Received: 20 November 2024 | Revised: 13 January 2025 | Accepted: 23 January 2025

DOI: 10.1111/andr.70007

ANDROLOGY

WILEY

REVIEW ARTICLE

Motile cilia: Key developmental and functional roles in reproductive systems

Shiyu Yang[1] | **Xiaoli Wang**[1] | **Huihui Gao**[1] | **Shuiqiao Yuan**[1,2,3]

[1]Institute of Reproductive Health, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, China

[2]Laboratory of Animal Center, Huazhong University of Science and Technology, Wuhan, China

[3]Shenzhen Huazhong University of Science and Technology Research Institute, Shenzhen, China

Correspondence
Dr. Shuiqiao Yuan, Institute of Reproductive Health, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei 430030, China.
Email: shuiqiaoyuan@hust.edu.cn

Funding information
National Natural Science Foundation of China, Grant/Award Number: 82371625; National Key Research and Development Program of China, Grant/Award Number: 2024YFA1108100

Abstract

Background: Cilia are specialized microtubule-based organelles that extend from the cell surface and are classified into non-motile and motile types. The assembly and function of cilia are regulated by a complex molecular network that enables motile cilia to generate fluid flow across epithelial surfaces through coordinated beating. These motile cilia are found in the respiratory, nervous, and reproductive systems. In males, motile cilia are found in the efferent ducts and facilitate the transport of sperm from the testis to the epididymis. In females, they are mainly found in the oviducts, where they help to transport, nourish and fertilize eggs, and are also present in the endometrial epithelium.

Material-Methods: This review compares the common factors that affect motile cilia in both male and female reproductive tracts, discusses the origin and development of multiciliated cell and cilia within the efferent ducts and oviducts, and enumerates the infertility or related reproductive diseases that may arise due to motile cilia defects.

Results-Discussion: In males, motile cilia in the efferent ducts create turbulence through their beating, which keeps semen suspended and prevents ductal obstruction. In females, motile cilia are distributed on the epithelia of the oviducts and the endometrium. Specifically, motile cilia in the infundibulum of the oviduct aid in capturing oocytes, while cilia in the isthmus region have been found to bind to sperm heads, facilitating the formation of the sperm reservoir. Several common factors, such as miR-34b/c and miR-449, TAp73, Gemc1, and estrogen, etc., have been shown to play crucial regulatory roles in motile cilia within the efferent ducts and oviducts, thereby further influencing fertility outcomes.

Conclusions: Pathogenic mutations that disrupt ciliary function can impair ciliogenesis or alter the structure of sperm flagella, potentially resulting in infertility. Consequently, motile cilia in both the male and female reproductive tracts are crucial for fertility. There are still numerous unresolved mysteries surrounding these cilia that merit

Shiyu Yang, and Xiaoli Wang authors contributed equally to this work.

further investigation by researchers, as they hold great significance for the clinical diagnosis and treatment of infertility and related reproductive disorders.

KEYWORDS
efferent ducts, fertility, motile cilia, oviducts

1 | INTRODUCTION

Cilia, microtubule-based organelles protruding from the cell surface, arise by centriole migration and form axonemal structures with either a '9+2' or '9+0' microtubule arrangement, depending on the presence or the absence of the central doublet[1] (Figure 1A). Primary cilia are observed in most cells with a sensory and signaling function,[2] whereas motile cilia are generally found in multiciliated cells (MCCs) in the epithelium of the respiratory, nervous, and reproductive systems.[3,4] These MCCs are adorned with an array of tens to hundreds of centrioles positioned on their apical surfaces, facilitating the outgrowth of an equivalent multitude of motile cilia.[5] These motile cilia are indispensable for fostering fluid movement within the lumen and facilitating the efficient transport of contents. Of note, multiciliogenesis involves multiple stages, including cell-cycle exit, centriole expansion and migration, and basal body docking.[6] This complex sequence of events is governed by intricate regulatory networks that ensure the precise coordination and execution of each step. In addition, cilia assembly is orchestrated by a bidirectional transport mechanism known as intraflagellar transport (IFT).[7,8]

In male mammals, spermatozoa are produced by the seminiferous tubules and collected in the rete testis. The efferent ducts (EDs) collectively serve as a vital conduit for the transport of spermatozoa, whose epithelium consists of MCCs and non-ciliated cells.[9] The motile cilia in the MCCs beat vigorously to create a turbulent flow that keeps the sperm in suspension.[10] Meanwhile, the non-ciliated cells mainly reabsorb components of the seminal fluid to produce highly concentrated semen. This harmonious collaboration ensures smooth transport of sperm to the epididymis, providing an optimal environment for sperm transfer.[11] In mice, the rete testis bifurcates laterally into 2–5 straight EDs, which subsequently develop into highly tortuous forms and eventually anastomose to form a single duct that connects to the caput of the epididymis.[12,13] At embryonic day 15.5 (E15.5), connections are established between the testicular cords and the mesonephric tubules (MTs), and these MTs undergo remodeling to form coiled EDs at birth.[14,15] The development of the MTs in mice, along with the expression patterns of steroid receptors during this process, such as the androgen receptor (AR) and the estrogen receptor (ESR1), has been thoroughly studied and proven to be crucial for the formation of the testes and EDs.[16,17] A previous study found that ciliated cells in the EDs of mice begin to differentiate around the fifth day after birth.[13] However, the definitive timing of ciliogenesis and the identity of the progenitor cells that give rise to MCCs in the EDs require further investigation. Additionally, the female oviduct, a delicate conduit connecting the ovary to the uterus, is segmented into four functional parts: the infundibulum, ampulla, isthmus, and uterotubal junction, each of which plays a vital role in the transport, nutrition, and fertilization of gametes, highlighting its central role in reproduction.[18–20] The oviductal epithelium is also composed of ciliated and non-ciliated secretory cells, the latter acting as progenitor cells capable of self-renewal and differentiation into ciliated cells.[21] Moreover, motile cilia are also present in the human endometrial epithelium, highlighting their broad functional importance in reproductive processes.[22]

In particular, recent studies have highlighted the critical role of motile cilia in male fertility, with some male patients diagnosed with primary ciliary dyskinesia (PCD) presenting with obstructive ozoospermia-associated infertility but normal sperm structure.[23–25] Infertility in female patients with PCD has also been observed, further emphasizing the cross-sexual importance of motile cilia.[26] This review summarizes recent studies on motile cilia, highlighting their indispensable functions within the reproductive system. It provides insight into the causes of male and female infertility and highlights areas that require further investigation, thereby enhancing our understanding of motile cilia and promoting progress in reproductive medicine research.

2 | AN OVERVIEW OF CILIOGENESIS

2.1 | The fundamental architecture of cilia

Cilia are microtubule-based, hair-like organelles on the cell surface that consist mainly of the basal body, transition zone, axoneme, and ciliary membrane[27] (Figure 1A). The core structure of cilia is the axoneme, which is enveloped by the ciliary membrane.[28] Depending on the configuration of the axoneme, cilia can generally be classified as either primary cilia, with a '9+0' arrangement, or motile cilia, characterized by a '9+2' microtubule arrangement.[1] Nonetheless, a small number of specialized cilia, such as nodal cilia, display motility despite having a '9+0' arrangement.[29] Primary cilia, which are ubiquitous on the surface of most cells, serve as sensors and transducers of extracellular signals.[2] In contrast, motile cilia are found specifically in the respiratory tract, brain ventricles (ependymal cells), and the epithelia of the male and female reproductive systems, where they exhibit their motility functions.[3,4]

In post-mitotic MCCs, an extraordinary simultaneous proliferation of hundreds of centrioles occurs, driven primarily by two distinct pathways. The mother–daughter pathway of centriole duplication is similar to the canonical pathway observed in cycling cells, but is estimated to contribute only about 10% of the basal body production in MCCs.[30] In

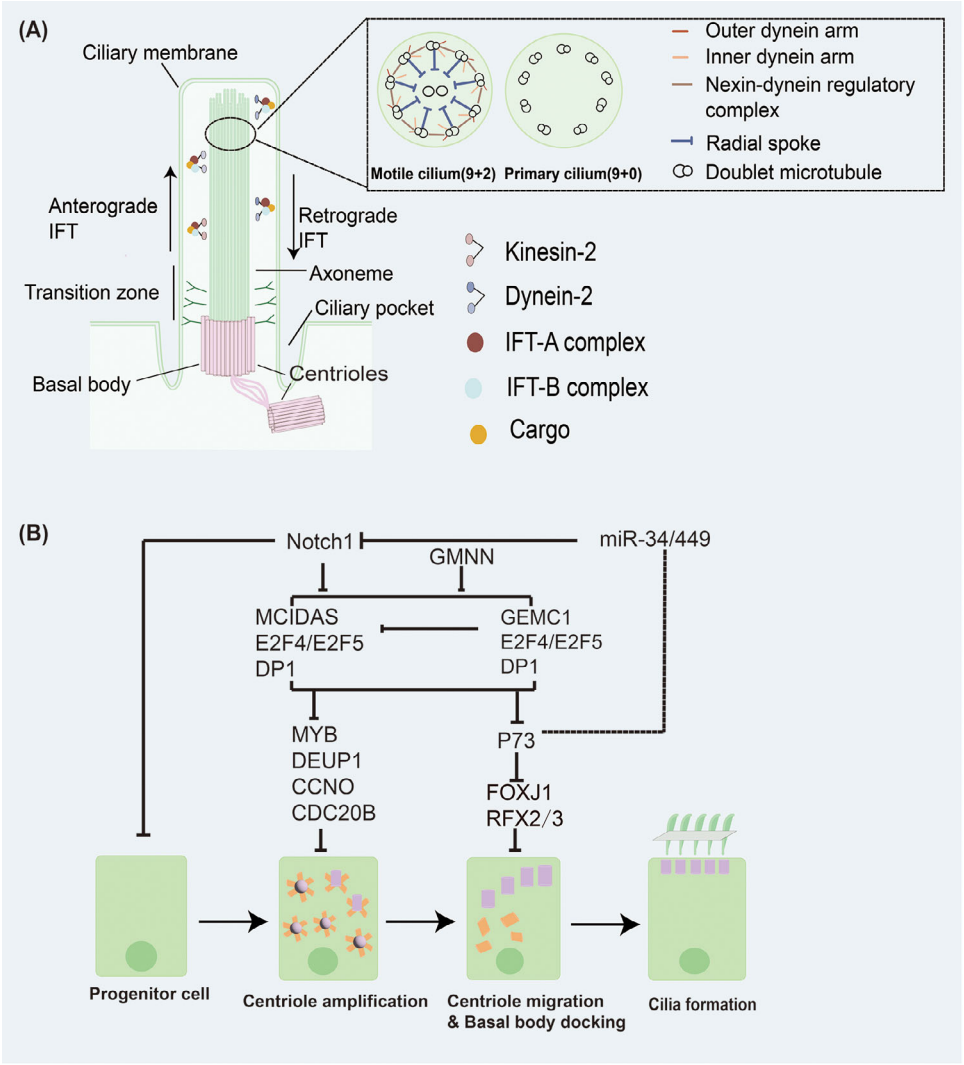

FIGURE 1 Multiciliated cell differentiation and the basic structure of cilia. (A) Cilia are composed of the ciliary membrane, axoneme, basal body, transition zone, ciliary pocket, and IFT protein particles. The axoneme of the motile cilia is assembled with nine peripheral doublet microtubules, two central singlet microtubules, inner and outer dynein arm, radial spokes, and nexin-dynein regulatory complex, whereas primary cilia lack central microtubules and dynein components. (B) Key factors involved in transcriptional regulation of multiciliogenesis and MCC differentiation. miR-34/449 can inhibit the Notch signaling pathway, thereby promoting the differentiation of MCC progenitor cells. This inhibition activates key regulators GEMC1 and MCIDAS, which collaborate with E2F4/5 and DP1 to positively regulate expression of centriole amplification genes (MYB, CCNO, DEUP1, CDC20B) and activate transcription factors FOJX1 and RFX2/3. GMNN inhibits GEMC1 and MCIDAS expression during MCC differentiation, while P73 functions downstream of MCIDAS/E2F4, regulating FOXJ1, RFX2/3, and miR-34b/c expression. IFT, Intraflagellar transport; MCC, multiciliated cell.

contrast, the deuterosome-dependent pathway involves the formation of deuterosomes—ring-shaped, electron-dense structures that arise independently of pre-existing mother centrioles and contain proteins such as DEUP1, CCDC78, and CDC20B, all of which are critical for rapid and efficient centriole expansion.[31,32] However, a recent study has cast doubt on the necessity of deuterosomes for centriole amplification in MCCs, as mice were observed to develop normal MCCs even in the absence of deuterosomes.[33] The apical surface of MCCs characteristically harbors an array of tens to hundreds of motile cilia, the number of which is closely linked to the number of centrioles. Nonetheless, the definitive pathways that meticulously govern the regulation of centriole abundance in MCCs remain elusive.[5] Following centriole amplification, these centrioles migrate to the cell surface, where they transform into basal bodies for cilia synthesis and acquire a rotational orientation that dictates the pattern of planar cilia beating. Securely anchored by the basal body, the axoneme extends outward from the cell and, in motile cilia, has an intricate structure comprising nine peripheral doublet microtubules (DMTs), two central singlet microtubules, inner and outer dynein arms (IDA and ODA), radial spokes (RS), and the nexin-dynein regulatory complex (N-DRC)[34] (Figure 1A). During axoneme assembly, many proteins need to be transported to the axoneme tip along microtubules, which rely on IFT.[7,8] IFT is a bidirectional mode of transport of protein particles such as kinesin-2 and dynein-2 from the base to the tip (anterograde) and from the tip back

to the base (retrograde), which plays a critical role in the assembly and maintenance of cilia and flagella.[35] Defects in IFT components lead to the development of ciliopathies such as polycystic nephropathy, skeletal ciliopathies and retinal degeneration. For example, mutation of the IFT particle protein IFT88 causes defective cilia and polycystic nephropathy in mice.[36]

It is noteworthy that the wave-like motion produced by the beating of the motile cilia is associated with mucus clearance in the respiratory tract,[37] the circulation of cerebrospinal fluid,[38] and the transport of gametes in the oviduct.[39] Specifically, motile cilia are equipped with dynein arms that use energy from ATP hydrolysis to orchestrate the axonemal movement through the sliding of DMTs against each other.[40] The mechanism by which cilia obtain energy through ATP regeneration and ADP depletion also relies on adenylate kinase, which catalyzes the exchange reaction of nucleotide phosphate groups.[41,42] A recent study has identified three distinct adenylate kinases located on the RS of the DMTs. This physical interaction ensures a constant level of ATP along the cilium.[43] Meanwhile, the central microtubules and RS are able to regulate the activity of dynein, which is also essential for cilia motility.[44,45]

2.2 | Pertinent regulatory factors involved in ciliogenesis

Transcriptional regulation of cilia-related genes is critical for MCC differentiation and ciliogenesis, and is mediated by numerous transcription factors and complex regulatory networks[46,47] (Figure 1B). In laevis epidermis and human airways, microRNAs such as miR-34/449 can initiate MCC differentiation by inhibiting the Notch/Delta pathway to facilitate the centriole proliferation and promote MCC differentiation.[48–50] In addition, the *Geminin* family consists of three distinct members: *Geminin*, *Mcidas*, and *Gemc1*, each of which occupy strategic positions within the regulatory network governing MCC differentiation. Geminin inhibits the expression of *Gemc1* and *Mcidas* during MCC differentiation, a function that is consistent with Notch signaling. Downstream of Notch signaling and *Geminin*, *Gemc1* and *Mcidas* work with the E2F4/5 transcription factors and their cofactor DP1 to positively regulate the expression of genes critical for centriole amplification, such as *Ccno* and *Myb*, and to activate the *Foxj1* and *Rfx2/3* transcription factors associated with ciliogenesis.[51–57] p73, a member of the p53 protein family, is expressed in p63$^+$ basal cells, which serve as progenitor cells in the airway epithelium, suggesting that p73 plays a pivotal role in the early fate determination of MCCs.[58,59] *TAp73*, an isoform of the *TP73* gene, acts downstream of E2F4/MCIDAS and also regulates the expression of *Foxj1*, *Rfx2/3*, and *miR34b/c* in the respiratory system, thus serving as a novel central regulator of multiciliogenesis.[60,61] Of note, the upregulation of *miR-449* following *TAp73* deletion suggests a complementary regulatory relationship between miR-449 and TAp73 in promoting multiciliogenesis in the brain.[62] *Foxj1* is a master transcription factor essential for MCC differentiation and motile cilia formation. It is specifically expressed in tissues containing motile cilia, making it a widely recognized marker for MCCs.[63–65] *Foxj1* deficiency in mice results in an absence of motile cilia structure and abnormal basal body anchoring.[66–68] In zebrafish and xenopus, ectopic expression of *Foxj1* in epithelial cells is sufficient to activate motile cilia growth in tissues, further confirming its central regulatory function in ciliogenesis.[69,70]

Additionally, the RFX family has also been shown to be a key cluster of transcription factors regulating motile ciliogenesis.[71–73] Similarly, RFX can regulate the transcription of genes encoding proteins involved in cilia assembly and function.[72,74] In particular, the transcription factor RFX can control the expression of IFT-related components and Bardet–Biedl syndrome proteins, which are essential for the IFT machinery.[75,76] In conclusion, the precise differentiation of MCCs and the subsequent ciliogenesis process requires the coordinated regulation of a large number of transcription factors and needs to be further investigated.

3 | MOTILE CILIA IN THE MALE EFFERENT DUCTS

3.1 | The transport of sperm in efferent ducts

The EDs serve as vital conduits connecting the rete testis to the epididymis (Figure 2A,B). Of note, small mammals like mice have fused EDs extending into the epididymal duct, whereas in large mammals like humans, each ED connects separately to the epididymal duct. This difference may be related to the folding of the cranial wolffian duct and requires further investigation.[77] In humans, the proximal segment of the epididymis consists mainly of a network of 6–15 EDs.[78,79] Upon release from the seminiferous tubules, sperm are collected in the rete testis and transported through the EDs toward the caput epididymis.[9] Previous studies have consistently found that the epithelial lining of the EDs is composed of ciliated and non-ciliated cells. Further research has shown that the 'non-ciliated' cells within the EDs actually contain primary cilia that originate from the microvillus boundary. The epithelial lining of the human EDs consists of 63% primary ciliated cells (FoxJ1$^-$/ERα^{+++}), 33% MCCs (FoxJ1$^+$/ER$\alpha^{+/-}$), and 4% FoxJ1$^-$/ERα^- cells. Notably, immunofluorescence staining further confirmed the presence of T lymphocytes (CD3$^+$) and macrophages (CD68$^+$ and CD163$^+$) within the EDs[79–81] Figure 2B, a distribution of cell types that has not been well defined in mouse EDs and female oviducts. A recent study used the single-cell RNA sequencing (scRNA-seq) technology to identify two distinct populations of epithelial cells within human EDs. One population exhibited a marker gene profile similar to that of kidney and bladder epithelium, while the other had marker genes associated with innate immunity and inflammation. Additionally, the study distinguished ciliated cells as a separate cluster characterized by specific marker genes including *CAPS*, *CAPSL*, *CFAP43/WDR96*, *TPPP3*, *DNAH5*, *DNAH6*, *DNAH12*, *DNAH9*, *and DNAI1/2*.[82] However, the progenitor that gives rise to MCCs within EDs, along with the precise developmental timing and differentiation trajectories of MCCs, remain poorly understood.

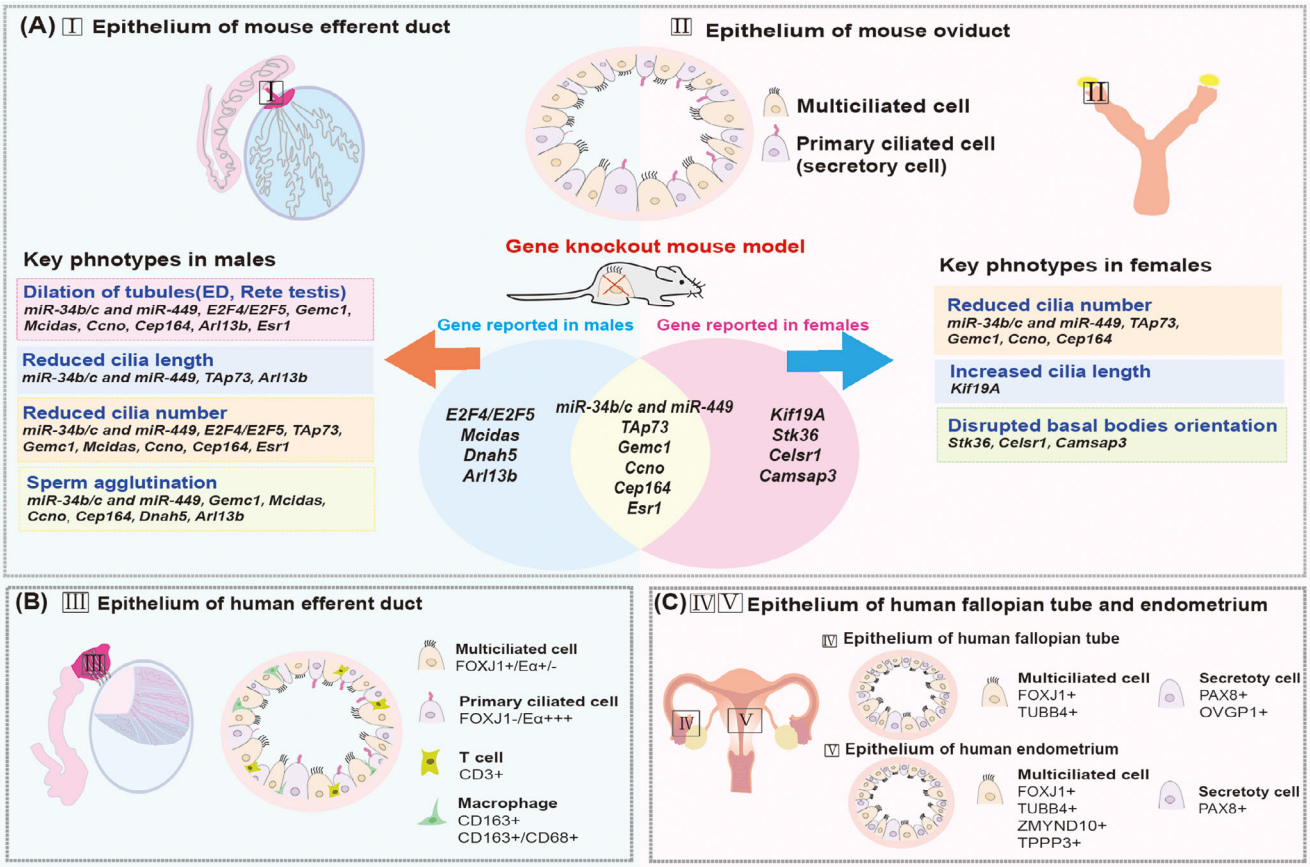

FIGURE 2 Schematic diagram of mouse and human reproductive tracts. (A) Diagrammatic representation of the mouse testis and oviduct, along with their luminal epithelium, which consists of multiciliated cells and primary ciliated cells (secretory cells). Genes commonly reported in both male and female mice (miR-34b/c and miR-449, TAp73, Gemc1, Ccno, Cep164, and Esr1), as well as genes studied exclusively in male mice (E2F4/E2F5, Mcidas, Dnah5, Arl13b) and in female mice (Kif19A, Stk36, Celsr1, Camsap3), are shown. Typical phenotypes observed in knockout mice for some of these genes are also summarized. (B) Diagram of the human male reproductive tract, highlighting the epithelium of the EDs. The lumen epithelium is composed of MCCs, primary ciliated cells, T cells, and macrophages, with characteristic markers identified. (C) Illustration of the human female reproductive tract, showing the epithelial lining of the fallopian tubes and endometrium. The epithelium lining both the fallopian tube and endometrium is composed of MCCs and secretory cells, and examples of their characteristic markers are shown. ED, efferent ducts; MCC, multiciliated cell.

The discovery of cilia in the male reproductive system dates back approximately 160 years, when Becker proposed the existence of motile cilia lining the epithelium of the EDs in the male reproductive tract.[83] The ED is anatomically segmented into three distinct regions: proximal, conical, and distal. Notably, the distal segment, close to the epididymis, has a higher density of ciliated cells than the proximal EDs, despite the larger diameter of the proximal tubule. This structure allows slower fluid flow in the proximal EDs, aiding fluid reabsorption and increasing sperm concentration by 25-fold. As sperm move into the narrower distal EDs, the increased number of cilia creates turbulence, maintaining sperm suspension for proper transport.[11,84,85] These cilia were originally thought to facilitate the transport of sperm to the epididymis by their rhythmic beating motions. However, recent studies have shown that the motile cilia in the EDs do not exhibit a coordinated wave-like movement. Instead, they maintain sperm suspension by agitating the luminal raw seminal fluid, thereby preventing sperm from clogging during the transport. In addition, it appears that peritubular smooth muscle contraction, rather than ciliary motion, is the primary force driving the sperm transport.[10,86] Thus, the precise function of motile cilia within the EDs and the intricate mode of the sperm transport have been elucidated.

3.2 | Male infertility due to ciliary defects in efferent ducts

Mice with ciliary defects in EDs show sperm agglutination, ductal occlusion, reduced epididymal sperm count, rete testis expansion, and male infertility. A previous study showed that the double KO (dKO) male mice lacking *miR-34b/c* and *miR-449* showed a significant decrease in sperm count and male sterility.[87] Our recent study shows that male mice with dKO and double conditional knockout of *miR-34b/c* and *miR-449* in multi-ciliated cells (*Foxj1*-Cre-dcKO) exhibit reduced cilia number, increased sperm agglutination, and clogging of the EDs, highlighting the pivotal role of these five microRNAs in ciliogenesis and male fertility.[10] Another study has shown that

miR-34/449 regulates multiciliogenesis in the EDs by modulating cell-cycle exit. Forcing cell-cycle exit by cyclin-dependent kinase inhibitors can promote the differentiation of MCCs and partially rescue the defective multiciliogenesis in the EDs of dKO mice. This finding advances our understanding of the regulatory pathways and mechanisms of miR-34/449 in ciliogenesis.[74]

Interestingly, conditional mutation of E2F4 specifically in the epithelial cells of the EDs using *Villin*^{Cre}, coupled with a heterozygous mutation in *E2F5* (*E2F4*$^{fl/fl}$/*E2F5*$^{+/-}$; *Villin*^{Cre}), leads to loss of MCCs within the EDs and dilation of the rete testis. In addition, this genetic alteration down-regulates the expression of aquaporin1 (AQP1), a key water channel protein, thereby inhibiting the resorption process within the EDs.[88] Therefore, it reminds us that both ciliary motility and the reabsorption capacity of adjacent non-ciliated cells are crucial for maintaining tubule patency. Knockout of other known transcription factors such as *TAp73*, *Gemc1*, *Mcidas*, and *Ccno* in mice results in reduced cilia number, dilated seminiferous tubules, sperm blockage in the EDs and ultimately male infertility.[62,86] Another study further demonstrated that *Mcidas* acts upstream of *Ccno* to regulate basal body biogenesis in the epithelium of the reproductive tract (EDs and oviducts), consistent with the classical ciliogenesis regulatory network.[23] CEP164, a key distal appendage protein, orchestrates ciliogenesis by recruiting small vesicles to basal bodies, which ensure anchoring of basal bodies to the apical cell surface. Similar phenotypic defects are observed in *Foxj1-Cre*; *CEP164*$^{fl/fl}$ mice, including impaired ciliogenesis, rete testis enlargement, and sperm aggregation within the EDs,[89] suggesting that CEP164 is essential for ciliogenesis and male fertility. Additionally, the absence of the axonemal motor protein DNAH5 in mice disrupts the structure of the ODAs in motile cilia, resulting in dilatation of the rete testis and sperm stasis within the EDs. Similarly, in humans, males with mutated *DNAH5* show a severe reduction in sperm count and dilatation of the epididymal head, further confirming the critical role of motile cilia in male fertility.[24]

Unlike other models of infertility caused by the complete absence of cilia, the ductal occlusions in *Arl13b* cKO male mice are attributed to shorter motile cilia and impaired fluid reabsorption, because disruption of ARL13B (*Villin*^{Cre}; *Arl13b*$^{Flox/Flox}$) leads to impairments in cilia architecture, fluid reabsorption, and immune homeostasis, ultimately resulting in fertility issues.[81] In addition, estrogens synthesized by germ cells play a crucial role in the male reproductive system, with the EDs and epididymis being the primary targets for estrogen receptors, including ESR1 and ESR2.[90,91] Research has also shown the presence of estrogen receptors in the ciliated cells of the EDs. Significant abnormalities have been observed in the *Esr1* knockout mouse (*Esr1* KO), including reduced ciliary density per cell, disrupted ciliary arrangement, impaired fluid reabsorption and subsequent fluid accumulation in the rete testis and seminiferous tubules.[9,92,93] However, it is the altered water reabsorption capacity specifically in non-ciliated cells that is suggested to be the primary driver of fluid accumulation in *Esr1* KO mice. Thus, the precise molecular pathway involving Esr1 and estrogen in the regulation of ciliary function in EDs remains elusive.

3.3 | Comparison of sperm flagella and motile cilia

In clinical practice, dysfunction of both the motile cilia in ED and the sperm flagellum can lead to male infertility. For example, PCD is a rare genetic disorder caused by cilia motility dysfunction with pathological symptoms such as recurrent respiratory infections, hydrocephalus, and male infertility.[94,95] Infertile males often present with sperm motility disorders, which result from the fact that the sperm flagellum has an axoneme structure that is reminiscent of that found in cilia. Both structures have a '9+2' microtubule arrangement in their axonemes, and mutations in PCD-related genes can alter the structure of the axoneme, leading to reduced sperm viability and infertility. However, some male patients with PCD and mouse models with defective cilia suggest that male infertility may be caused by damage to the motile cilia in the ejaculate and is not related to the structure of the sperm itself. For example, *Dnah5* deficient mice show an impaired ciliary structure in the EDs, whereas the sperm flagella display a normal microstructural appearance.[24] In contrast, the absence of *Dnah10*, a heavy chain component of the endokinetic protein arm, results in abnormal sperm morphology and subsequent male infertility without other typical PCD symptoms.[96] Additionally, *Odad3* has been reported to be associated with PCD; however, deletion of *Odad3* in mice results in multiple morphological abnormalities of sperm flagella, while the EDs exhibit normal ciliogenesis.[97] The causes of these phenotypic differences may be related to the genes that are expressed specifically in cilia and flagella. For example, the components of the ODA heavy chain, DNAH5, DNAH9 and DNAH11, are uniquely expressed in cilia, whereas DNAH17 and DNAH8 are found exclusively in mature sperm flagella. Accordingly, individuals carrying DNAH17 mutations show axonemal defects in sperm flagella without other clinical symptoms of PCD.[98] In conclusion, in clinical practice it is important to recognize that there is no direct and inevitable relationship between impaired cilia and reduced sperm motility. The function of the ED cilia should also be an important factor for clinicians to consider when diagnosing oligozoospermia and asthenozoospermia.

Differences in their surrounding accessory structures, axoneme assembly mechanisms, and motility patterns further distinguish their functions and regulatory mechanisms.[27,99] In terms of structure, the fibrous sheath, outer dense fibers, and mitochondrial sheath, which are absent in ciliary structures, surround the axoneme of the sperm flagellum and play a crucial role in sperm energy metabolism, viability and stability of the axoneme structure.[100,101] Furthermore, the motile cilia in EDs have a unique structure at their tips known as the ciliary crown, a claw-like complex that protrudes from the plasma membrane. This feature, first observed in oviduct cilia, remains enigmatic in its specific function within the EDs, but is absent in sperm flagella, further emphasizing the structural and functional differences between motile cilia and sperm flagella.[9] Notably, a recent study has analyzed the 96-nm modular repeat of axonemal DMTs in epithelial motile cilia and sperm flagella using cryo-electron microscopy, cryo-electron tomography, and proteomics. It shows that sperm flagella have more complex DMTs and specifically contain over 30 types of proteins. They propose that sperm-specific proteins are sequentially linked to general

axonemal proteins, which may explain why mutations in these conserved axonemal proteins have a more pronounced effect on sperm flagella compared to other motile cilia. In addition, structurally, sperm flagella have a TRiC chaperone specifically suspended between RS1 and RS2, which plays a mechanical regulatory role and assists in microtubule assembly.[43] Despite their differences, the structural integrity of both ED cilia and sperm flagella is critical for efficient sperm transport and overall male fertility.

4 | MOTILE CILIA IN THE FEMALE REPRODUCTIVE SYSTEM

4.1 | Functional significance of motile cilia in the oviduct

The mouse oviduct (analogous to the fallopian tube in humans) consists of four parts: infundibulum, ampulla, isthmus, and uterotubal junction, and the inner mucosa has a columnar epithelium with non-ciliated secretory and ciliated cells. The specific markers identifying the cell types are shown in Figure 2C.[102,103] Through scRNA-seq analysis of the human fallopian tubes (FTs), four subpopulations of ciliated cells were identified, distinguished by high expression of genes associated with motile cilia, such as *FOXJ1*, *CAPS*, *CFAP157*, *CFAP73*, *CFAP52*, *CFAP126*, and various dynein components, including *DYNLL1*, *DNAI1*, *DNAI2*, *DNAH11*, *KIF21A*, etc. [102] The motile cilia extending from the surface of the MCCs are prominently located at the tips of the mucosal folds.[18] The infundibulum harbors the highest density of ciliated cells, comprising approximately 80% of the epithelial cell population.[19,20] In human FTs, the initial presence of ciliated cells is documented at 18 weeks of gestation.[104] However, in mice, ciliated cells are observed histologically at P4, characterized by the first appearance of acetylated tubulin (ac-TUB), whereas mature cilia are presented at P12.[105] Furthermore, as development progresses, secretory cells gradually differentiate into ciliated cells,[21] a phenomenon that has not been documented in male EDs.

Of note, the oviduct efficiently transports gametes via three coordinated pathways: smooth muscle contraction, motile cilia beating, and fluid flow.[106] Interestingly, studies have shown that inhibition of smooth muscle motility in vitro does not affect the transport of oocytes and embryos, suggesting that smooth muscle contractions may not be essential for the gamete transport.[107–109] In addition, studies in patients with Kartagener syndrome have shown that ciliary axoneme defects impair ciliary motility but paradoxically preserve fertility in some females, suggesting a dispensable role for FT cilia.[110–112] However, research in infertile patients has shown that FTs have only 20% of the normal number of cilia and this, combined with a possible reduction in cilia beating frequency, may contribute to infertility.[113–115] Moreover, reduced levels of DNAH5 in cervical fluid have been associated with ectopic pregnancy and endometriosis, suggesting the wider impact of cilia dysfunction on reproductive health.[39,116] In particular, our recent landmark study has shown that mouse motile cilia defects result in the retention of cumulus–oocyte complexes in the ovary, highlighting the critical role of infundibular motile cilia in oocyte retrieval. In contrast, the absence of motile cilia in other regions of the oviduct does not affect sperm or embryo transport, suggesting a non-essential role for motile cilia in these processes.[117] Thus, the paramount importance of motile cilia in the oviduct epithelium for oocyte capture and female fertility has been widely recognized.

Interestingly, research has shown that in the oviduct of female mammals, sperm heads can bind to the cilia of isthmic epithelial cells, resulting in the formation of a sperm reservoir.[118] Further studies confirmed that after capacitation, the increased exposure of phosphatidylserine on the heads of spermatozoa facilitates their binding to ANXA5, which is specifically expressed on the cilia of isthmic epithelial cells.[119] This discovery reveals a new mechanism by which sperm bind to oviductal cilia and form a sperm reservoir. As a result, this research has deepened our understanding of the cilia function by showing that, in addition to their role in capturing oocytes, cilia also have the potential to bind to the heads of spermatozoa.

4.2 | Factors influencing the ciliary activity in the oviduct

Research suggests that a number of factors, including progesterone (P4), estrogen-17 (E2), angiotensin II, prostaglandins, interleukin-6, Ca^{2+}, and epidermal growth factor, have been identified as influencing cilia beating frequency or ciliogenesis in the female FT.[120–127] In contrast to male reproductive physiology, the ratio of ciliated to secretory cells in the FT epithelium fluctuates throughout the menstrual cycle in response to hormonal changes. The role of steroid hormones in regulating the function of MCCs in the female reproductive tract has been extensively reviewed.[128] For example, E2 has been shown to promote the differentiation of epithelial cells into ciliated cells in vitro and to promote ciliogenesis in rat epithelial cells in vivo by upregulating the expression of *Foxj1*.[129] In addition, ERα and ERβ have been reported to be expressed in oviductal epithelial cells, with ERβ uniquely localized to the ciliary stalk of mouse MCCs.[130,131] However, specific deletion of *Esr1* in MCCs has no effect on ciliogenesis or female fertility ($Foxj1^{Cre/+}$; $Esr1^{f/f}$), suggesting that ERα is dispensable for oviductal ciliogenesis and may primarily play a role in secretory epithelial cells.[132,133] Furthermore, using the FT organoid model, the negative regulatory role of Notch signaling in tubal ciliogenesis was demonstrated.[134] Building on this, further studies show that E2 promotes MCC differentiation in the tubal epithelium via ERβ activation and Notch signaling repression, accompanied by *Foxj1* upregulation. Conversely, epidermal growth factor, a key regulator of epithelial homeostasis and differentiation, inhibits ciliogenesis through activation of the Notch signaling pathway.[127] In cattle, the peak abundance of ciliated cells in the oviductal ampulla occurs around ovulation, a time when E2 levels are highest.[135,136] Given this temporal correlation, it is reasonable to speculate that E2 may play a role in stimulating the transformation of secretory cells into ciliated cells within the oviduct during ovulation. Moreover, both progesterone and testosterone levels may also affect ciliary motility. For example, high levels of testosterone

have been shown to decrease the expression of *Foxj1* and subsequently reduce cilia beating frequency in human FT ciliated cells.[120,121,137,138]

It has been demonstrated that the TRPV4 channel, located in the female reproductive tract, can be activated by viscous loading, resulting in an elevation of intracellular Ca^{2+} levels and an increase in ciliary beating frequency.[139] A recent study proposes that under conditions of elevated viscosity in the culture medium, the proportion of ciliated epithelial cells increases significantly, accompanied by activation of the TRPV4 expression, which increases intracellular Ca^{2+} levels, enhances ATP production, and regulates the biophysics of ciliary beating and coordination.[140] Given that steroid hormones, such as P4 and E2, exert control over the secretory function of epithelial cells, resulting in the rheological properties of FT fluid.[141,142] It is reasonable to assume that an increase in estrogen levels during ovulation increases the viscosity of the tubal fluid. This, in turn, facilitates the differentiation of epithelial cells into ciliated cells and simultaneously activates the expression of TRPV4, which affects ciliogenesis and coordination.

Genetic factors also influence the ciliary activity. The kinesin protein Kif19A regulates ciliary length, and its absence leads to excessively long cilia, obstruction of fluid flow, and infertility in female mice.[143] The serine-threonine kinase *Stk36* (Fused or Fu) is essential for the regulation of central microtubules of the axoneme, and its deletion results in random ciliary orientation and impaired motility.[144] In addition, deletion of the planar cell polarity gene *Celsr1* also results in abnormal ciliary motility orientation and female infertility. While it is hypothesized that the primary underlying cause of this infertility is due to the disrupted reproductive tract development, the direct link to ciliary polarity abnormalities remains elusive.[145] Further studies have shown that CAMSAP3, a microtubule regulator located at the base of the cilium, plays a crucial role in regulating intracellular cilia orientation and microtubule assembly associated with basal bodies, while *Celsr1* is mainly responsible for regulating cilia orientation between cells.[146] Furthermore, using the oviduct organoid model, the researchers have discovered that CDC42 coordinates the transformation of secretory cells into MCCs independently of the well-known Notch pathway and that the CDC42-AKT signaling pathway plays a key role in this transformation, uncovering a novel pathway underlying oviduct multiciliogenesis.[147]

4.3 | Motile cilia in human endometrial epithelium

In female mammals, the endometrium is the inner layer of the uterus, including the luminal epithelium and the glandular epithelium. Notably, the luminal epithelial cells within the layers are also composed of secretory and ciliated cells, and the distinctive markers for these two cell types have been demonstrated[148–150] (Figure 2C). In the endometrium of patients with recurrent miscarriages, ciliary abnormalities (such as shortening and fusion) have been observed in MCCs in addition to microvillar abnormalities in secretory epithelial cells. This suggests a possible link between motile cilia in the endometrium and pregnancy outcome.[151] However, the role of motile endometrial cilia is still poorly understood. Research has shown that E2 is the primary driver of ciliogenesis in endometrial epithelial cells.[152] This process may involve the downregulation of Notch signaling or the upregulation of *Gemc1* expression, both of which could contribute to the development of motile cilia in the endometrium.[150] We can therefore conclude that E2 and its receptors (ERα and ERβ) play a pivotal role in the regulation of ciliary function in both the male and female reproductive systems. In addition, a recent study shows that the expression of endometrial ciliogenesis-related genes declines after the age of 35 years, demonstrating that increasing age is responsible for a progressive dysregulation of the endometrial ciliary function.[153] Interestingly, a recent study has come to the opposite conclusion, confirming that older women have an increased abundance of MCCs and increased expression of cilia genes compared to younger women. This phenomenon may be related to cell-cycle regulation and may be influenced by cellular ageing.[154] These findings suggest that ageing, together with possible hormonal imbalances, has a direct influence on the development of MCCs in human endometrial cells. Therefore, it remains an open question whether ageing causes functional changes in ciliated endometrial epithelial cells and whether these changes subsequently contribute to implantation failure. Despite the relative paucity of studies on endometrial cilia compared to those in the FT and ED, recent investigations have highlighted their indispensable role in the endometrial function, which is intricately linked to hormonal fluctuations throughout the menstrual cycle. In particular, the use of endometrial organoid models has emerged as a robust in vitro platform that provides unique insights into the functional mechanisms of endometrial ciliated cells. [150,155]

5 | COMPARISON OF THE ROLE AND REGULATION OF MOTILE CILIA IN THE MALE AND FEMALE REPRODUCTIVE SYSTEMS

5.1 | The progenitor cells of multiciliated cells

Extensive research has been conducted on the progenitor cells and cell fate determination of MCCs in the respiratory tract and brain. Specifically, radial glial cells have been identified as the precursors of ependymal cells in the mouse brain, as evidenced by cell fate tracing experiments.[156] In the large airways, basal cells expressing p63 (also known as TP63) serve as pluripotent progenitors that differentiate into club cells and ciliated cells. These differentiated club cells retain their stem-like properties in the bronchioles, enabling them to further differentiate into ciliated cells.[157,158] A study has shown that during development and injury repair, primary cilia appear transiently on these basal cells. Subsequently, these basal cells specifically differentiate into MCCs, suggesting a potential role for primary cilia in the MCC development.[159] Similarly, basal cells in the epididymis have been shown to have the ability to self-renew and develop into organoids in vitro, suggesting their potential as resident stem cells.[160,161] Studies have revealed that knocking out the primary cilia component ARL13B in these cells leads to impaired Hedgehog signaling and reduced

expression of basal cell markers such as KRT5, KRT14, and p63, which affects basal cell stemness and results in failure to properly regenerate of the epididymal epithelium in vivo.[162] This further suggests a role for primary cilia signaling in maintaining basal cell stemness. However, scRNA-seq data and immunofluorescence staining results indicate the absence of a KRT5-expressing basal cell population in human EDs.[82]

Pseudotime trajectory analysis reveals THY1^{+}/PAX8^{+} early secretory cells in human FTs differentiate into mature secretory cells and subsequently into ciliated cells.[163] Additionally, another study using RNA velocity analysis has shown that mature secretory cells originate from LGR5^{+} or PGR^{+} progenitors and that both mature secretory cells and epithelial–mesenchymal transition cells have the potential to become ciliated cells, supporting the notion of multipotent differentiation of FT epithelial cells.[102] However, the differentiation fate of epithelial stem cells in EDs and the genes and signaling pathways that control their differentiation remain elusive. Increased attention to this puzzle should be a priority for future research efforts.

5.2 | Shared genes influencing ciliogenesis across male and female reproductive systems

Ciliogenesis in both sexes relies on hormonal signals, a delicate balance of local environmental factors and some common regulatory pathways to maintain the optimal ciliary function (Table 1). In particular, some genes can directly regulate the ciliary function in both male and female reproductive tracts. For example, simultaneous deletion of *miR-34b/c* and *miR-449* also results in a large absence of cilia in the oviductal epithelium, which could potentially impede oocyte capture or transport and thus lead to female infertility.[87,117] In *TAp73* KO female mice, reduced cilia coverage, reduced levels of *Dnali1*, *Foxj1*, and *Rfx2*, and mislocalized basal bodies are observed, along with potential defects in the oocyte development that may contribute to female infertility.[62] However, in contrast to Foxj1-Cre; *CEP164*$^{fl/fl}$ male mice, females of this genotype are fertile despite significant loss of cilia in the oviduct epithelium, suggesting that residual cilia are sufficient to maintain the physiological function of the oviduct.[164] In addition, *Gemc1*$^{-/-}$ female mice also exhibited sterility and complete loss of cilia, likely due to impaired differentiation of MCCs. Interestingly, these mutant females also have smaller ovaries and fewer degenerated antral follicles, suggesting that female sterility in this context results from a complex interplay of factors.[52] One study has reported that *Ccno*$^{-/-}$ female mice are also infertile, characterized by a lack of cilia per cell. However, the underlying mechanism responsible for this infertility phenotype remains elusive.[165] In addition, a marked reduction in motile cilia and basal bodies within the infundibulum has been observed in female patients with *Ccno* mutations, leading to failure of oocyte pickup and female infertility.[23] Taken together, key regulatory genes for ciliogenesis and MCC differentiation also affect ciliogenesis in the oviducts and EDs, thereby influencing fertility.

5.3 | The functions of motile cilia in the reproductive tract in relation to human fertility

The current concept suggests that the function of motile cilia in the reproductive system is critical for both male and female fertility. Specifically, in the female oviduct, motile cilia direct unidirectional fluid flow, ensuring efficient capture of the egg and its transport to the ampulla.[34,139] In the male ED, motile cilia employ irregular whipping motions to exert centripetal forces, creating turbulence that maintains sperm suspension and prevents occlusion.[10] However, given that deletion of certain key ciliary genes in females does not produce the profound infertility phenotype observed in males, it is reasonable to conclude that only ciliary dysfunction specifically in the infundibular region of the FT contributes to female infertility. And the motile cilia in the EDs are more necessary than those in the oviduct for maintaining normal fertility.

In clinical practice, multiple cases of infertility have been linked to ciliopathies. Infertility is a common feature of PCD, affecting 71 out of 119 female patients and 167 out of 192 male patients.[26] Although sperm flagellar abnormalities may contribute to reduced fertility in male PCD patients, one study found that PCD males with loss-of-function DNAH5 mutations had caput epididymal dilatation and obstructive azoospermia (or oligozoospermia), but their sperm had normal ultrastructure and motility, suggesting that impaired cilia in the EDs are a direct cause of infertility.[24] However, the specific mechanisms underlying infertility in PCD patients remain poorly understood. In men, possible mechanisms include obstruction of the EDs due to damaged motile cilia or impaired sperm motility. In women, it has been speculated that abnormal ciliary motility in the FTs and endometrium may impair oocyte and early embryo transport, interfering with fertilization and embryo implantation.[26] Furthermore, in addition to infertility directly caused by ciliary dysfunction, several gynecological diseases in women are closely related to FT cilia, such as salpingitis, endometriosis and polycystic ovary syndrome.[39,166] After Neisseria gonorrhoeae infects the FTs, it triggers an inflammatory response, inducing an increase in tumor necrosis factor alpha levels, which in turn leads to reduced ciliary activity and the death of MCCs.[167] In the peritoneal fluid of patients with endometriosis, a macromolecular ovum capture inhibitor forms a membrane on the surface of cilia, thereby inhibiting ciliary activity. In addition, components of the proinflammatory peritoneal fluid may also have a direct effect on ciliary motility.[168] Patients with polycystic ovary syndrome are known to have high levels of testosterone.[169] Under high testosterone exposure, the expression of FOXJ1 in human FT epithelial cells is inhibited,[137] suggesting that these patients may have an increased risk of infertility due to high testosterone levels. It is worth noting that DNAH5 levels are reduced in cervical fluid from patients with ectopic pregnancy, suggesting possible ciliary abnormalities. Therefore, the level of DNAH5 is promising as an effective basis for early diagnosis of ectopic pregnancy.[116] On the other hand, it has been reported that various drugs are known to alter the beat frequency of respiratory cilia.[170] Therefore, when administering drugs, particular attention should be paid to the potential impact

TABLE 1 Common regulators affecting the cilia function within male and female reproductive tract.

Name	Functions in MCC	Description	Phenotypes in mutant individuals	References
miR-34b/c and miR-449	Enforcing cell cycle exit; MCC differentiation	*miR34b/c*$^{-/-}$/*miR449*$^{-/-}$; Foxj1-dcKO male mice	Reduced cilia number and ciliated cells in EDs; Sperm aggregation clogged the EDs; Fluid accumulation in the germinal tubules; Large sperm granulomas in the rete testis; infertile	10,87,171
		miR34b/c$^{-/-}$/*miR449*$^{-/-}$ female mice	Reduced number of the oviduct epithelial cilia; infertile	87
TAp73	Transcription factor; MCC differentiation; Upstream of Foxj1, Rfx2/3, and miR34b/c	*TAp73* knockout male mice	Reduced sperm number in testis and no mature spermatozoa in epididymis; reduced number and length of cilia; almost absent expression of Dnali1, Foxj1, Rfx2, and Rfx3; infertile	62
		TAp73 knockout female mice	Reduced cilia coverage of the oviduct epithelium; mislocated basal bodies; reduced levels of Dnali1, Foxj1 and Rfx2; defects of oocyte development; infertile	62
Gemc1	Transcription factor; MCC differentiation	*Gemc1*$^{-/-}$ male mice	Thinning of the seminiferous germinal epithelia; decreased number of round spermatids and elongated spermatids; dilation of the seminiferous tubules and rete testes; sperm agglutinations in the EDs; Sertoli cell degeneration; infertile	52,86
		Gemc1$^{-/-}$ female mice	Complete loss of cilia; no Foxj1 or Rfx3 signals in the oviduct; smaller ovaries and few degenerated antral follicles; infertile	52]
CCNO	Deuterosome formation and centriole amplification	*Ccno*$^{-/-}$ male mice	Dilation of the seminiferous tubules and rete testes; sperm agglutinations in the EDs; no sperm in epididymis; infertile	86
		Ccno$^{-/-}$ female mice	Very few cilia per cell of the oviduct epithelium; the expression of Foxj1 has no significant change; infertile	165
		Female patients with *Ccno* mutations	Decreased number of cilia and basal bodies of epithelial cells in infundibulum; failure of oocyte pickup; infertile	23
CEP164	Distal appendage component; basal body docking	Foxj1-Cre; *CEP164*$^{fl/fl}$ male mice	Dilation of the seminiferous tubules and rete testes; sperm aggregation and agglutination in the seminiferous tubules and EDs; loss of multicilia; reduced sperm counts; infertile	89
		Foxj1-Cre; *CEP164*$^{fl/fl}$ female mice	Reduced number of multicilia; fertile	164
DNAH5	Axonemal motor protein	*Dnah5*$^{mut/mut}$ male mice	Dilatation of the rete testis; sperm stasis in EDs; ciliary dysmotility; infertile	24
		Male patients with *DNAH5* mutations	Reduced sperm numbers; dilatation of the epididymal head; infertile	24
		Female patients with *DNAH5* mutations	A proportion of the patients are infertile	172
E2(or estrogen receptor)	Inducing differentiation of epithelial cells into MCC	*Esr1* knockout male mice	Reduced number of cilia per cell; disorganized arrangement of cilia; the random beating of cilia; impaired fluid reabsorption and accumulation in rete testis and seminiferous tubules; infertile	9,92,93
		Foxj1$^{Cre/+}$; *Esr1*$^{f/f}$ female mice	Normal cilia formation; fertile	132
		E2 treatment of human fallopian tube epithelial cells cultured in vitro	Differentiation of epithelial cells into ciliated cells	129
		E2 treatment of porcine fallopian tube epithelial cells cultured in vitro	MCCs appeared after E2 treatment, while very few of them were observed when treated with progesterone; downregulation of Notch ligand and Notch intracellular domain; upregulation of Foxj1	127
		Endometrial organoids cultured in the presence of E2	Upregulation of genes related to cilia formation and function; ciliated cells are observed in the endometrial organoids treated with E2; downregulation of the NOTCH1 receptor	150

Abbreviations: E2, estrogen; ED, efferent duct; MCC, multiciliated cell.

on the function of cilia in the reproductive tract, which may lead to fertility-related side effects.

6 | CONCLUSIONS AND PERSPECTIVES

Recent studies of motile cilia in the reproductive system, coupled with the use of knockout models for cilia-associated proteins, have unequivocally illuminated their pivotal role in the transportation of gametes. Despite differences in ciliary beating patterns and ciliary functions between ED and oviductal cilia, they share some key regulatory factors such as *miR-34/449*, *Gemc1* and *Ccno*. The functions of motile cilia in the human reproductive tract have recently been reviewed.[173] We further summarize the common regulatory factors affecting cilia in both male and female reproductive tracts, with a focus on the origin and development of MCC and cilia in the ED and oviducts. We also cite the recent article to discuss the molecular structural differences between cilia and sperm flagella, providing a reference for further in-depth research on cilia functions and infertility treatment in clinical practice. Nevertheless, many questions remain unanswered, particularly regarding the ancestral origins and developmental trajectories of ciliated cells in EDs, the precise timing of ciliogenesis, and whether additional, undiscovered cell populations reside in these reproductive tracts. Future endeavors ought to harness sophisticated techniques such as organoid modeling, scRNA-seq, and spatial transcriptome profiling. These advanced methods have great potential to elucidate diverse cell types, intricate cell–cell communication and dynamic developmental trajectories, thereby improving our understanding of the MCC development and ciliogenesis within the reproductive system. It may also provide new insights into the causes of obstructive oligospermia and facilitate genetic counseling and prenatal diagnosis.

AUTHOR CONTRIBUTIONS

Shiyu Yang, Xiaoli Wang, and Huihui Gao reviewed the literature. Shuiqiao Yuan wrote and revised the manuscript.

ACKNOWLEDGMENTS

We thank members of the Yuan laboratory for their constructive critique of this manuscript. We apologize to any authors whose work we have unfortunately omitted due to space constraints.This work was supported by grants from the National Natural Science Foundation of China (No. 82371625) and National Key Research and Development Program of China (No. 2024YFA1108100).

CONSENT FOR PUBLICATION

The authors declare no competing interests and agree with the publication.

ORCID

Shuiqiao Yuan https://orcid.org/0000-0003-1460-7682

REFERENCES

1. Wheatley DN, Wang AM, Strugnell GE. Expression of primary cilia in mammalian cells. *Cell Biol Int*. 1996;20(1):73-81.
2. Singla V, Reiter JF. The primary cilium as the cell's antenna: signaling at a sensory organelle. *Science*. 2006;313(5787):629-633.
3. Spassky N, Meunier A. The development and functions of multiciliated epithelia. *Nat Rev Mol Cell Biol*. 2017;18(7):423-436.
4. Roberson EC, Tran NK, Konjikusic MJ, Fitch RD, Gray RS, Wallingford JB. A comparative study of the turnover of multiciliated cells in the mouse trachea, oviduct, and brain. *Dev Dyn*. 2020;249(7):898-905.
5. Mahjoub MR, Nanjundappa R, Harvey MN. Development of a multiciliated cell. *Curr Opin Cell Biol*. 2022;77:102105.
6. Hagiwara H, Ohwada N, Takata K. Cell biology of normal and abnormal ciliogenesis in the ciliated epithelium. *Int Rev Cytol*. 2004;234:101-141.
7. Rosenbaum JL, Child FM. Flagellar regeneration in protozoan flagellates. *J Cell Biol*. 1967;34(1):345-364.
8. Witman GB. The site of in vivo assembly of flagellar microtubules. *Ann N Y Acad Sci*. 1975;253:178-191.
9. Hess RA. Small tubules, surprising discoveries: from efferent ductules in the turkey to the discovery that estrogen receptor alpha is essential for fertility in the male. *Anim Reprod*. 2015;12(1):7-23.
10. Yuan S, Liu Y, Peng H, et al. Motile cilia of the male reproductive system require miR-34/miR-449 for development and function to generate luminal turbulence. *Proc Natl Acad Sci USA*. 2019;116(9):3584-3593.
11. Clulow J, Jones RC, Hansen LA. Micropuncture and cannulation studies of fluid composition and transport in the ductuli efferentes testis of the rat: comparisons with the homologous metanephric proximal tubule. *Exp Physiol*. 1994;79(6):915-928.
12. Nakata H, Iseki S. Three-dimensional structure of efferent and epididymal ducts in mice. *J Anat*. 2019;235(2):271-280.
13. Benoît J. Recherches anatomiques, cytologiques et histophysiologiques, sur les voies excrétices du testicules chez les mammiferes. *Arch Anat Histol Embryol*. 1926;5:173.
14. Omotehara T, Wu Xi, Kuramasu M, Itoh M. Connection between seminiferous tubules and epididymal duct is originally induced before sex differentiation in a sex-independent manner. *Dev Dyn*. 2020;249(6):754-764.
15. Omotehara T, Nakata H, Nagahori K, Itoh M. Comparative anatomy on the development of sperm transporting pathway between the testis and mesonephros. *Histochem Cell Biol*. 2022;157(3):321-332.
16. Omotehara T, Hess RA, Nakata H, Birch LA, Prins GS, Itoh M. Expression patterns of sex steroid receptors in developing mesonephros of the male mouse: three-dimensional analysis. *Cell Tissue Res*. 2023;393(3):577-593.
17. Yoshimura S, Omotehara T, Nakata H, et al. Mesonephric tubules expressing estrogen and androgen receptors remain in the rete ovarii of adult mice. *Cell Tissue Res*. 2024;399:85-96.
18. Patek E. The epithelium of the human fallopian tube. A surface ultrastructural and cytochemical study. *Acta Obstet Gynecol Scand Suppl*. 1974;31(0):1-28.
19. Yamanouchi H, Umezu T, Tomooka Y. Reconstruction of oviduct and demonstration of epithelial fate determination in mice. *Biol Reprod*. 2010;82(3):528-533.
20. Stewart CA, Behringer RR. Mouse oviduct development. *Results Probl Cell Differ*. 2012;55:247-262.
21. Ghosh A, Syed SM, Tanwar PS. In vivo genetic cell lineage tracing reveals that oviductal secretory cells self-renew and give rise to ciliated cells. *Development*. 2017;144(17):3031-3041.
22. Hando T, Okada DM, Zamboni L. Atypical cilia in human endometrium. *J Cell Biol*. 1968;39(2):475-481.
23. Ma C, Wu H, Zhu D, et al. Bi-allelic mutations in MCIDAS and CCNO cause human infertility associated with abnormal gamete transport. *Clin Genet*. 2021;100(6):731-742.
24. Aprea I, Nöthe-Menchen T, Dougherty GW, et al. Motility of efferent duct cilia aids passage of sperm cells through the male reproductive system. *Mol Hum Reprod*. 2021;27(3):gaab009.

25. Phillips DM, Jow WW, Goldstein M. Testis factors that may regulate gene expression: evidence from a patient with Kartagener's syndrome. *J Androl*. 1995;16(2):158-162.
26. Newman L, Chopra J, Dossett C, et al. The impact of primary ciliary dyskinesia on female and male fertility: a narrative review. *Hum Reprod Update*. 2023;29(3):347-367.
27. Fliegauf M, Benzing T, Omran H. When cilia go bad: cilia defects and ciliopathies. *Nat Rev Mol Cell Biol*. 2007;8(11):880-893.
28. Ishikawa H, Marshall WF. Ciliogenesis: building the cell's antenna. *Nat Rev Mol Cell Biol*. 2011;12(4):222-234.
29. Hirokawa N, Tanaka Y, Okada Y, Takeda S. Nodal flow and the generation of left–right asymmetry. *Cell*. 2006;125(1):33-45.
30. Al Jord A, Lemaître A-I, Delgehyr N, Faucourt M, Spassky N, Meunier A. Centriole amplification by mother and daughter centrioles differs in multiciliated cells. *Nature*. 2014;516(7529):104-107.
31. Zhao H, Zhu L, Zhu Y, et al. The Cep63 paralogue Deup1 enables massive de novo centriole biogenesis for vertebrate multiciliogenesis. *Nat Cell Biol*. 2013;15(12):1434-1444.
32. Klos Dehring DA, Vladar EK, Werner ME, Mitchell JW, Hwang P, Mitchell BJ. Deuterosome-mediated centriole biogenesis. *Dev Cell*. 2013;27(1):103-112.
33. Mercey O, Levine MS, Lomastro GM, et al. Massive centriole production can occur in the absence of deuterosomes in multiciliated cells. *Nat Cell Biol*. 2019;21(12):1544-1552.
34. Mizuno N, Taschner M, Engel BD, Lorentzen E. Structural studies of ciliary components. *J Mol Biol*. 2012;422(2):163-180.
35. Rosenbaum JL, Witman GB. Intraflagellar transport. *Nat Rev Mol Cell Biol*. 2002;3(11):813-825.
36. Pazour GJ, Dickert BL, Vucica Y, et al. Chlamydomonas IFT88 and its mouse homologue, polycystic kidney disease gene tg737, are required for assembly of cilia and flagella. *J Cell Biol*. 2000;151(3):709-718.
37. Wong LB, Miller IF, Yeates DB. Nature of the mammalian ciliary metachronal wave. *J Appl Physiol*. 1993;75(1):458-467.
38. O'Callaghan C, Sikand K, Chilvers MA. Analysis of ependymal ciliary beat pattern and beat frequency using high speed imaging: comparison with the photomultiplier and photodiode methods. *Cilia*. 2012;1(1):8.
39. Lyons RA, Saridogan E, Djahanbakhch O. The reproductive significance of human fallopian tube cilia. *Hum Reprod Update*. 2006;12(4):363-372.
40. Huitorel P. From cilia and flagella to intracellular motility and back again: a review of a few aspects of microtubule-based motility. *Biol Cell*. 1988;63(2):249-258.
41. Otokawa M. Adenylate kinase activity. In ciliary axonemes isolated from Tetrahymena pyriformis. *J Biochem*. 1974;76(2):335-340.
42. Schoff PK, Cheetham J, Lardy HA. Adenylate kinase activity in ejaculated bovine sperm flagella. *J Biol Chem*. 1989;264(11):6086-6091.
43. Leung MR, Sun C, Zeng J, et al. Structural diversity of axonemes across mammalian motile cilia. *Nature*. 2025;637:1170-1177. Advanced online publication. doi: doi.org/10.1038/s41586-024-08337-5
44. Witman GB, Plummer J, Sander G. Chlamydomonas flagellar mutants lacking radial spokes and central tubules. Structure, composition, and function of specific axonemal components. *J Cell Biol*. 1978;76(3):729-747.
45. Wargo MJ, Smith EF. Asymmetry of the central apparatus defines the location of active microtubule sliding in Chlamydomonas flagella. *Proc Natl Acad Sci USA*. 2003;100(1):137-142.
46. Thomas J, Morlé L, Soulavie F, Laurençon A, Sagnol S, Durand B. Transcriptional control of genes involved in ciliogenesis: a first step in making cilia. *Biol Cell*. 2010;102(9):499-513.
47. Choksi SP, Lauter G, Swoboda P, Roy S. Switching on cilia: transcriptional networks regulating ciliogenesis. *Development*. 2014;141(7):1427-1441.
48. Marcet B, Chevalier B, Luxardi G, et al. Control of vertebrate multiciliogenesis by miR-449 through direct repression of the Delta/Notch pathway. *Nat Cell Biol*. 2011;13(6):693-699.
49. Tsao Po-N, Vasconcelos M, Izvolsky KI, Qian J, Lu J, Cardoso WV. Notch signaling controls the balance of ciliated and secretory cell fates in developing airways. *Development*. 2009;136(13):2297-2307.
50. Rock JR, Gao X, Xue Y, Randell SH, Kong Y-Y, Hogan BLM. Notch-dependent differentiation of adult airway basal stem cells. *Cell Stem Cell*. 2011;8(6):639-648.
51. Arbi M, Pefani D-E, Kyrousi C, et al. GemC1 controls multiciliogenesis in the airway epithelium. *EMBO Rep*. 2016;17(3):400-413.
52. Terré B, Piergiovanni G, Segura-Bayona S, et al. GEMC1 is a critical regulator of multiciliated cell differentiation. *EMBO J*. 2016;35(9):942-960.
53. Stubbs JL, Vladar EK, Axelrod JD, Kintner C. Multicilin promotes centriole assembly and ciliogenesis during multiciliate cell differentiation. *Nat Cell Biol*. 2012;14(2):140-147.
54. Ma L, Quigley I, Omran H, Kintner C. Multicilin drives centriole biogenesis via E2f proteins. *Genes Dev*. 2014;28(13):1461-1471.
55. Funk MC, Bera AN, Menchen T, et al. Cyclin O (Ccno) functions during deuterosome-mediated centriole amplification of multiciliated cells. *EMBO J*. 2015;34(8):1078-1089.
56. Tan FE, Vladar EK, Ma L, Fuentealba LC, et al. Myb promotes centriole amplification and later steps of the multiciliogenesis program. *Development*. 2013;140(20):4277-4286.
57. Arbi M, Pefani D-E, Taraviras S, Lygerou Z. Controlling centriole numbers: geminin family members as master regulators of centriole amplification and multiciliogenesis. *Chromosoma*. 2018;127(2):151-174.
58. Pardo-Saganta A, Tata PR, Law BM, et al. Parent stem cells can serve as niches for their daughter cells. *Nature*. 2015;523(7562):597-601.
59. Marques MM, Villoch-Fernandez J, Maeso-Alonso L, Fuertes-Alvarez S, Marin MC. The Trp73 mutant mice: a ciliopathy model that uncouples ciliogenesis from planar cell polarity. *Front Genet*. 2019;10:154.
60. Nemajerova A, Kramer D, Siller SS, et al. TAp73 is a central transcriptional regulator of airway multiciliogenesis. *Genes Dev*. 2016;30(11):1300-1312.
61. Marshall CB, Mays DJ, Beeler JS, et al. 73 is required for multiciliogenesis and regulates the Foxj1-associated gene network. *Cell Rep*. 2016;14(10):2289-2300.
62. Wildung M, Esser TU, Grausam KB, et al. Transcription factor TAp73 and microRNA-449 complement each other to support multiciliogenesis. *Cell Death Differ*. 2019;26(12):2740-2757.
63. Clevidence DE, Overdier DG, Peterson RS, et al. Members of the HNF-3/forkhead family of transcription factors exhibit distinct cellular expression patterns in lung and regulate the surfactant protein B promoter. *Dev Biol*. 1994;166(1):195-209.
64. Hackett BP, Brody SL, Liang M, Zeitz ID, Bruns LA, Gitlin JD. Primary structure of hepatocyte nuclear factor/forkhead homologue 4 and characterization of gene expression in the developing respiratory and reproductive epithelium. *Proc Natl Acad Sci USA*. 1995;92(10):4249-4253.
65. Murphy DB, Seemann S, Wiese S, Kirschner R, Grzeschik KH, Thies U. The human hepatocyte nuclear factor 3/fork head gene FKHL13: genomic structure and pattern of expression. *Genomics*. 1997;40(3):462-469.
66. Chen J, Knowles HJ, Hebert JL, Hackett BP. Mutation of the mouse hepatocyte nuclear factor/forkhead homologue 4 gene results in an absence of cilia and random left–right asymmetry. *J Clin Invest*. 1998;102(6):1077-1082.

67. Brody SL, Yan XH, Wuerffel MK, Song S-K, Shapiro SD. Ciliogenesis and left–right axis defects in forkhead factor HFH-4-null mice. *Am J Respir Cell Mol Biol*. 2000;23(1):45-51.
68. Gomperts BN, Gong-Cooper X, Hackett BP. Foxj1 regulates basal body anchoring to the cytoskeleton of ciliated pulmonary epithelial cells. *J Cell Sci*. 2004;117:1329-1337.
69. Stubbs JL, Oishi I, Izpisúa Belmonte JC, Kintner C. The forkhead protein Foxj1 specifies node-like cilia in Xenopus and zebrafish embryos. *Nat Genet*. 2008;40(12):1454-1460.
70. Yu X, Ng CP, Habacher H, Roy S. Foxj1 transcription factors are master regulators of the motile ciliogenic program. *Nat Genet*. 2008;40(12):1445-1453.
71. Bonnafe E, Touka M, Aitlounis A, et al. The transcription factor RFX3 directs nodal cilium development and left–right asymmetry specification. *Mol Cell Biol*. 2004;24(10):4417-4427.
72. Chung M-I, Peyrot SM, Leboeuf S, et al. RFX2 is broadly required for ciliogenesis during vertebrate development. *Dev Biol*. 2012;363(1):155-165.
73. Chung MI, Kwon T, Tu F, et al. Coordinated genomic control of ciliogenesis and cell movement by RFX2. *Elife*. 2014;3: e01439.
74. Jerber J, Thomas J, Durand B. Transcriptional control of ciliogenesis in animal development. *Biol Aujourdhui*. 2012;206(3):205-218.
75. Ou G, E Blacque O, Snow JJ, Leroux MR, Scholey JM. Functional coordination of intraflagellar transport motors. *Nature*. 2005;436(7050):583-587.
76. Fort C, Bastin P. Elongation of the axoneme and dynamics of intraflagellar transport. *Med Sci*. 2014;30(11):955-961.
77. Omotehara T, Nakata H, Itoh M. Three-dimensional analysis of mesonephric tubules remodeling into efferent tubules in the male mouse embryo. *Dev Dyn*. 2022;251(3):513-524.
78. Nakata H, Iseki S, Mizokami A. Three-dimensional analysis of junctions between efferent and epididymal ducts in the human caput epididymis. *Andrology*. 2024;12(1):87-97.
79. Vinay L, Hess RA, Belleannée C. Human efferent ductules and epididymis display unique cell lineages with motile and primary cilia. *Andrology*. 2024, Advance onlie publication. doi: doi.org/10.1111/andr.13732
80. Girardet L, Augière C, Asselin M-P, Belleannée C. Primary cilia: biosensors of the male reproductive tract. *Andrology*. 2019;7(5):588-602.
81. Augière C, Campolina-Silva G, Vijayakumaran A, et al. ARL13B controls male reproductive tract physiology through primary and Motile Cilia. *Commun Biol*. 2024;7(1):1318.
82. Leir SH, Paranjapye A, Harris A. Functional genomics of the human epididymis: further characterization of efferent ducts and model systems by single-cell RNA sequencing analysis. *Andrology*. 2024;12(5):991-1000.
83. Becker O. Uber flimmerepithelium im nebenhoden des menschen. *Wien Wchnschr*. 1856;6:184.
84. Talo A. In-vitro spontaneous electrical activity of rat efferent ductules. *J Reprod Fertil*. 1981;63(1):17-20.
85. Ilio KY, Hess RA. Structure and function of the ductuli efferentes: a review. *Microsc Res Tech*. 1994;29(6):432-467.
86. Terré B, Lewis M, Gil-Gómez G, et al. Defects in efferent duct multiciliogenesis underlie male infertility in GEMC1-, MCIDAS- or CCNO-deficient mice. *Development*. 2019;146(8):dev162628.
87. Wu J, Bao J, Kim M, et al. Two miRNA clusters, miR-34b/c and miR-449, are essential for normal brain development, motile ciliogenesis, and spermatogenesis. *Proc Natl Acad Sci USA*. 2014;111(28):E2851-E2857.
88. Danielian PS, Hess RA, Lees JA. E2f4 and E2f5 are essential for the development of the male reproductive system. *Cell Cycle*. 2016;15(2):250-260.
89. Hoque M, Chen D, Hess RA, Li F-Q, Takemaru K-I. CEP164 is essential for efferent duct multiciliogenesis and male fertility. *Reproduction*. 2021;162(2):129-139.
90. Hess RA, Fernandes SAF, Gomes GRO, Oliveira CA, Lazari MFM, Porto CS. Estrogen and its receptors in efferent ductules and epididymis. *J Androl*. 2011;32(6):600-613.
91. Hess RA, Sharpe RM, Hinton BT. Estrogens and development of the rete testis, efferent ductules, epididymis and vas deferens. *Differentiation*. 2021;118:41-71.
92. Hess RA, Bunick D, Lee Ki-Ho, et al. A role for oestrogens in the male reproductive system. *Nature*. 1997;390(6659):509-512.
93. Hess RA, Bunick D, Lubahn DB, Zhou Q, Bouma J. Morphologic changes in efferent ductules and epididymis in estrogen receptor-alpha knockout mice. *J Androl*. 2000;21(1):107-121.
94. Leigh MW, Pittman JE, Carson JL, et al. Clinical and genetic aspects of primary ciliary dyskinesia/Kartagener syndrome. *Genet Med*. 2009;11(7):473-487.
95. Horani A, Ferkol TW, Dutcher SK, Brody SL. Genetics and biology of primary ciliary dyskinesia. *Paediatr Respir Rev*. 2016;18:18-24.
96. Tu C, Cong J, Zhang Q, et al. Bi-allelic mutations of DNAH10 cause primary male infertility with asthenoteratozoospermia in humans and mice. *Am J Hum Genet*. 2021;108(8):1466-1477.
97. Pasquini M, Chiani F, Gambadoro A, et al. The Odad3 gene is necessary for spermatozoa development and male fertility in mice. *Cells*. 2024;13(12):1053.
98. Whitfield M, Thomas L, Bequignon E, et al. Mutations in DNAH17, encoding a sperm-specific axonemal outer dynein arm heavy chain, cause isolated male infertility due to asthenozoospermia. *Am J Hum Genet*. 2019;105(1):198-212.
99. Sironen A, Shoemark A, Patel M, Loebinger MR, Mitchison HM. Sperm defects in primary ciliary dyskinesia and related causes of male infertility. *Cell Mol Life Sci*. 2020;77(11):2029-2048.
100. Cao W, Gerton GL, Moss SB. Proteomic profiling of accessory structures from the mouse sperm flagellum. *Mol Cell Proteomics*. 2006;5(5):801-810.
101. Lehti MS, Sironen A. Formation and function of sperm tail structures in association with sperm motility defects. *Biol Reprod*. 2017;97(4):522-536.
102. Ulrich ND, Shen Yu-C, Ma Q. Cellular heterogeneity of human fallopian tubes in normal and hydrosalpinx disease states identified using scRNA-seq. *Dev Cell*. 2022;57(7):914-929.
103. Paik DY, Janzen DM, Schafenacker AM, et al. Stem-like epithelial cells are concentrated in the distal end of the fallopian tube: a site for injury and serous cancer initiation. *Stem Cells*. 2012;30(11):2487-2497.
104. Perlman S, Hertweck P, Fallat ME. Paratubal and tubal abnormalities. *Semin Pediatr Surg*. 2005;14:124-134.
105. Agduhr E. Studies on the structure and development of the bursa ovarica and the tuba uterina in the mouse. *Acta Zool*. 1927:8:1-133.
106. Blandau RJ, Boling JL, Halbert S, Verdugo P. Methods for studying oviductal physiology. *Gynecol Invest*. 1975;6(3-4):123-145.
107. Halbert SA, Tam PY, Blandau RJ. Egg transport in the rabbit oviduct: the roles of cilia and muscle. *Science*. 1976;191(4231):1052-1053.
108. Halbert SA, Becker DR, Szal SE. Ovum transport in the rat oviductal ampulla in the absence of muscle contractility. *Biol Reprod*. 1989;40(6):1131-1136.
109. Salathe M. Regulation of mammalian ciliary beating. *Annu Rev Physiol*. 2007;69:401-422.
110. Afzelius BA, Camner P, Mossberg B. On the function of cilia in the female reproductive tract. *Fertil Steril*. 1978;29(1):72-74.
111. Bleau G, Richer CL, Bousquet D. Absence of dynein arms in cilia of endocervical cells in a fertile woman. *Fertil Steril*. 1978;30(3):362-363.
112. Raidt J, Werner C, Menchen T, et al. Ciliary function and motor protein composition of human fallopian tubes. *Hum Reprod*. 2015;30(12):2871-2880.
113. Ul Hassan A, Hassan G, Khan SJ, Rasool Z, Abida A. Ciliopathy with special emphasis on kartageners syndrome. *Int J Health Sci*. 2009;3(1):65-69.

114. Mccomb P, Langley L, Villalon M, Verdugo P. The oviductal cilia and Kartagener's syndrome. *Fertil Steril*. 1986;46(3):412-416.
115. Halbert SA, Patton DL, Zarutskie PW, Soules MR. Function and structure of cilia in the fallopian tube of an infertile woman with Kartagener's syndrome. *Hum Reprod*. 1997;12(1):55-58.
116. Sahin C, Uygun ZO, Hortu I, et al. Using dynein heavy chain 5 and creatine kinase levels in cervical fluid and blood for early diagnosing of ectopic pregnancy. *J Obstet Gynaecol Res*. 2021;47(3):921-927.
117. Yuan S, Wang Z, Peng H, et al. Oviductal motile cilia are essential for oocyte pickup but dispensable for sperm and embryo transport. *Proc Natl Acad Sci*. 2021;118(22):e2102940118.
118. Kölle S. Sperm–oviduct interactions: key factors for sperm survival and maintenance of sperm fertilizing capacity. *Andrology*. 2022;10(5):837-843.
119. Schmaltz L, Barakat E, Fleurot R, et al. Phosphatidylserine on sperm head interact with Annexin A5 on oviduct luminal cilia to form a sperm reservoir in pigs. *Eur J Cell Biol*. 2024;104(1):151471.
120. Mahmood T, Saridogan E, Smutna S, Habib AM, Djahanbakhch O. The effect of ovarian steroids on epithelial ciliary beat frequency in the human fallopian tube. *Hum Reprod*. 1998;13(11):2991-2994.
121. Bylander A, Nutu M, Wellander R, Goksör M, Billig H, Larsson DgJ. Rapid effects of progesterone on ciliary beat frequency in the mouse fallopian tube. *Reprod Biol Endocrinol*. 2010;8:48.
122. Nishimura A, Sakuma Ko, Shimamoto C, et al. Ciliary beat frequency controlled by oestradiol and progesterone during ovarian cycle in guinea-pig fallopian tube. *Exp Physiol*. 2010;95(7):819-828.
123. Saridogan E, Djahanbakhch O, Puddefoot JR, et al. Angiotensin II receptors and angiotensin II stimulation of ciliary activity in human fallopian tube. *J Clin Endocrinol Metab*. 1996;81(7):2719-2725.
124. Spilman CH, Harper MJ. Effects of prostaglandins on oviductal motility and egg transport. *Gynecol Invest*. 1975;6(3-4):186-205.
125. Papathanasiou A, Djahanbakhch O, Saridogan E, Lyons RA. The effect of interleukin-6 on ciliary beat frequency in the human fallopian tube. *Fertil Steril*. 2008;90(2):391-394.
126. Verdugo P. Ca2+-dependent hormonal stimulation of ciliary activity. *Nature*. 1980;283(5749):764-765.
127. Zhu M, Iwano T, Takeda S. Estrogen and EGFR pathways regulate notch signaling in opposing directions for multi-ciliogenesis in the fallopian tube. *Cells*. 2019;8(8):933.
128. Hunter MI, Thies KM, Winuthayanon W. Hormonal regulation of cilia in the female reproductive tract. *Curr Opin Endocr Metab Res*. 2024:34:100503.
129. Comer MT, Leese HJ, Southgate J. Induction of a differentiated ciliated cell phenotype in primary cultures of fallopian tube epithelium. *Hum Reprod*. 1998;13(11):3114-3120.
130. Okada A, Ohta Y, Brody SI, et al. Role of foxj1 and estrogen receptor alpha in ciliated epithelial cell differentiation of the neonatal oviduct. *J Mol Endocrinol*. 2004;32(3):615-625.
131. Shao R, Weijdegård B, Fernandez-Rodriguez J, et al. Ciliated epithelial-specific and regional-specific expression and regulation of the estrogen receptor-beta2 in the fallopian tubes of immature rats: a possible mechanism for estrogen-mediated transport process in vivo. *Am J Physiol Endocrinol Metab*. 2007;293(1):E147-E158.
132. Mcglade EA, Stephens KK, Winuthayanon S, Anamthathmakula P, Holtzman MJ, Winuthayanon W. Classical estrogen signaling in ciliated epithelial cells of the oviduct is nonessential for fertility in female mice. *Endocrinology*. 2023;165(1):bqad163.
133. Li S, O'neill SRS, Zhang Y, et al. Estrogen receptor α is required for oviductal transport of embryos. *Faseb J*. 2017;31(4):1595-1607.
134. Kessler M, Hoffmann K, Brinkmann V, et al. The Notch and Wnt pathways regulate stemness and differentiation in human fallopian tube organoids. *Nat Commun*. 2015;6:8989.
135. Ito S, Kobayashi Y, Yamamoto Y, Kimura K, Okuda K. Remodeling of bovine oviductal epithelium by mitosis of secretory cells. *Cell Tissue Res*. 2016;366(2):403-410.
136. Wijayagunawardane MPB, Miyamoto A, Cerbito WA, Acosta TJ, Takagi M, Sato K. Local distributions of oviductal estradiol, progesterone, prostaglandins, oxytocin and endothelin-1 in the cyclic cow. *Theriogenology*. 1998;49(3):607-618.
137. Jackson-Bey T, Colina J, Isenberg BC, et al. Exposure of human fallopian tube epithelium to elevated testosterone results in alteration of cilia gene expression and beating. *Hum Reprod*. 2020;35(9):2086-2096.
138. Nutu M, Weijdegård B, Thomas P, Thurin-Kjellberg A, Billig H, Larsson DgJ. Distribution and hormonal regulation of membrane progesterone receptors beta and gamma in ciliated epithelial cells of mouse and human fallopian tubes. *Reprod Biol Endocrinol*. 2009;7:89.
139. Andrade YaniréN, Fernandes J, Vázquez E, et al. TRPV4 channel is involved in the coupling of fluid viscosity changes to epithelial ciliary activity. *J Cell Biol*. 2005;168(6):869-874.
140. Abdul Halim MS, Dyson JM, Gong MM, O'bryan MK, Nosrati R. Fallopian tube rheology regulates epithelial cell differentiation and function to enhance cilia formation and coordination. *Nat Commun*. 2024;15(1):7411.
141. Leese Hj, Tay Ji, Reischl J, Downing Sj. Formation of fallopian tubal fluid: role of a neglected epithelium. *Reproduction*. 2001;121(3):339-346.
142. Barton BE, Herrera GG, Anamthathmakula P, et al. Roles of steroid hormones in oviductal function. *Reproduction*. 2020;159(3):R125.
143. Niwa S, Nakajima K, Miki H, Minato Y, Wang D, Hirokawa N. KIF19A is a microtubule-depolymerizing kinesin for ciliary length control. *Dev Cell*. 2012;23(6):1167-1175.
144. Nozawa YI, Yao E, Lin C, et al. Fused (Stk36) is a ciliary protein required for central pair assembly and motile cilia orientation in the mammalian oviduct. *Dev Dyn*. 2013;242(11):1307-1319.
145. Shi D, Komatsu K, Hirao M, et al. Celsr1 is required for the generation of polarity at multiple levels of the mouse oviduct. *Development*. 2014;141(23):4558-4568.
146. Usami FM, Arata M, Shi D, et al. Intercellular and intracellular cilia orientation is coordinated by CELSR1 and CAMSAP3 in oviduct multi-ciliated cells. *J Cell Sci*. 2021;134(4):jcs257006.
147. Jiang R, Tang X, Pan J, et al. CDC42 governs normal oviduct multiciliogenesis through activating AKT to ensure timely embryo transport. *Cell Death Dis*. 2022;13(9):757.
148. Hafez ES, et al. Scanning electron microscopy of human female reproductive tract and amniotic fluid cells. *Int J Fertil*. 1977;22(4):193-205.
149. Cochrane DR, Campbell KR, Greening K, et al. Single cell transcriptomes of normal endometrial derived organoids uncover novel cell type markers and cryptic differentiation of primary tumours. *J Pathol*. 2020;252(2):201-214.
150. Haider S, Gamperl M, Burkard TR, et al. Estrogen signaling drives ciliogenesis in human endometrial organoids. *Endocrinology*. 2019;160(10):2282-2297.
151. Farghaly LM, Atwa KA. Histological changes in the endometrium of women suffering from recurrent pregnancy loss: light and electron microscopic study. *Egypt J Histol*. 2010;33(2):224-235.
152. More IA, Masterton RG. The role of oestrogen in the control of ciliated cells of the human endometrium. *J Reprod Fertil*. 1976;47(1):19-24.
153. Devesa-Peiro A, Sebastian-Leon P, Parraga-Leo A, Pellicer A, Diaz-Gimeno P. Breaking the ageing paradigm in endometrium: endometrial gene expression related to cilia and ageing hallmarks in women over 35 years. *Hum Reprod*. 2022;37(4):762-776.
154. Loid M, Obukhova D, Kask K, et al. Aging promotes accumulation of senescent and multiciliated cells in human endometrial epithelium. *Hum Reprod Open*. 2024;2024(3):hoae048.
155. Abbas Y, Brunel LG, Hollinshead MS, et al. Generation of a three-dimensional collagen scaffold-based model of the human endometrium. *Interface Focus*. 2020;10(2):20190079.

156. Spassky N, Merkle FT, Flames N, et al. Adult ependymal cells are postmitotic and are derived from radial glial cells during embryogenesis. *J Neurosci*. 2005;25(1):10-18.
157. Hong KU, Reynolds SD, Watkins S, Fuchs E, Stripp BR. Basal cells are a multipotent progenitor capable of renewing the bronchial epithelium. *Am J Pathol*. 2004;164(2):577-588.
158. Rock JR, Onaitis MW, Rawlins EL, et al. Basal cells as stem cells of the mouse trachea and human airway epithelium. *Proc Natl Acad Sci USA*. 2009;106(31):12771-12775.
159. Jain R, Pan J, Driscoll JA, et al. Temporal relationship between primary and motile ciliogenesis in airway epithelial cells. *Am J Respir Cell Mol Biol*. 2010;43(6):731-739.
160. Pinel L, Cyr DG. Self-renewal and differentiation of rat epididymal basal cells using a novel in vitro organoid model†. *Biol Reprod*. 2021;105(4):987-1001.
161. Pinel L, Mandon M, Cyr DG. Tissue regeneration and the epididymal stem cell. *Andrology*. 2019;7(5):618-630.
162. Girardet L, Cyr DG, Belleannée C. Arl13b controls basal cell stemness properties and Hedgehog signaling in the mouse epididymis. *Cell Mol Life Sci*. 2022;79(11):556.
163. Dinh HQ, Lin X, Abbasi F, et al. Single-cell transcriptomics identifies gene expression networks driving differentiation and tumorigenesis in the human fallopian tube. *Cell Rep*. 2021;35(2):108978.
164. Siller SS, Sharma H, Li S, et al. Conditional knockout mice for the distal appendage protein CEP164 reveal its essential roles in airway multiciliated cell differentiation. *PLoS Genet*. 2017;13(12):e1007128.
165. Núnez-Ollé M, Jung C, Terré B, et al. Constitutive Cyclin O deficiency results in penetrant hydrocephalus, impaired growth and infertility. *Oncotarget*. 2017;8(59):99261-99273.
166. He L, Xu H, Liu M, et al. The ignored structure in female fertility: cilia in the fallopian tubes. *Reprod Biomed Online*. 2024;50(2):104346.
167. Lenz JD, Dillard JP. Pathogenesis of Neisseria gonorrhoeae and the host defense in ascending infections of human fallopian tube. *Front Immunol*. 2018;9:2710.
168. Suginami H, Yano K. An ovum capture inhibitor (OCI) in endometriosis peritoneal fluid: an OCI-related membrane responsible for fimbrial failure of ovum capture. *Fertil Steril*. 1988;50(4):648-653.
169. Dulohery K, Trottmann M, Bour S, et al. How do elevated levels of testosterone affect the function of the human fallopian tube and fertility? New insights. *Mol Reprod Dev*. 2020;87(1):30-44.
170. Workman AD, Cohen NA. The effect of drugs and other compounds on the ciliary beat frequency of human respiratory epithelium. *Am J Rhinol Allergy*. 2014;28(6):454-464.
171. Wu Yu-J, Liu Y, Hu Y-Q, et al. Control of multiciliogenesis by miR-34/449 in the male reproductive tract through enforcing cell cycle exit. *J Cell Sci*. 2021;134(9):jcs253450.
172. Schreck LD, Pedersen ESL, Dexter K, et al. Infertility and pregnancy outcomes among adults with primary ciliary dyskinesia. *Hum Reprod Open*. 2024;2024(3):hoae039.
173. Wloga D, Joachimiak E, Osinka A, Ahmadi S, Majhi S. Motile cilia in female and male reproductive tracts and fertility. *Cells*. 2024;13(23):1974.

How to cite this article: Yang S, Wang X, Gao H, Yuan S. Motile cilia: Key developmental and functional roles in reproductive systems. *Andrology*. 2025;1-15.
https://doi.org/10.1111/andr.70007