of these complexities, this literature review aims to explore the multifaceted relevance of MRKH research. The review will delve into various aspects of this syndrome, including diagnostic challenges, genetic and molecular insights, clinical implications, available treatment options, impact on quality of life, and indispensable need for psychosocial support. A critical analysis is performed on the most recent psychological studies available, engaging in a comprehensive exploration of the psychosocial implications for MRKH individuals concerning diverse fertility options. This includes an in-depth examination of the psychological impact of the latest advancements in Uterine Transplantation (UTx) and recent developments in uterine Tissue Engineering (TE) studies. Furthermore, qualitative data obtained from interviews *(see section 7.0)* is incorporated to target the most needed proactive measures on specific subjects. By examining the existing body of research across these dimensions, our aim is to underscore the pressing need for further investigation into this rare syndrome and highlight its profound consequences for the affected individuals.

2.1 MRKH Syndrome: an Overview

MRKH syndrome is classified as a disorder of sex development, which is also referred to as Müllerian Aplasia, Vaginal agenesis and Müllerian agenesis (Gilfillan & Carter, 2023). The syndrome is classified into MRKH type I and type II (Chen et al., 2022). MRKH type I accounts for 69.6% of MRKH individuals and is characterized by isolated uterovaginal aplasia, whereas the remaining 30.4% belong to type II, which is also associated with extragenital malformations such as in the kidneys, skeleton and heart (Chen et al., 2022). As depicted in *Figure 1*, components of the female reproductive tract, including the uterine tube, uterus, cervix, ovaries and upper two-thirds of the vagina, originate from the paramesonephric (Müllerian) ducts (PMD) (Herlin et al., 2020). Around the 5th to 6th week of pregnancy PMDs start forming under the guidance of mesonephric (Wolffian) ducts. Following female-specific signals, the Wolffian ducts degenerate, and due to the absence of the anti-Müllerian hormone, Müllerian ducts develop and differentiate into the uterus (Herlin et al., 2020). The uterus is an organ responsible for fundamental functions such as implantation, gestation, menstruation, and labor. Accordingly, as seen in *Figure 1*, it is composed of three main layers, the perimetrium (outermost layer), myometrium and, the endometrium (innermost layer) (Habiba et al., 2020). The three layers of the uterus are distinctly structured to fulfill different functions: the perimetrium serves as a serous membrane, the myometrium is composed of smooth muscle tissue, and the endometrium functions as a mucous membrane (Habiba et al., 2020).

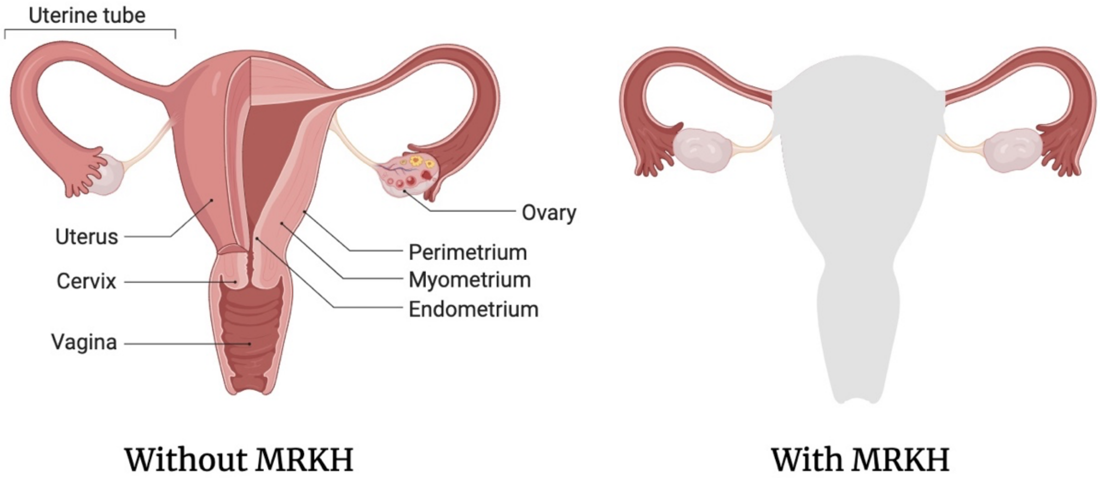

Figure 1: *Anatomical Representation of the Female Reproductive System: contrasting individuals without MRKH syndrome (depicted in natural colors) with those affected by MRKH (depicted in grey the missing or underdeveloped parts of the uterus and upper vagina) (Created with Biorender).*

Moreover, the intricacies of the endometrium, still not fully understood, unfold through its two essential layers portrayed in *Figure 2*. The upper stratum, known as the endometrial functionalis layer and populated by the lumen epithelium, stromal cells and uterine glands, serves as the site for embryo implantation and undergoes regular shedding during menstruation (Tempest et al., 2021). The lower stratum, the basalis layer, contains stem cells to restore the uterine glands that play a pivotal role in regenerating the functionalis layer (Tempest et al., 2021).

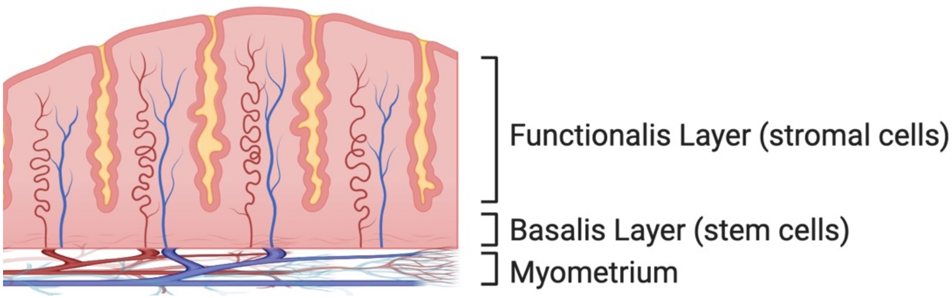

Figure 2: *Graphical Representation of the Endometrial Layers (Created with Biorender).*

Hence, MRKH arises from anomalies during the uterus embryogenesis, manifesting as either complete agenesis or aplasia of the PMDs (Herlin et al., 2020). Consequently, one of the hallmark features of MRKH is the absence of menstruation, leading to a diagnosis usually around the age of 15 years due to primary amenorrhea (Gilfillan & Carter, 2023). Beyond this primary symptom, MRKH is also characterized by an inability to engage in penile-vaginal intercourse without treatment, and inability of natural pregnancy and childbirth (Gilfillan & Carter, 2023). The aetiopathogenesis of MRKH is still not fully understood, primarily due to its intrinsic heterogeneity and complex interplay of various factors (Chen et al., 2022). Most MRKH cases occur sporadically, prompting a hypothesis centered on two pivotal factors that interact during embryonic development: environmental (epigenetic) and genetic causes (Chen et al., 2022). In terms of epigenetic factors, correlations have been drawn between MRKH and the use of drugs such as diethylstilbesterol, which was utilized to prevent miscarriages, and thalidomide, which was prescribed for morning sickness treatment (Chen et al., 2022). Even though this syndrome prevents genetic mutations from being passed vertically via females from one generation to the next, the genetic role in MRKH syndrome might be undervalued or not fully recognized. Based on recent research that has compiled data on the most commonly observed genetic anomalies linked to MRKH syndrome, the most frequently identified chromosomal regions and potentially implicated genes include 1q21.1 (*RBM8A* gene), 1p31-1p35 (*WNT4* gene), 7p15.3 (*HOXA* gene cluster), 16p11 (*TBX6* gene), 17q12 (*LHX1* and *HNF1B* genes), 22q11.21, and Xp22 (Triantafyllidi et al., 2022). However, further research is imperative to gain a more comprehensive understanding of the specific causes and underlying mechanisms of this condition.

Addressing the challenges posed by vaginal agenesis, characterized by the absence of a fully developed vagina, different minimally invasive and surgical methods are currently being implemented. One non-surgical approach is Frank's method, also known as self-dilation. It involves the use of progressive dilators multiple times a day for 20-30 minutes, and the procedure typically spans for several months (Herlin et al., 2020). Another non-surgical approach is d'Alberton's method, which involves dilation through sexual intercourse (Herlin et al., 2020). However, it is important to note that this approach may not be suitable for all MRKH individuals. Surgical procedures use different types of autografts such as split-skin graft, bowel graft, peritoneal graft and labia majora flaps (Herlin et al., 2020). In addition, recent advancements in the field have explored the use of tissue-engineered biomaterials in vaginoplasties, aiming to address the limitations associated with autografts, including compatibility issues, rejection, and the prevention of cross-talk between signaling pathways from other cell types (Sueters et al., 2023). Biodegradable or synthetic nonabsorbable grafts can be implemented (Sueters et al., 2023). As an example, Zhang et al., (2017) performed a less invasive vaginoplasty in MRKH patients by implementing a tissue-engineered biological mesh derived from animal’s dermal tissues. The findings from this study demonstrated an anatomical success rate of 100%, with 80% of the patients expressing satisfaction with the sexual functional outcomes.

3.0 Sociopsychological Impact of Individuals with MRKH Syndrome

By looking at the aforementioned traits that characterize MRKH, it is imaginable the social and psychological implications these features may have on affected individuals. Research comparing psychological evaluations between individuals with MRKH syndrome and healthy control females has consistently revealed elevated scores in psychological disorder categories such as, psychoticism, depression, phobic and generalized anxiety, among those affected by the syndrome (Heller-Boersma et al., 2009). A study conducted by Chen et al. (2020), found that 75,2% of MRKH individuals suffer from depressive symptoms, and one-third of these individuals even reached moderate to severe levels. In particular, the depressive symptoms are more pronounced in patients in their 20s (Chen et al., 2020). The psychological challenges faced by individuals with MRKH syndrome encompass several key themes: diagnosis, treatments for vaginal agenesis, and issues related to infertility and pregnancy at the forefront.

3.1 The Psychological Toll of Diagnosis

The diagnostic journey for MRKH, often perceived as traumatic by affected individuals, initiates a cascade of emotional struggles. This process prompts profound questions about female identity, body image, and social and sexual roles (Heller-Boersma et al., 2009; Patterson et al., 2014). The majority of diagnoses occur later than desired, often stemming from healthcare professionals' limited awareness of this condition (Heller-Boersma et al., 2009). Regrettably, this lack of awareness is not only perceived as a traumatic experience for MRKH individuals but also presents a significant challenge due to the absence of a universally recognized diagnostic toolbox. This absence further exacerbates patients' feelings of isolation and hinders their ability to seek support, whether within their community or from expert healthcare providers (Hatim et al., 2021; Gilfillan & Carter, 2023). The diagnosis given around the years where the building blocks of gender identity are being created, contribute to low self-esteem, causing affected individuals to perceive themselves as defective, inferior, or even unlovable, particularly when faced with the first sexual "milestones" (Heller-Boersma et al., 2007). Additionally, these negative emotions felt by MRKH individuals contribute to feelings of apprehension about future romantic relationships and prompts introspection regarding their gender identity (Patterson et al., 2014; Hatim et al., 2021).

From a cultural and social perspective, some MRKH individuals took a positive view of the diagnosis, appreciating the absence of a need for menstrual pads or need to interrupt Ramadhan fasting (Hatim et al., 2021). However, MRKH individuals often expressed a desire to keep their diagnosis private, as they perceived MRKH as a social taboo and a stigmatizing condition (Patterson et al., 2014). Additionally, the family's response to the diagnosis is frequently characterized by an overprotective role played by the mother, which may include self-blame during the pregnancy (Patterson et al., 2014). Generally, MRKH individuals struggle with disclosing their diagnosis due to feelings of shame and a sense of being markedly different from cisgender females (Patterson et al., 2014).

3.2 Psychological Hurdles in Pursuit of Vaginal Agenesis Treatments

The journey to seek treatments for vaginal agenesis introduces another layer of psychological difficulties. Surgical interventions and the use of dilators, although essential for medical reasons, are still stigmatized in society and can be perceived as shameful by MRKH individuals (Heller-Boersma et al., 2009). Unfortunately, studies have demonstrated that even after the creation of a neovagina, many MRKH individuals continue to experience high levels of psychological distress and low self-esteem (Heller-Boersma et al., 2007). This is often because they undergo these procedures under societal pressure to conform to normative notions of 'womanhood', seeking to alleviate negative self-beliefs and emotional issues (Roen et al., 2018). Consequently, even after surgical intervention, some MRKH individuals struggle with sexual inhibition, anxiety, and discomfort during sexual experiences (Roen et al., 2018). Furthermore, non-surgical options such as self-dilation or coital dilation can be emotionally challenging due to daily discomfort and pain endured over several months or even years (Herlin et al., 2018). Regardless of the type and success of treatments, it has been demonstrated that MRKH individuals continue to grapple with long-lasting negative psychological side-effects stemming from the syndrome (Ismail-Pratt et al., 2007; Djordjević et al., 2011). Moreover, from a cultural standpoint, Malaysian MRKH individuals are often advised to return for treatment only after marriage, as premarital sex is assumed to be uncommon, and discussions about vaginal dilator techniques are avoided due to the social taboo surrounding virginity (Hatim et al., 2021).

3.3 Infertility and Parenthood Concerns

Beyond these challenges, the profound and intricate topic of infertility and parenthood remains a central concern for MRKH individuals. The definition of motherhood formulated by Russo (1976) is a cisgender female's "raison d'être". This perception significantly influences today's societal perspective on feeling like a cisgender female, and consequently, it imposes a profound emotional burden on MRKH individuals, particularly with regard to the infertility side-effect. For MRKH individuals, infertility stands as one of the most profound anxieties, casting uncertainty not only over the realization of their parenthood dreams but also over the potential reactions of prospective partners (Patterson et al., 2014). In the context of cultural differences, Malaysian MRKH individuals often contemplated entering into polygamous marriages (Hatim et al., 2021). As a matter of fact, many Malaysian MRKH individuals had their engagements terminated once their fiancés became aware of the MRKH condition (Hatim et al., 2021).

The most prevalent current choices for MRKH individuals aiming to have children are adoption (non-biological children) and surrogacy (biological children), however, it is important to acknowledge the myriad psychological, technical, and cultural challenges inherent to pursuing these options (Hatim et al., 2021; Patterson et al., 2014). As of now, MRKH individuals seeking to accomplish genetic parenthood, are primarily presented with two options: surrogacy or Uterine Transplantation (UTx). Notably, surrogacy is still prohibited in many European countries, which elevates the importance of UTx. Sweden, in particular, has conducted a highly successful UTx clinical trial, with a substantial number of participants being MRKH individuals (Brännström et al., 2014; Brännström et al., 2015). Nevertheless, it is imperative to acknowledge that such a complex procedure can impose significant psychological stress on both the MRKH individuals, their partners, and the uterine donor.

Researchers have pinpointed several challenging psychological phases in the context of UTx, including post-surgery recovery, reintegration into daily life, and adherence to treatment (Järvholm et al., 2015). Within the first year following the UTx procedure, both recipients and their partners exhibited remarkable psychological resilience regarding this life-altering transplantation, with only a minor decrease observed three-months post-UTx (Järvholm et al., 2015). However, the emotionally sensitive period associated with pregnancy attempts commences one year after UTx (Järvholm et al., 2020). Investigating a later post-transplantation timeframe, a reversible decline is noted in recipients during the second year, likely attributed to difficulties in achieving pregnancy and parenthood (Järvholm et al., 2020). The uterine donors also experienced certain emotional challenges, although the majority maintained relatively stable health-related quality of life, mental well-being, and marital relationships post-donation (Järvholm et al., 2019). By five-year post-transplantation, recipients reported a positive impact on their self-image and shared mixed feelings about UTx in terms of achieving a sense of completeness as a cisgender female (Järvholm, Enskog et al., 2020). The Swedish UTx clinical trial resulted in all seven participants achieving pregnancy. Recipients encountered common challenges akin to those described by cisgender females undergoing infertility treatments, such as “The yoke of childlessness”, “Going through the uncertain”, and “Motherhood as surreal and normal” (Järvholm et al., 2021).

4.0 Exploring New Paths to Parenthood: Advancements in Uterine Transplantation and Uterus Tissue Engineering

The forthcoming sections of this review will delve into the latest findings on UTx and explore various approaches currently under investigation in uterine Tissue Engineering (TE) enabling pregnancy for MRKH individuals. While these recent findings are still in the early stages of development and are not yet ready for human clinical application, they represent exciting prospects that may reshape the landscape of reproductive medicine, offering novel possibilities for individuals facing challenges such as MRKH individuals or transfeminine people.

4.1 Developments in Uterus Transplantation Procedures

UTx represents a ground-breaking advancement for individuals afflicted by absolute uterine factor infertility (AUFI), a condition that, prior to this technique's development, left them with little hope of ever experiencing the joy of childbirth (Brännström et al., 2023). As of now, UTx can be performed from a live donor (LD) or a deceased donor (DD). According to Brännström et al. (2023), a typical UTx procedure with a LD begins with a comprehensive evaluation and health screening of both the donor and recipient, followed by committee approval to initiate *in-vitro* fertilization (IVF) treatment. Subsequently, the recipient undergoes immunosuppression to prepare for the UTx, embryo transfer, and the aspiration of a successful pregnancy. The delivery occurs via a caesarean section, followed by a hysterectomy to discontinue immunosuppressive medication intake. Lastly, the patient is advised to undergo long-term follow-up to monitor potential complications. Following these steps, the first clinical trial for UTx took place in Sweden where between 2012 and 2013, nine LD UTx procedures were conducted and resulted in the historic first live UTx birth in 2014 at the Sahlgrenska University Hospital in Gothenburg (Brännström et al., 2014; Brännström et al., 2015). By 2022, out of 80 UTx procedures, 40 live births were achieved worldwide (Brännström et al., 2023). Moreover, a second clinical trial using LD UTx has started using robotic-assisted laparoscopic surgery on the donor to reduce the invasiveness of the procedure and the recovery period (Brännström et al., 2020). Regarding DD UTx, the first worldwide live birth from an MRKH individual was registered in in 2017 in Brazil at the Hospital das Clínicas, University of São Paulo School of Medicine, which opened a path for AUFI individuals to become pregnant without needing uterus from living donors or live donor surgery (Ejzenberg et al., 2018). The procedure parallels LD UTx, with the key distinction being that the donor in DD UTx is deceased due to brain injury, and the organ retrieval must occur within a critically time-sensitive window (Ejzenberg et al., 2018).

Even though LD UTx and DD UTx have demonstrated positive outcomes, there is still much research needed in this area to address numerous limitations and enhance our understanding of the topic, such as research not only on human uterus transplantation but also on animal models from rodents to larger animals and then to non-human primates (Brännström et al., 2023). The research started with mice in 2002 and progressed to rats, sheep, pigs, and non-human primates (Brännström et al., 2023). While uterine transplantation has shown promise in aiding infertile cisgender females to attain their reproductive goals, it comes with a set of significant health concerns. Notably, individuals undergoing this procedure must undergo immunosuppression to halt progression of organ (uterus) rejection, which elevates their susceptibility to nephrotoxicity, heightened risk of severe infections, and diabetes (Hellström et al., 2016). As a result, the field of regenerative medicine has embarked on exploring alternative strategies to address AUFI.

4.2 Latest Uterus Tissue Engineering Findings

Innovative TE practices have emerged, focusing on replacing the allogenic sources used in UTx. For uterine TE, several steps need to be considered in order to engineer a transplantable uterus. To start, it needs to be taken into consideration the highly dynamic native endometrium extracellular matrix (ECM), responsible for connecting the cellular components of a tissue by translating biochemical and biomechanical cues into fundamental cellular processes for tissue survival and regeneration (De Vriendt et al., 2023). Specifically, the endometrial ECM consists of proteins that endow the uterus with a dynamic composition capable of withstanding the changes it goes through continuous remodeling during the menstrual cycle and pregnancy (De Vriendt et al., 2023). Hence, regarding uterus TE, the viscoelastic properties of ECM must be taken into consideration (De Vriendt et al., 2023). Additionally, the cell types used are fundamental. The optimal choice of cell type would be uterus-specific stem cells derived from the endometrium and myometrium (Yoshimasa & Maruyama, 2021). However, since endometrial and myometrial stem/progenitor cells do not expand properly *in-vitro* and *in-vivo*, embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs) have become good alternatives, since they can proliferate indefinitely while keeping their stemness (Yoshimasa & Maruyama, 2021). Song et al. (2015) successfully generated endometrium epithelial-like cells from hESCs by co-culture with endometrial stromal cells and specific cytokines. The cells were transplanted onto a collagen scaffold and repaired uterine damage in a rat model. Moreover, Miyazaki et al. (2018) differentiated hiPSCs to endometrial stromal fibroblasts (EMSFs) by investigating the molecular pathway WNT/CTNNB1 and hormonal stimulation.

Regarding the scaffolds needed for the bioengineering of the uterus, current studies have used biological materials such as biological scaffolds, decellularized biological tissue, or synthetic scaffolds (Yoshimasa & Maruyama, 2021). Biological scaffolds are often characterized by enhanced biocompatibility when compared to synthetic materials, as they are comprised of pre-existing macromolecules found in the ECM, thus also exhibiting mechanical and adhesive properties akin to the ECM (Yoshimasa & Maruyama, 2021). In particular, collagen-based materials hold great promise for reproductive medicine (H. Chen et al., 2022). Collagen plays a vital role for the bio-inks used in 3D bio-printing, which are fundamental for the structural and biochemical support for cell viability and growth (H. Chen et al., 2022). A porous collagen scaffold seeded with umbilical cord-derived mesenchymal stem cells (UC-MSCs) was shown to promote human endometrial stromal cell (HESC) proliferation and resulted in endometrial structural reconstruction and functional recovery in rats (Xin et al., 2019). Porous collagen scaffolds can also be used to mimic the endometrial structure through a hormone-responsive co-culture model based on the addition of stromal and epithelial cells, and formation of luminal-like epithelial layers with apical polarization (Abbas et al., 2020).

An alternative method involves the decellularization and recellularization strategy of biological tissue, initially emerging in 2013, enabling the creation of scaffolds with reduced antigenicity to minimize immune reactions post-transplantation (Yoshimasa & Maruyama, 2021; Daryabari et al., 2022). Decellularization is a multi-step process wherein living cells are eliminated from human or animal organs or tissues to create a scaffold that still mimics the macrostructure and microstructure of fundamental ECM components of that organ or tissue (Yoshimasa & Maruyama, 2021). These decellularized scaffolds play a crucial role in offering structural support to the cells to be integrated, and they can subsequently undergo recellularization with diverse types of autologous somatic and stem cells (Hellström et al., 2016). The resulting engineered structure must be sustained either within advanced perfusion bioreactors *in-vitro* or grown ectopically *in-vivo* (Hellström et al., 2016). Studies on bioengineering uterine patches using rat models are further investigating the best scaffold decellularizing process (Hellström, Moreno-Moya, et al., 2016). A study confirmed that by decellularizing biological tissues with triton-X100 in DMSO and repopulating the decellularized scaffold with primary uterine cells and green fluorescent protein–labeled bone marrow–derived mesenchymal stem cells (GFP-MSCs) supported a rat pregnancy (Hellström, Moreno-Moya, et al., 2016). This decellularizing process was further tested on larger animal models that closely mimic the size, shape, pregnancy characteristics, and vascular anatomy of the human uterus, such as sheep, and demonstrated promising outcomes (Tiemann et al., 2020). As a result of these encouraging results from animal studies, researchers proceeded to create human uterine whole-organ bio-scaffolds and optimize the decellularization process of the human uterus to then implant it in rats (Daryabari et al., 2022). This approach demonstrated that after the decellularization process, the collagen fibers in the bio-scaffold was maintained, as well as the ECM's composition and structure and its content was similar to the native human uterus (Daryabari et al., 2022). Furthermore, the preservation of the ECM for the major uterine arteries and vascular network in the decellularized uteri, showed its feasibility for *in-vitro* applications and *in-vivo* recellularization (Daryabari et al., 2022).

While previous research has demonstrated the effectiveness of biological scaffolds, scientists have encountered challenges such as batch variability, a short degradation period, and difficulties in quality control and purification (Yoshimasa & Maruyama, 2021). In response to these hurdles, researchers are increasingly exploring synthetic scaffolds as promising alternatives. Synthetic materials like polycaprolactone (PCL) nanofibers have shown promise in engineering scaffolds for the uterine myometrium smooth muscle tissue, due to the scaffold's similarity to the cells ECM (Hanuman & Nune, 2021). Hence, a study first explored PCL nanofibrous scaffolds by electrospinning and modified its surface through processes involving aminolysis, followed by maltose conjugation to enhance cellular adhesion, proliferation, and differentiation in both *in-vitro* and *in-vivo* conditions (Hanuman & Nune, 2021). These maltose-conjugated PCL scaffolds demonstrated that human uterine fibroblast (HUF) cells proliferation and attachment was better compared to other type of scaffolds (Hanuman & Nune, 2021). Moreover, the elastic properties of the PCL polymer, made it a promising human uterine myometrial patch that mimicked the intrauterine microenvironment to treat uterine injuries and supporting regeneration (Hanuman & Nune, 2021). Furthermore, alternative methods involving the modification of PCL nanofibers through hydrolysis and aminolysis treatments also present promising options for potential use as tissue engineering scaffolds (Yaseri et al., 2023).

A hybrid approach between the two types of scaffolds was executed by Y. Chen et al. (2020), by creating a scaffold based on human amniotic extracellular matrix (HAECM) with synthetic components such as poly (lactic acid-co-glycolic acid) (PLGA) microspheres loaded with 17β-estradiol (E2) (E2-MS), integrated for drug delivery purposes. This study showed promising results as a disease model to develop treatment to intrauterine adhesions since the E2-MS-HAECM scaffold replicated menstrual cycle's hormonal changes while supporting the growth and proliferation of endometrial cells. Furthermore, the upregulation of growth factors, such as EGF and IGF1, known contributors to endometrial regeneration, provides additional confirmation of the potential effectiveness of this drug-loaded bioactive scaffold (Y. Chen et al., 2020).

Moreover, the aforementioned studies concentrate on regenerating the endometrium, a crucial factor for successful embryo implantation and pregnancy. However, these studies face limitations in their ability to fully restore or regenerate the complex structure and functionality of the entire multi-layered uterus. Therefore, S. Chen et al. (2023) opted for a strategy that closely mimicks native uterine tissue by employing a combination of 4D printing, electrospinning, and 3D bioprinting. To reproduce the stretchable myometrium, poly L-lactide-co-trimethylene carbonate (PTMC) and thermoplastic polyurethane (TPU) were mixed through fused deposition modeling (FDM). A second layer to regulate cell behaviour was created by electrospinning and was fused with a mixture of PLGA and gelatin methacryloyl (GelMA), as well as encapsulated E2. Lastly, on top of the second layer, a bone marrow-derived mesenchymal stem cell (BMSC)-laden GelMA/Gel hydrogel was 3D bioprinted. These trilayers exhibited remarkable elasticity, transitioning from a planar configuration to tubular structures during culture at 37°C, as well as displaying impressive stretchability, high BMSCs survival rate, and enabled controlled release of E2. In summary, this strategy may successfully achieve the optimal biological and mechanical advantages necessary for the full restoration or regeneration of the complex structure and functionality of the entire multi-layered uterus.

5.0 Discussion

MRKH syndrome is a complex condition that carries significant sociopsychological implications such as the gender identity, body image and the pursuit of goals set by society, such as intimacy and parenthood. Hence, the following section will critically discuss the main topics influencing the psychological wellbeing of MRKH individuals as well as, consider potential avenues for societal modernization to foster greater inclusivity towards individuals with rare conditions.

5.1 Heterogeneity in MRKH Psychological Studies Outcomes

The psychological impact of MRKH syndrome has yielded different findings in different studies. Some research, based on psychological questionnaires, suggests that MRKH individuals exhibited no significant differences in depression, anxiety, self-esteem, or body image when compared to a control group of cisgender females (Gatti et al., 2010; Weijenborg et al., 2019; Rall et al., 2021). However, there is notable heterogeneity across these studies, and several limitations should be considered. One key factor is the difference in age intervals considered in each study, which may influence the outcomes. Psychological well-being in MRKH individuals tends to fluctuate during pivotal periods, such as the years surrounding diagnosis, initial sexual experiences, and attempts at pregnancy. Furthermore, these studies may exhibit selection bias due to the following reasons. MRKH individuals facing severe psychological disturbances might opt not to participate and, MRKH individuals with low socioeconomic status, who are potentially less engaged with healthcare professionals, might be underrepresented (Heller-Boersma et al., 2009). Additionally, in several studies, a higher proportion of MRKH individuals reported having a partner compared to the control group, and they were all in heterosexual relationships. Furthermore, the majority of studies are limited to a maximum of three countries. For instance, Hatim et al (2021) examination of Malaysian MRKH individuals highlighted how cultural traditions profoundly shape perspectives on the syndrome in distinct ways.

5.2 Navigating the Research Gap: The Underexplored Landscape of MRKH Syndrome in Female Health Studies

An outside factor that influences the psychological well-being of MRKH individuals is the very common issue of under researched female and rare diseases. This is a general societal issue where females suffering from rare diseases are victims of a two-fold bias. Improvements into research and fundings for MRKH could potentially have an impact on the psychological well-being of MRKH individuals. Firstly, considering that the diagnosis delivers life-changing news, it should be imperative for healthcare professionals to undergo specialized training in its delivery, ensuring optimal conditions for the patient. Therefore, the implementation of universally standardized guidelines are crucial, outlining steps aimed at providing comprehensive medical and psychological support to the individual. Analogous to established procedures for conditions like a heart attack, where specific steps are systematically executed, addressing the psychological well-being of MRKH individuals should be accorded with the same significance as their physical health. In this regard, the development of a cognitive-behavioral model and group treatment for MRKH has proven effective in enhancing the psychological well-being of affected individuals (Heller-Boersma et al., 2007). Furthermore, increased awareness of this condition among medical doctors would mitigate the tendency to leave MRKH individuals without answers and also foster greater compassion toward them (Gilfillan & Carter, 2023). From a societal perspective, it would be advantageous to employ additional tools for sharing information about the syndrome to family and friends, aiming to enhance their understanding of the emotional challenges faced by MRKH individuals. Given the absence of a visible physical representation, comprehending the intensity of these struggles might prove challenging. As one interviewee expressed, "You suddenly feel like you're broken in some way."

5.3 Considerations for Vaginal Agenesis Treatments

Furthermore, regarding the treatment for vaginal agenesis, experts have identified key psychological observations that need to be made before treating the condition. As an example, a lot of emphasis is made on the language the medical doctors use when addressing MRKH individuals and discussing the deeply emotional journey for vaginal reconstruction (Roen et al., 2018). In particular, it is highlighted how instead of measuring the success of vaginal reconstruction based on physical changes, like anatomy, the focus should be shifted to the individual's personal feelings of control and empowerment (Roen et al., 2018).

Furthermore, while non-surgical dilation stands out as one of the initial choices due to its lower complication risks, it is not without them. These risks still include potential issues like vaginal prolapse, mental distress, diminished satisfaction, and prolonged discomfort in the long-term (Sueters et al., 2023). In an effort to address these limitations, various TE approaches based on decellularizing tissue and synthetic biomaterials have been developed (Elia et al., 2022). These techniques show promising results in their ability to be tissue-specific and replicate the biological properties of a native vagina such as mucus production, and maintaining the natural microbiome (Elia et al., 2022). By addressing contemporary societal needs, these TE procedures could also find clinical application in the context of transfeminine individuals. Conclusively, as emphasized by an interviewed individual, MRKH individuals should not be subjected to pressure to hastily make decisions about the procedure merely to conform to societal expectations of what the ideal characteristics of a vagina should be.

5.4 Ethical Dilemmas in Uterine Transplantation

In the pursuit of achieving parenthood, significant strides have been made through the development of UTx. However, addressing ethical considerations is crucial as we navigate the frontiers of medically assisted reproduction. In 2012, “The Montreal Criteria”, composed by a set of criteria that the recipient needs to reach to be eligible for a UTx, was issued (Gullo et al., 2022). Hence, the following ethical questions should be considered. Firstly, gestation holds profound value in the context of human reproduction. Thus, relying on medically assisted reproduction has the potential to perpetuate the social stigma associating parenthood with a core aspect of female identity (O’Donovan et al., 2019). Moreover, cisgender females who lack access to these procedures may even experience an amplified sense of inferiority. Therefore, societal efforts should focus on evolving beyond traditional or genetic family norms, ensuring that alternatives such as adoption, surrogacy, or even childlessness are not unfairly viewed as inferior choices by society (O’Donovan et al., 2019). One of the interviewed persons stated, "I think there is a significant need for progress in redefining our understanding of what constitutes a family."

Secondly, the clinical and practical risks inherent to UTx must be considered for all involved parties. For the recipient, the extended use of immunosuppressant treatments poses potential health hazards, including an elevated risk of conditions such as skin cancer or haematological malignancies (Brännström & Dahm-Kähler, 2019). Additionally, from a psychological standpoint, recipients undergo a profound adjustment period as they navigate the experience of having a uterus, followed by the potential challenge of returning to their pre-transplant emotional state (Järvholm et al., 2015). Furthermore, there is ongoing debate regarding the risks faced by the LD for a non-life-saving surgery, the donor's psychological awareness of their decision, and the possibility of financial motivations (O’Donovan et al., 2019). While the use of a DD could potentially alleviate some of these ethical issues, medical professionals agree that using a LD is preferable due to greater benefits post-transplantation (O’Donovan et al., 2019).

Lastly, discussions have emerged concerning the welfare of the born child. It is suggested that, before undergoing the procedure, comprehensive assessments including criminal background checks, financial evaluations, sociopsychological evaluations, and an assessment of the couple's stability should be conducted (O'Donovan et al., 2019). However, it is important to acknowledge that such criteria may introduce additional challenges, including the potential reinforcement of pre-existing societal biases related to subjective judgments of what constitutes "good parents" (O'Donovan et al., 2019). In conclusion, there is a pressing need for further research and the establishment of comprehensive guidelines to optimize the UTx procedure both medically and ethically. Despite being in an early experimental stage, uterine TE holds promise as a potential avenue for advancing reproductive technologies.

5.5 Innovative Uterine Tissue Engineering: Addressing Ethical and Medical Concerns in Lieu of Uterine Transplantation

New TE methods to replace UTx procedures could help with the ethical and medical issues linked to it. Instead of involving four parties (the recipient, the donor, the born child, the recipient's partner), these techniques aim to streamline it to three. This not only simplifies the ethical issues, but also addresses the common risks of making a donor undergo surgery for a non-life-threatening condition. However, it has to be taken into consideration that the progress that has been made so far in uterus TE is limited, most studies are still in the preclinical stage, and it will still take several years, possibly decades, until the principle of bioengineered uterus transplantation reaches the starting point for a human clinical trial.

As an example, further investigations on the decellularization process revealed that this process is heavily dependent on the animal species used. As an example, the reagent Triton has shown successful results on rat models, but not on sheep models (Hellström et al., 2016; Tiemann et al., 2020). Nevertheless, sheeps are considered by researchers as optimal non-primate large animal model for studies related to the uterus due to its close similarity to humans in terms of uterus size, shape, pregnancy characteristics, and vascular anatomy (Tiemann et al., 2020). Additionally, the decellularization strategy is a very complex process since the chosen detergent must remove all cellular components to create an immune system-inert scaffold to avoid rejection by the recipient, while also ensuring that the native ECM structure is preserved (Tiemann et al., 2020). Also, in the context of the recellularization process across the scaffold, researchers have yet to identify the optimal cell type. Therefore, considering the option of combining multiple cell types remains a viable approach to be tested. Furthermore, many bioengineered uteri have not undergone testing for the hormonal cycles and alterations in the animals receiving the graft. This oversight could have significant implications for the subsequent *in-vivo* studies (Daryabari et al., 2022).

Bioengineering techniques should extend their focus beyond seeking solutions for novel medically assisted reproduction, but rather delve deeper into the study of the syndrome's pathogenesis. Hence, from cells derived by uterine rudiment horns from MRKH individuals, hormone-responsive organoid cultures were developed (Brucker et al., 2022). This investigation enabled to shed more light on the etiology of MRKH syndrome by identifying, through RNA sequencing, differences in *LHX1* and *HOX* genes between diseased and healthy organoids (Brucker et al., 2022). Discoveries related to the genetics of the disease would also provide clarity regarding the health status of a child born with the inheritance of the mother's

DNA. Additionally, a study combined various aforementioned TE approaches to delve deeper into the 28-day human menstrual cycle (Gnecco et al., 2023). The researchers used organoid models on polyethylene glycol (PEG)-based hydrogel scaffold with matrix metalloproteinases (MMPs) responsible for degrading PEG and promote organoid remodeling and growth (Gnecco et al., 2023). To ensure hormone-responsive organoids *in-vitro*, synthetic biochemical cues like integrin ITGα2β1 or ITGα5β1binding peptide based on collagen (GFOGER) were added (Gnecco et al., 2023). In conclusion, it is crucial to emphasize the necessity for additional research in developing reproducible *in-vitro* tissue models. This not only enhances the safety of animal testing in preclinical research trials but also contributes to minimizing animal use in scientific research.

5.6 Considerations on Parenthood Choices between MRKH Individuals

To summarize, as depicted in *Figure 3*, nowadays individuals with MRKH syndrome have four technical choices in terms of parenthood, although accessibility and feasibility vary among individuals. These options include adoption, surrogacy, UTx, and uterus TE. However, recent developments in new possibilities for childbearing among MRKH individuals have a dual impact. On the one hand, they undoubtedly provide reassurance to MRKH individuals by showcasing a variety of options for achieving pregnancy and parenthood, that can be specifically tailored to the individual's needs. On the other hand, these advancements may present adjustment challenges for MRKH individuals, given the distinct psychological impacts associated with each procedure, leading to uncertainty in decision-making. An additional noteworthy point raised by the interviewed MRKH individual, is the limited awareness in the general public about these recent TE techniques. This lack of information could potentially amplify feelings of insecurity and skepticism towards these procedures.

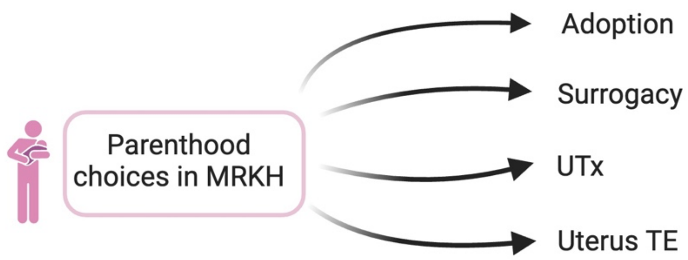

Figure 3: *Navigating Parenthood Choices in MRKH: exploring adoption, surrogacy, UTx, and uterus TE alternatives (Created with Biorender).*

6.0 Concluding remarks

In conclusion, the MRKH syndrome presents a highly intricate case that needs increased attention from both research, medical, and psychological perspectives. This review underscores the crucial need for sustained mental health support across the various life stages that pose distinct psychological challenges for the affected individuals. Additionally, societal modernization is fundamental to mitigate the isolation experienced by individuals facing sexual development disorders. Moreover, it is imperative for studies to enhance inclusivity by considering the diverse cultural and socioeconomic backgrounds of individuals, ensuring more accurate and representative conclusions. Furthermore, the field of uterus TE holds significant promise in addressing the challenges associated with vaginal reconstruction procedures and overcoming clinical and ethical limitations in achieving genetic parenthood. A holistic approach is essential, with the overarching research goal being the development of pivotal tools to provide comprehensive psychological assistance to individuals navigating the complexities of MRKH syndrome.

7.0 Methodology: Strategies for Literature Review and Interview Procedures

The literature review utilized PubMed, Google Scholar, and OMIM databases. In the search engines, the following keywords were employed: MRKH syndrome, Rokitansky syndrome, uterovaginal agenesis, uterus anatomy, MRKH genetics, psychology MRKH, quality of life MRKH, motherhood MRKH, uterus transplantation, vaginoplasty, uterus tissue engineering, uterus-specific stem cells, uterus collagen scaffold, uterus decellularization, and organoids MRKH.

To integrate qualitative data into the discussion, interviews were arranged to engage individuals directly affected by MRKH, medical professionals with specialization in MRKH individuals care, clinical psychologists, and advocates associated with MRKH organizations. A total of nine individuals were contacted via e-mail, resulting in responses from four. Ultimately, two interviews were successfully conducted online. The participants consisted of 1) a clinical psychologist, highly experienced in guiding patients through the uterus transplantation process, and 2) a MRKH individual who is also the director of an MRKH organization. The online interviews ranged between 45-60 minutes, and were recorded with consent. The interview questions, presented below, were shared with participants a few days prior to the interview, and relevant quotes have been integrated into the Discussion section of this review.

Interview questions:

1. In just a couple of sentences, could you please provide an overview of the MRKH syndrome?
    a. What are the main medical challenges faced by MRKH individuals?
2. What sociopsychological factors affect individuals with MRKH?
    a. Coping with the shock of diagnosis
    b. Navigating treatments for vaginal agenesis
    c. Dealing with the emotional challenges of infertility
    d. Making decisions about parenting options, such as adoption, surrogacy, or uterus transplantation.
3. Can you discuss the importance of psychological support for MRKH individuals?
4. In light of the recent progress in performing uterine transplants from living donors to MRKH individuals, enabling them to have children, it is worth noting that this procedure is intricate and involves a range of medical and ethical considerations.
    a. What are your first thoughts when thinking about this procedure? Do they lean more towards a positive or negative perspective?

b. How do you perceive the potential impact of this procedure on the psychological and social well-being of MRKH individuals?
c. Are there any ethical concerns or issues you find important to address in relation to this procedure?

5. In recent years, significant progress has been made in the field of regenerative medicine, particularly in uterus tissue engineering. These innovative approaches aim to tackle the challenges associated with uterine transplantation, including the necessity for immunosuppressants and donor availability. Given these advancements, do you think that this emerging approach could lead to substantial psychological differences for individuals receiving this treatment as compared to the more traditional approach in uterine transplantation?
6. Among the many medical challenges confronting MRKH individuals, which one do you believe has the most significant psychological impact? Additionally, do you think that the availability of numerous medical options itself has a psychological impact on MRKH individuals?
7. Which aspect of MRKH do you think deserves further research attention?

Reference List:

1. Abbas, Y., Brunel, L. G., Hollinshead, M., Fernando, R. C., Gardner, L., Duncan, I., Moffett, A., Best, S. M., Turco, M. Y., Burton, G. J., & Cameron, R. E. (2020). Generation of a three-dimensional collagen scaffold-based model of the human endometrium. *Interface Focus*, *10*(2), 20190079. https://doi.org/10.1098/rsfs.2019.0079
2. Brännström, M., & Dahm-Kähler, P. (2019). Uterus transplantation and fertility preservation. *Best Practice & Research in Clinical Obstetrics & Gynaecology*, *55*, 109–116. https://doi.org/10.1016/j.bpobgyn.2018.12.006
3. Brännström, M., Dahm-Kähler, P., Kvarnström, N., Akouri, R., Rova, K., Olausson, M., Groth, K., Ekberg, J., Enskog, A., Sheikhi, M., Mölne, J., & Bokström, H. (2020). Live birth after robotic-assisted live donor uterus transplantation. *Acta Obstetricia Et Gynecologica Scandinavica*, *99*(9), 1222–1229. https://doi.org/10.1111/aogs.13853
4. Brännström, M., Johannesson, L., Bokström, H., Kvarnström, N., Mölne, J., Dahm-Kähler, P., Enskog, A., Milenković, M., Ekberg, J., Díaz-García, C., Gäbel, M., Hanafy, A., Hagberg, H., Olausson, M., & Nilsson, L. (2015). Livebirth after uterus transplantation. *The Lancet*, *385*(9968), 607–616. https://doi.org/10.1016/s0140-6736(14)61728-1
5. Brännström, M., Johannesson, L., Dahm-Kähler, P., Enskog, A., Mölne, J., Kvarnström, N., Díaz-García, C., Hanafy, A., Lundmark, C., Marcickiewicz, J., Gäbel, M., Groth, K., Akouri, R., Eklind, S., Holgersson, J., Tzakis, A. G., & Olausson, M. (2014). First clinical uterus transplantation trial: a six-month report. *Fertility and Sterility*, *101*(5), 1228–1236. https://doi.org/10.1016/j.fertnstert.2014.02.024
6. Brännström, M., Racowsky, C., Carbonnel, M., Wu, J. H., Gargiulo, A. R., Adashi, E. Y., & Ayoubi, J. M. (2023). Uterus transplantation: from research, through human

trials and into the future. *Human Reproduction Update*, *29*(5), 521–544. https://doi.org/10.1093/humupd/dmad012

7. Brucker, S., Hentrich, T., Schulze-Hentrich, J. M., Pietzsch, M., Wajngarten, N., Singh, A. R., Rall, K., & Koch, A. (2022). Endometrial organoids derived from Mayer–Rokitansky–Küster–Hauser syndrome patients provide insights into disease-causing pathways. *Disease Models & Mechanisms*, *15*(5). https://doi.org/10.1242/dmm.049379
8. Chen, H., Li, X., Gong, G., Pan, J., Wang, X., Zhang, Y., Guo, J., & Qin, L. (2022). Collagen-based materials in reproductive medicine and engineered reproductive tissues. *Journal of Leather Science and Engineering*, *4*(1). https://doi.org/10.1186/s42825-021-00075-y
9. Chen, N., Song, S., Bao, X., & Zhu, L. (2022). Update on Mayer—Rokitansky—Küster—Hauser syndrome. *Frontiers of Medicine*, *16*(6), 859–872. https://doi.org/10.1007/s11684-022-0969-3
10. Chen, N., Song, S., Duan, Y., Kang, J., Deng, S., Pan, H., & Zhu, L. (2020). Study on depressive symptoms in patients with Mayer-Rokitansky-Küster-Hauser syndrome: an analysis of 141 cases. *Orphanet Journal of Rare Diseases*, *15*(1). https://doi.org/10.1186/s13023-020-01405-9
11. Chen, S., Li, J., Zheng, L. W., & Wang, M. (2023). Biomimicking Trilayer Scaffolds with Controlled Estradiol Release for Uterine Tissue Regeneration. *Authorea (Authorea)*. https://doi.org/10.22541/au.169674869.96372261/v1
12. Chen, Y., Fei, W., Zhao, Y., Wang, F., Zheng, X., Luan, X., & Zheng, C. (2020). Sustained delivery of 17β-estradiol by human amniotic extracellular matrix (HAECM) scaffold integrated with PLGA microspheres for endometrium

regeneration. *Drug Delivery*, *27*(1), 1165–1175. https://doi.org/10.1080/10717544.2020.1801891

13. Daryabari, S. S., Fendereski, K., Ghorbani, F., Dehnavi, M., Shafikhani, Y., Omranipour, A., Davani, S. Z., Zolbin, M. M., Tavangar, S. M., & Kajbafzadeh, A. (2022). Whole-organ decellularization of the human uterus and in vivo application of the bio-scaffolds in animal models. *Journal of Assisted Reproduction and Genetics*, *39*(6), 1237–1247. https://doi.org/10.1007/s10815-022-02492-2
14. De Vriendt, S., Casares, C., Rocha, S., & Vankelecom, H. (2023). Matrix scaffolds for endometrium-derived organoid models. *Frontiers in Endocrinology*, *14*. https://doi.org/10.3389/fendo.2023.1240064
15. Djordjević, M. L., Stanojević, D., & Bizic, M. (2011). Rectosigmoid vaginoplasty: clinical experience and outcomes in 86 cases. *The Journal of Sexual Medicine*, *8*(12), 3487–3494. https://doi.org/10.1111/j.1743-6109.2011.02494.x
16. Ejzenberg, D., Andraus, W., Mendes, L. R. B. C., Ducatti, L., Song, A., Tanigawa, R., Rocha-Santos, V., Arantes, R. M., Soares, J. M., Serafini, P., De Paiva Haddad, L. B., Francisco, R. P. V., D'Albuquerque, L. a. C., & Baracat, E. C. (2018). Livebirth after uterus transplantation from a deceased donor in a recipient with uterine infertility. *The Lancet*, *392*(10165), 2697–2704. https://doi.org/10.1016/s0140-6736(18)31766-5
17. Elia, E., Brownell, D., Chabaud, S., & Bolduc, S. (2022). Tissue engineering for gastrointestinal and genitourinary tracts. *International Journal of Molecular Sciences*, *24*(1), 9. https://doi.org/10.3390/ijms24010009
18. Gatti, C., Del Rossi, C., Lombardi, L., Caravaggi, F., Casolari, E., & Casadio, G. (2010). Sexuality and psychosocial functioning in young women after colovaginoplasty. *The Journal of Urology*, *184*(4S), 1799–1803. https://doi.org/10.1016/j.juro.2010.03.078

19. Gilfillan, R., & Carter, P. (2023). Issues of identity, perceptions and isolation: An interpretative phenomenological analysis of women's experience of Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome. *Journal of Health Psychology*. https://doi.org/10.1177/13591053231199253

20. Gnecco, J., Brown, A., Buttrey, K., Ives, C., Goods, B. A., Baugh, L., Hernandez-Gordillo, V., Loring, M., Isaacson, K., & Griffith, L. G. (2023). Organoid co-culture model of the human endometrium in a fully synthetic extracellular matrix enables the study of epithelial-stromal crosstalk. *Med*, *4*(8), 554-579.e9. https://doi.org/10.1016/j.medj.2023.07.004

21. Gullo, G., Etrusco, A., Fabio, M., Cucinella, G., Rossi, C., & Billone, V. (2022). The reproductive potential of uterus transplantation: future prospects. *PubMed*, *93*(2), e2022138. https://doi.org/10.23750/abm.v93i2.12868

22. Habiba, M., Heyn, R., Bianchi, P., Brosens, I., & Benagiano, G. (2020). The development of the human uterus: morphogenesis to menarche. *Human Reproduction Update*, *27*(1), 1–26. https://doi.org/10.1093/humupd/dmaa036

23. Hanuman, S., & Nune, M. (2021). Design and characterization of Maltose-Conjugated Polycaprolactone nanofibrous scaffolds for uterine tissue engineering. *Regenerative Engineering and Translational Medicine*, *8*(2), 334–344. https://doi.org/10.1007/s40883-021-00231-0

24. Hatim, H., Zainuddin, A. A., Anizah, A., Kalok, A., Daud, T. I. M., Ismail, A., Nurazurah, A., & Grover, S. (2021). The missing uterus, the missed diagnosis, and the missing care. Mayer-Rokitansky-Küster-Hauser Syndrome in the lives of Women in Malaysia. *Journal of Pediatric and Adolescent Gynecology*, *34*(2), 161–167. https://doi.org/10.1016/j.jpag.2020.11.009

25. Heller-Boersma, J. G., Schmidt, U., & Edmonds, D. K. (2007). A randomized controlled trial of a cognitive-behavioural group intervention versus waiting-list control for women with uterovaginal agenesis (Mayer–Rokitansky–Küster–Hauser syndrome: MRKH). *Human Reproduction*, *22*(8), 2296–2301. https://doi.org/10.1093/humrep/dem167
26. Heller-Boersma, J. G., Schmidt, U., & Edmonds, D. K. (2009). Psychological distress in women with uterovaginal agenesis (Mayer-Rokitansky-Küster-Hauser Syndrome, MRKH). *Psychosomatics*, *50*(3), 277–281. https://doi.org/10.1176/appi.psy.50.3.277
27. Hellström, M., Bandstein, S., & Brännström, M. (2016). Uterine tissue engineering and the future of uterus transplantation. *Annals of Biomedical Engineering*, *45*(7), 1718–1730. https://doi.org/10.1007/s10439-016-1776-2
28. Hellström, M., Moreno-Moya, J. M., Bandstein, S., Bom, E., Akouri, R., Miyazaki, K., Maruyama, T., & Brännström, M. (2016). Bioengineered uterine tissue supports pregnancy in a rat model. *Fertility and Sterility*, *106*(2), 487-496.e1. https://doi.org/10.1016/j.fertnstert.2016.03.048
29. Herlin, M., Bjørn, A. B., Jørgensen, L. K., Trolle, B., & Petersen, M. B. (2018). Treatment of vaginal agenesis in Mayer-Rokitansky-Küster-Hauser syndrome in Denmark: a nationwide comparative study of anatomical outcome and complications. *Fertility and Sterility*, *110*(4), 746–753. https://doi.org/10.1016/j.fertnstert.2018.05.015
30. Herlin, M., Petersen, M. B., & Brännström, M. (2020). Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome: a comprehensive update. *Orphanet Journal of Rare Diseases*, *15*(1). https://doi.org/10.1186/s13023-020-01491-9
31. Ismail-Pratt, I., Bikoo, M., Liao, L., Conway, G. S., & Creighton, S. M. (2007). Normalization of the vagina by dilator treatment alone in Complete Androgen

Insensitivity Syndrome and Mayer-Rokitansky-Kuster-Hauser Syndrome. *Human Reproduction*, *22*(7), 2020–2024. https://doi.org/10.1093/humrep/dem074

32. Järvholm, S., Bokström, H., Enskog, A., Hammarling, C., Dahm-Kähler, P., & Brännström, M. (2021). Striving for motherhood after uterus transplantation: a qualitative study concerning pregnancy attempts, and the first years of parenthood after transplantation. *Human Reproduction*, *37*(2), 274–283. https://doi.org/10.1093/humrep/deab260
33. Järvholm, S., Dahm-Kähler, P., Kvarnström, N., & Brännström, M. (2020). Psychosocial outcomes of uterine transplant recipients and partners up to 3 years after transplantation: results from the Swedish trial. *Fertility and Sterility*, *114*(2), 407–415. https://doi.org/10.1016/j.fertnstert.2020.03.043
34. Järvholm, S., Enskog, A., Hammarling, C., Dahm-Kähler, P., & Brännström, M. (2020). Uterus transplantation: joys and frustrations of becoming a 'complete' woman—a qualitative study regarding self-image in the 5-year period after transplantation. *Human Reproduction*, *35*(8), 1855–1863. https://doi.org/10.1093/humrep/deaa143
35. Järvholm, S., Johannesson, L., Clarke, A., & Brännström, M. (2015). Uterus transplantation trial: Psychological evaluation of recipients and partners during the post-transplantation year. *Fertility and Sterility*, *104*(4), 1010–1015. https://doi.org/10.1016/j.fertnstert.2015.06.038
36. Järvholm, S., Kvarnström, N., Dahm-Kähler, P., & Brännström, M. (2019). Donors' health-related quality-of-life and psychosocial outcomes 3 years after uterus donation for transplantation. *Human Reproduction*, *34*(7), 1270–1277. https://doi.org/10.1093/humrep/dez087

37. O'Donovan, L., Williams, N. J., & Wilkinson, S. (2019). Ethical and policy issues raised by uterus transplants. *British Medical Bulletin*, *131*(1), 19–28. https://doi.org/10.1093/bmb/ldz022

38. Patterson, C. J., Crawford, R., & Jahoda, A. (2014). Exploring the psychological impact of Mayer–Rokitansky–Küster–Hauser syndrome on young women: An interpretative phenomenological analysis. *Journal of Health Psychology*, *21*(7), 1228–1240. https://doi.org/10.1177/1359105314551077

39. Rall, K., Schenk, B., Schäffeler, N., Schöller, D., Kölle, A., Schönfisch, B., & Brucker, S. Y. (2021). Long Term Findings Concerning the Mental and Physical Condition, Quality of Life and Sexuality after Laparoscopically Assisted Creation of a Neovagina (Modified Vecchietti Technique) in Young MRKHS (Mayer-Rokitansky-Küster-Hauser-Syndrome) Patients. *Journal of Clinical Medicine*, *10*(6), 1269. https://doi.org/10.3390/jcm10061269

40. Roen, K., Creighton, S. M., Hegarty, P., & Liao, L. (2018). Vaginal Construction and Treatment Providers' experiences: A Qualitative analysis. *Journal of Pediatric and Adolescent Gynecology*, *31*(3), 247–251. https://doi.org/10.1016/j.jpag.2018.01.001

41. Russo, N. F. (1976). The motherhood mandate. *Journal of Social Issues*, *32*(3), 143–153. https://doi.org/10.1111/j.1540-4560.1976.tb02603.x

42. Song, T., Zhao, X., Sun, H., Li, X., Lin, N., Ding, L., Dai, J., & Hu, Y. (2015). Regeneration of Uterine Horns in Rats Using Collagen Scaffolds Loaded with Human Embryonic Stem Cell-Derived Endometrium-Like Cells. *Tissue Engineering Part A*, *21*(1–2), 353–361. https://doi.org/10.1089/ten.tea.2014.0052

43. Sueters, J., Groenman, F., Bouman, M., Roovers, J. P., De Vries, R., Smit, T., & Huirne, J. a. F. (2023). Tissue Engineering Neovagina for Vaginoplasty in Mayer–Rokitansky–Küster–Hauser Syndrome and Gender Dysphoria Patients: A Systematic

review. *Tissue Engineering Part B-reviews*, *29*(1), 28–46. https://doi.org/10.1089/ten.teb.2022.0067

44. Tempest, N., Hill, C. J., Maclean, A., Marston, K., Powell, S., Al-Lamee, H., & Hapangama, D. K. (2021). Novel microarchitecture of human endometrial glands: implications in endometrial regeneration and pathologies. *Human Reproduction Update*, *28*(2), 153–171. https://doi.org/10.1093/humupd/dmab039
45. Tiemann, T. T., Padma, A. M., Sehic, E., Bäckdahl, H., Oltean, M., Song, M. J., Brännström, M., & Hellström, M. (2020). Towards uterus tissue engineering: a comparative study of sheep uterus decellularisation. *Molecular Human Reproduction*, *26*(3), 167–178. https://doi.org/10.1093/molehr/gaaa009
46. Triantafyllidi, V. E., Mavrogianni, D., Kalampalikis, A., Litos, M., Roidi, S., & Michala, L. (2022). Identification of Genetic Causes in Mayer-Rokitansky-Küster-Hauser (MRKH) Syndrome: A Systematic Review of the literature. *Children (Basel)*, *9*(7), 961. https://doi.org/10.3390/children9070961
47. Tsarna, E., Eleftheriades, A., Eleftheriades, M., Kalampokas, E., Liakopoulou, M., & Christopoulos, P. (2022). The impact of Mayer–Rokitansky–Küster–Hauser Syndrome on Psychology, Quality of Life, and Sexual Life of Patients: A Systematic Review. *Children (Basel)*, *9*(4), 484. https://doi.org/10.3390/children9040484
48. Weijenborg, P., Kluivers, K. B., Dessens, A. B., Kate-Booij, M. T., & Both, S. (2019). Sexual functioning, sexual esteem, genital self-image and psychological and relational functioning in women with Mayer–Rokitansky–Küster–Hauser syndrome: a case–control study. *Human Reproduction*, *34*(9), 1661–1673. https://doi.org/10.1093/humrep/dez130
49. Xin, L., Lin, X., Pan, Y. B., Zheng, X., Shi, L., Zhang, Y., Ma, L., Gao, C., & Zhang, S. (2019). A collagen scaffold loaded with human umbilical cord-derived

mesenchymal stem cells facilitates endometrial regeneration and restores fertility. *Acta Biomaterialia*, *92*, 160–171. https://doi.org/10.1016/j.actbio.2019.05.012

50. Yaseri, R., Fadaie, M., Mirzaei, E., Samadian, H., & Ebrahiminezhad, A. (2023). Surface modification of polycaprolactone nanofibers through hydrolysis and aminolysis: a comparative study on structural characteristics, mechanical properties, and cellular performance. *Scientific Reports*, *13*(1). https://doi.org/10.1038/s41598-023-36563-w
51. Yoshimasa, Y., & Maruyama, T. (2021). Bioengineering of the uterus. *Reproductive Sciences*, *28*(6), 1596–1611. https://doi.org/10.1007/s43032-021-00503-8
52. Zhang, X., Liu, Z., Yang, Y., Yao, Y., & Tao, Y. (2017). The clinical outcomes of vaginoplasty using tissue-engineered biomaterial mesh in patients with Mayer-Rokitansky-Küster-Hauser syndrome. *International Journal of Surgery*, *44*, 9–14. https://doi.org/10.1016/j.ijsu.2017.06.026