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Mucus and the mare: How little we know

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Abstract

Uterine infections are a major cause of infertility, but the role of mucus in equine uterine defense is not well understood. Mucociliary currents play an important role in protecting mucous membranes, including the upper and lower respiratory tracts of mammals, and are required for feeding and oxygenation of many aquatic invertebrates. Although phagocytosis has long been considered the first line of uterine defense in the mare, there are concerns about its efficacy in the uterine lumen. Additional local defenses, such as mucociliary currents, have therefore been proposed. The uterine epithelium exhibits alternating mucus-secreting and ciliated cells supporting a mucopolysaccharide blanket, features shared with mucociliary membranes throughout the animal kingdom. Gross uterine anatomy, such as continuity of uterine and cervical folds, may indicate adaptations to mucociliary clearance. In addition, ciliated cells obtained in uterine lavages often display motility. Disruptions of mucociliary clearance play major roles in pathogenesis of mucosal infections in humans, including pneumonia, chronic sinusitis, and otitis media. Establishing drainage is a major goal of therapy in treatment of chronic sinusitis, hastening return of mucociliary function. Similar disruptions may occur in equine uterine infections, associated with accumulations of uterine fluid, loss of endometrial folds, and cervical trauma. Possible clinical implications of mucociliary clearance in the mare are discussed, however the role of mucociliary clearance in the mare remains speculative.

Keywords: Mucus; Infertility; Cilia; Uterine defense mechanisms; Horse

1. Introduction

Infertility due to uterine infections is one of the most common problems faced by equine practitioners [1]. A typical clinical presentation is that of an older mare who fails to conceive because she has become susceptible to bacterial infection [2]. In contrast, most young mares, and some older mares, are resistant to uterine bacterial infections [3]. Several studies have implicated failure of physical clearance as underlying susceptibility to infection [4–10]. For example, if uterine secretions accumulate, complement is broken down, reducing the opsonic and bacteriostatic properties of uterine fluid [11]. Interestingly, uterine secretions also appear to contain varying amounts of mucus [12]. Although mucus is well recognized as protecting the respiratory tract [13,14], its role in uterine defense is not clear. The purpose of this review is to discuss available evidence suggesting a role for mucus in equine uterine defense, how those defenses may be disrupted, and possible clinical implications of mucus function and dysfunction. However, it will be seen from what follows that we remain mostly ignorant about the role of mucus in equine uterine defense.

2. Why mucus?

The resistant mare is capable of clearing large numbers of actively growing bacteria from the uterus, preserving her future fertility [3]. How she accomplishes

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this feat is not well understood. Phagocytosis has always been considered the first line of defense against uterine infection, but several lines of evidence (referenced below) suggest that phagocytic efficacy on a mucosal surface may be limited. Preliminary data on the clearance of charcoal and streptococci suggest that mucociliary clearance may supplement phagocytosis in removing bacteria and inflammatory cells from the uterus, but such studies are too limited to be conclusive.

2.1. Concerns regarding phagocytosis

Several lines of evidence suggest that phagocytic efficacy may be reduced in uterine secretions. First, uterine fluid appears to have poor opsonic ability, compared to serum [15], regardless of susceptibility to infection. This poor opsonic ability is not due to a lack of immunoglobulins [16], but the rapid break down of complement in uterine fluid [11,15]. In contrast, effective phagocytosis of streptococci requires a source of both immunoglobulin and complement [17,18]. Thus the neutrophil appears to be severely compromised as soon as it arrives in the uterine lumen, a situation made worse by fluid accumulation [8]. This break down of complement explains why mares showing strong systemic immune responses against *Streptococcus zooepidemicus* do not necessarily have improved protection against uterine infection [19]. However, it does not explain why mares with no measurable systemic immunity to *S. zooepidemicus* are capable of clearing a uterine challenge rapidly [20]. This lack of correlation between uterine clearance and apparent systemic immunity suggests that some additional local defense helps eliminate streptococci. Such local defenses would presumably be located in the endometrium.

2.2. Clearance of charcoal

To reveal possible endometrial clearance mechanisms, a suspension of charcoal powder was infused into the uterus of four reproductively healthy mares. Endometrial biopsy was performed at 4 and 48 h post-infusion, and the endometrial sections stained specifically for mucus (mucicarmine). By 48 h, two of the mares retained charcoal, whereas the remaining two did not [21]. The endometrium of mares which cleared charcoal was comprised of columnar, mucus-containing cells free of charcoal. In one of these biopsies taken at 4 h, charcoal still occupied the uterine lumen, adhered to the cut face of the specimen (exposed lamina propria), but did not adhere to the intact epithelial surface lined by mucus-secreting cells. In the two mares which retained carbon, endometria appeared to have lost the mucous layer, and the carbon was adhered to the endometrial surface. The preliminary interpretation of this small study was that mucus facilitated clearance of charcoal from the uterus by preventing its adhesion to the epithelium.

2.3. Clearance of streptococci

In addition to the charcoal evidence, mares displaying no measurable systemic immune responses against specific strains of *S. zooepidemicus* nevertheless appeared to be able to clear the organism from the uterus, at rates comparable to mares displaying such immunity [20]. Interestingly, some cervical cultures taken post-inoculation displayed steadily increasing numbers of streptococci, when subsequent uterine cultures, and published reports, suggested declining uterine streptococci at the same period post-inoculation [22]. However, concurrent cervical and uterine cultures were not performed in any of these studies. Although no firm conclusions can be drawn, a possible implication is that the uterus voids mostly viable organisms, bactericidal activity not necessarily being the cause of declining streptococcal numbers in the uterus.

2.4. Mucociliary clearance—a hypothesis

Taken as a whole, these preliminary findings raised the possibility that a process of physical bacterial clearance helps achieve bacteriologic sterility of the endometrium. In addition, Metchnikoff's statement that ''nature does not make use of antiseptics to protect the skin and mucous membranes'' might apply to the equine uterus [23]. For the sake of generating a testable hypothesis, an extreme (and probably inaccurate) position was therefore postulated, that bactericidal mechanisms play no role in uterine defense, and physical mechanisms alone achieve sterility of the uterus. If such were the case, an explanation of how the last remaining *Streptococcus* could be removed by physical means is needed. Uterine contractions could not by themselves be expected to achieve sterility. Directional flow of secretions would have to be active at the microscopic level on both the endometrial surface and within the uterine glands. It was the occasional observation of ciliated cells in clinical uterine cytologies that led to a hypothesis that the equine uterus has a mucociliary escalator, like the respiratory tract. An important difference between the two systems is that in the airways, mucus interfaces with air, whereas uterine mucus interfaces with a fluid environment. The microscopic mucous clearance mechanism would assist, and be assisted by, the macroscopic physical clearance achieved by uterine contractions.

3. Mucociliary clearance

Ciliary movement was first recognized almost two centuries ago [24]. Single cells, such as protozoa and spermatozoa, were seen to be motile, propelled by cilia and/or flagella. In addition, multicellular organisms possess a variety of ciliated epithelia propelling blankets of mucus. The respiratory tract of mammals, and the gills of bivalves, provide useful bench-marks to investigate mucociliary clearance in the mare.

3.1. The respiratory tract

Mucociliary clearance is essential for respiratory health. The flow of mucus not only removes potential pathogens, but also clears inflammatory debris from the airways. Mucus-secreting and ciliated cells support the mucus blanket which is propelled by coordinated ciliary beating. In the trachea and bronchi, mucus is carried up into the pharynx to be swallowed. Similarly, in the nose, sinuses and eustachian tubes, mucus is carried retrograde, towards the oropharynx for swallowing [25,26]. Propulsion of mucus is assisted by mechanisms such as coughing [27], to expel airway blockages, and chloride pumps to osmotically hydrate mucosal secretions [28].

3.2. Invertebrates

It is interesting that the overall pattern of mucociliary transport in bivalves is similar to that in mammals—a constant sweeping of material towards the digestive tract. In bivalves, water-borne food particles, such as phytoplankta, are swept over gill lamellae towards the gut [29]. Cilia beating in one direction can nevertheless create bi-directional flow, with the blanket at the cilia extremities carried in the opposite direction to fluid at the base of the cilia [30]. This provides a means by which non-desirable particles may be sorted and expelled, while desirable particles are ingested. To achieve effective currents, cilia will beat in different directions depending on their location, such as on an exposed ridge, the face of a gill lamellae, or in the trough created by two adjacent lamellae.

3.3. The properties of mucus

Mucus is a heavily glycosylated polypeptide, whose production is coded for by MUC genes [31]. The attached polysaccharides trap water to varying degrees, thus controlling mucus' rheological (flow) properties. Because mucus is water soluble, it will eventually disperse in free fluid. Mucus also contains a variety of bacteriostatic components, such as lysozyme to break down cell walls, and lactoferrin to limit iron availability to bacteria [13]. Furthermore, mucus coats receptors to which some opportunistic pathogens may bind, and helps maintain a well hydrated, intact mucous membrane. The immunoglobulin most prevalent in mucus, secretory IgA, when bound to its secretory component, does not fix complement [32]. Thus it does not promote phagocytosis or lysis of invading bacteria. Instead, it appears to prevent bacterial adhesion by binding to the bacterial cell surface, trapping pathogens within the mucus blanket. Mucus also contains various surfactants, which not only preserve the rheological properties of mucus, but also decrease inflammation, and mitigate oxygen free-radical damage [33].

4. Evidence for mucociliary transport in the mare's genital tract

To test the mucociliary hypothesis, the equine genital tract would first need to be searched for the microscopic components of a mucociliary escalator, namely alternating mucus-secreting and ciliated cells beneath a mucus blanket. Similarly, the mare's gross anatomy would need to be reviewed for possible adaptations to promote mucociliary flow. When reviewed in such a way, the anatomy of the mare appears potentially adapted for mucociliary clearance.

4.1. Mucus producing cells in the endometrium

Through endometrial biopsy it is possible to examine the mare's endometrium at various stages of the estrous cycle. Several stains specific for mucopolysaccharides may be used, including mucicarmine, periodic acid schiff (PAS), and alcian blue. (The common histopathologic stain of haematoxylin and eosin does not stain mucopolysaccharides.) In the author's experience, alcian blue at pH 2.5 specifically stains cells of the luminal epithelium, whereas PAS also stains cells in the stratum spongiosum, revealing glandular carbohydrate. In bouin's-fixed tissue, a mucus blanket appears preserved over the mucosal surface, appearing as an undulating ribbon in a tissue section. In formalin-fixed tissue the mucus ribbon is not so well preserved. The composition of the ribbon appears to change with the cycle. The mucus blanket takes up alcian blue more intensely during diestrus than estrus, possibly reflecting different degrees of mucus hydration [12].

Speculation on the relationship between the luminal and glandular epithelia is interesting, but data are lacking. Ciliated cells are plentiful within glands, especially in the proximal third close to the neck, and would appear to be well positioned for expulsion of glandular contents into the lumen. Although the glandular and luminal epithelia of the equine uterus share a common lineage [34], understanding how both epithelia turn-over, and if their turn-over is linked, may help our understanding of subfertility in the future [35]. Gene expression and hyperplasia in mucus-secreting cells is partly controlled by mediators of inflammation [31]. Understanding how these various processes unfold is necessary to understand disease processes in uterine glands.

4.2. Uterine folds

In the author's opinion, one of the most striking features of the equine uterus, which may represent adaptation to mucociliary clearance, are endometrial folds. Here, thoughts of mucociliary action in the equine uterus invite comparison with invertebrate systems, where mucus is propelled in a fluid environment, through channels, and/or over ridges [24]. The folds of the equine uterus are well-known to equine theriogenologists as radiating features seen on ultrasound. In estrus they generate the characteristic ''wagon wheel'', ''pizza pie'' or ''cut grapefruit'' effect of the uterine horn in cross-section. In gross specimens, the folds are seen to be oriented longitudinally, and continuous with the folds of the cervix [36]. In speculating on the arrangement of uterine folds, one might suspect that they may be adapted to sweep material from the top of the fold into the trough [24]. Once in the trough, material may be speedily transported retrograde towards the cervix. However, experimental evidence in support is not available; such currents have not yet been demonstrated in the equine uterus. If mucociliary clearance does play a role in uterine defense, it would probably require some form of gross adaptations to be effective in cleansing the entire endometrium.

4.3. The cervix

The cervix is literally the bottle-neck of uterine physical clearance. Blockage of the cervix, or failure of the cervix to dilate, predisposes the mare to uterine fluid accumulation and uterine infection. What hypothetical role would the cervix play in mucociliary clearance? The answer is, obviously, a critical one. Mucociliary currents in the uterus would be useless if currents stopped at the internal os. Instead, trans-cervical currents must be rapid to prevent accumulations of materials arriving from far-distant regions of the endometrial surface. One indication that the cervix is adapted to mucociliary clearance is that uterine folds are continuous with cervical folds. Thus particles may exit the uterus unimpeded. A second is that ciliary density in available equine cervical descriptions is high [36], possibly reflecting increased mucociliary flow through cervical channels. The third is that the edema of the cervix in estrus may increase mucociliary flow by providing a greater surface area available to mucociliary currents, and a final downward trajectory over the pendulous external os. It seems likely, therefore, that the importance of the cervix to uterine defense must extend to mucociliary clearance, if such clearance exists.

4.4. Evidence of possible mucociliary action

While the above anatomical descriptions are helpful, they nevertheless provide no physiological evidence of mucociliary clearance. To be proven, the mucociliary hypothesis requires demonstration of movement at the microscopic level, and such movement must be distinguished from the well-documented macroscopic movement of uterine contractions. It is a well-known property of ciliated cells that they can maintain their motility when removed from the host. Therefore, one would predict that rafts of endometrial cells collected during uterine lavage would contain motile cells. This indeed is the case, as may occasionally be seen when searching for equine embryos. Motile rafts of cells may sometimes be seen tumbling or rotating in flush solution, while searching for an embryo.

To specifically search for motile endometrial cells, one may use an embryo transfer filter to concentrate cells from a large volume flush, allow cells to fall by gravity to the bottom of 50-mL centrifuge tubes, and then pipette samples into wells of a 96-well plate and examine using inverted microscopy. Not all wells have motile cells, but motility is usually visible in some wells as vibration or movement of cell clumps. More careful examination under higher magnification (400$\times$) reveals cilia beating at a rate or about 13 beats/s. In the author's limited experience, modified dulbecco's PBS (including pyruvate, dextrose, calcium and magnesium), with added 5% equine serum, such as from the horse to be flushed, can be used for lavage and examination of cells. The author's use of autologous serum is arbitrary; further study would probably reveal a preferable additive. The author has not been successful in demonstrating ciliary movement in cells collected using sterile isotonic saline. In the PBS-type media described, cilia may be seen to beat for as long as 96 h, and it is possible to compare rates of beating between different wells under different treatments using video microscopy. One limitation of the technique is that most video cameras capture images at a rate of 30 frames/s, making it difficult to detect movement faster than 13 beats/s. However, small studies using this system have been useful in identifying potential inhibitors of ciliary movement (e.g. unbuffered gentamicin) [37].

Rarely, an intact membrane containing coordinated ranks of motile cells can be encountered, and visible markers, such as carbon powder, can be applied to reveal ciliary currents. Using this technique, the author has observed particle velocity of about 50 μm/s, within the expected rate of ciliary movement for other epithelia [25]. Assuming that the equine cervix is approximately 10 cm long, such a current would require approximately 30 min to traverse the cervix, and perhaps about 3–4 h to carry a particle from the tip of a uterine horn to the external os. The informal nature of these estimates should be emphasized, but they indicate a hypothetical clearance rate that a mucociliary escalator might be predicted to achieve in the equine uterus. This rate of clearance also has implications for potential disruption of mucus transport.

5. Disruption of mucociliary clearance

Efficient mucociliary transport requires mucus of appropriate composition for optimal flow, adequate numbers of functioning ciliated cells for propulsion, and appropriate membrane "topography". Disruptions of mucociliary clearance usually involve disruption of these requirements, either singly or in combination. These disruptions have been extensively explored in the respiratory tract, and experimentally in invertebrate systems. Understanding how mucociliary transport is altered in such systems allows us to speculate on potential causes and effects of ciliary disruption in the uterus of the mare.

5.1. The respiratory tract

The mammalian respiratory tract includes a variety of mucosal surfaces such as the nasopharynx, sinuses, eustachian tube, middle ear, and trachea, all displaying flow of material towards the esophagus. Disruption of such mucus flow is implicated in the pathogenesis of conditions such as sinusitis, otitis media, and pneumonia.

Altered mucus composition may be a primary or secondary cause of disruption of mucociliary transport. As a primary cause, hereditary impairment in chloride transport across mucous membranes prevents adequate hydration of mucus, leading to accumulations of the highly viscous secretions seen with cystic fibrosis. In addition to reducing air flow, such secretions fail to adequately clear pathogens. Cystic fibrosis patients are particularly susceptible to infections with adherent bacteria, such as *Pseudomonas aeruginosa* [28]. As a secondary cause, mucus composition may be altered during infection, leading to other post-infection sequelae. For example, following respiratory infections, accumulations of neutrophils and cellular debris increase the viscosity of mucus, and may prevent drainage of sinuses and/or the middle ear, leading to opportunistic infection at these sites [25]. Alternatively, microorganisms which cause overly abundant, serous secretions, may promote spread of pathogens throughout the respiratory tract, by flow in the opposite direction of mucociliary currents [38]. In addition, such secretions may be more easily aerosolized by sneezing, increasing likelihood of transmission.

Primary disruption of ciliary function is also well documented. For example, in primary ciliary dyskinesia, coordination of ciliary beating, and hence mucus flow, is impaired [39]. Similarly, cilia associated viruses may target ciliated cells directly, and influenza viruses are well-known for their destruction of ciliated respiratory epithelia [40]. Inhaled toxins from cigarette smoking, pollution, or poor ventilation, may all depress ciliary function, with subsequent depression of respiratory clearance [41]. Ciliary activity is also subject to pharmacological inhibition by certain antibiotics [42].

The topography of mucous membranes is essential for their adequate functioning as mucus transport systems. Essential features would be integrity of the epithelium over long distances, open drainage, and physical protection of the mucus blanket. In the nasal cavity, epithelial scarring may interrupt mucus flow. Inspissated mucus secretions may cause blockage of paranasal sinuses, or the eustachian tube. Endoscopic surgery to improve drainage in patients with chronic rhinosinusitis improves subsequent mucociliary clearance of sinus epithelia [25]. Importantly, if fluid accumulates over a ciliated epithelium, mucociliary currents will be rendered ineffective [24]. Also, because mucus is water soluble, the mucus blanket and the pathogens it contains may be dispersed throughout accumulated fluid.

5.2. Invertebrates

In invertebrates, disruption of ciliary activity has been experimentally studied to determine factors necessary for ciliary movement. Acidic pH appears to be the most potent physiological inhibitor of ciliary activity [24]. Similarly, elevated temperature, within physiological ranges, increases ciliary flow. Ciliary activity is often depressed by inhibitors of aerobic metabolic activity in many (but not all) systems. However, the intricate arrangement of folds and channels in invertebrates suggest that disruption of anatomy could gravely alter mucociliary function [29]. In ciliated tubes, transport in the center appears significantly reduced when the radius exceeds five times the length of the cilium [24].

6. Possible mucociliary disruption in the mare

At the time of writing, mucociliary function and dysfunction in the mare's genital tract are not well studied. However, considerations of mucociliary clearance might shed light on some conditions of the equine uterus.

6.1. Intrauterine fluid

In the mare, a tendency towards accumulation of fluid in the uterus has been linked to susceptibility to uterine infection through multiple studies. Susceptible mares show cyclic fluid accumulations, expel uterine fluid more slowly than resistant mares, and display improved fertility when fluid is removed [6,9,43]. Accumulations of uterine fluid clearly disrupt phagocytic processes by degradation of complement [11], but could also potentially disrupt mucociliary clearance in various ways. Firstly, mucociliary currents are most efficient through narrow channels, such as would be found when opposite faces of the endometrium are in contact. Based on the estimate that cilia would not be effective in moving fluid whose depth exceeded five ciliary lengths (approximately 50–100 μm in the mare) [24], the center of visible fluid accumulations on ultrasound would represent material beyond the immediate reach of ciliary motion. Whether the borders of such accumulations can be effectively "drained" by cilia is an open question. Accumulations of fluid of a few millimetres are normal in estrus, and may not represent overall break down of mucociliary clearance. Extensive accumulations, especially those which dilate the uterine lumen, might represent overall break down of mucociliary processes.

Fluid accumulations might also disrupt the composition of mucus. The ability of mucus to trap particles, and carry them long distances, requires integrity of the mucous continuum. The combined properties of viscosity and elasticity help hold mucus together, and allow it to transmit force over long distances. Excessive hydration of mucus will eventually cause its dispersal into solution. Thus protecting mucus from long-term exposure to large volumes of static free fluid seems essential for its mechanical function. The tendency for some uterine pathogens, such as *S. zooepidemicus*, to cause copious secretions [44,8] might disrupt mucociliary clearance by similar dispersal of mucus. Alternatively, tenacious secretions of capsule, perhaps by such organisms as *Klebsiella pneumoniae* [45], might cause excessive viscosity, inhibiting the movement of mucus.

6.2. Loss of intrauterine folds

As mentioned earlier, uterine folds would probably be an essential component of any uterine mucociliary clearance mechanisms in the mare. That mares susceptible to chronic uterine infections are reported to have reduced numbers of uterine folds would appear to support this idea, as would the increased susceptibility to uterine infection of progesterone-dominated mares, which also display reduction of uterine folds [46,47]. Mucociliary currents would be most efficient in the capillary spaces between folds, and would be disrupted when folds are separated by uterine fluid, or reduced in size by scarring, pathology, or endocrine disruption. Intrauterine particles have a tendency to be carried, and even trapped, between the folds of the uterus. By projecting into the uterine lumen, folds reduce the potential space for free uterine fluid. In contrast, when folds are reduced, free fluid will accumulate in the center of the uterine lumen. Alternatively, fluid distension of a uterine horn might obliterate the associated uterine folds.

6.3. Cervical trauma

The cervix is well-known to be an essential component of uterine defenses, serving as both a barrier to ascending infection, and a conduit for the expulsion of contaminated uterine fluid. Cervical adhesions, or failure of the cervix to adequately dilate, may lead to delays in uterine clearance and even pyometra [48]. Continuity of the cervical folds with uterine folds suggests that damage of cervical folds by scarring, adhesions, or hormonal imbalance may lead to impaired mucociliary clearance through the cervix. Indeed, as the final exit point of proposed mucociliary currents, cervical dysfunction would be especially deleterious. A large cervix, with generous folds in estrus, would theoretically be ideal for optimal clearance, since the cross-sectional area of available capillary spaces between the folds would be maximized. In contrast, a small cervix, with reduced folds, might have comparatively less mucociliary flow, a condition that might be expected to occur, for example, in aged maiden mares.

6.4. Loss of cilia

Scanning electron microscopy of the endometrium reveals various conditions which cause loss of cilia, and may indicate potential disruption of the mucociliary apparatus. For example, infusion of irritating substances can cause significant uterine scarring and damage to the uterine epithelium [49–51]. Electron microscopy of endometrial biopsies of mares treated with unbuffered gentamicin has shown loss of cilia, although the author is not aware of a corroborating study in another species [52]. Not only could this cause failure of clearance, but denuded epithelium can serve as a nidus for infection. Similar ultra-structural pathology, including cilia loss and epithelial defects, can be seen in mares susceptible to endometritis [53,54]. Studies in the post-partum mare show return to an intact epithelium of mucus producing and ciliated cells as part of normal uterine involution [55]. It would seem likely that the endometrium could suffer severe damage following metritis, retained placenta, or dystocia, all of which could potentially delay or prevent return of normal mucociliary clearance, as occurs in patients suffering damage to respiratory epithelia [25].

6.5. Mucus overproduction—a vicious circle?

Inflammation causes increased endometrial section of mucus, a response typical of mucous membranes [56]. Not only does the mucus trap and hopefully coat the source of inflammation, it also protects denuded areas of the epithelium. However, if mucus accumulates excessively, the inflammatory debris it contains may itself act as a source of inflammation, and in-turn cause more mucus production. Such a vicious circle may occur in many cases of chronic uterine infections. In such situations, the rheological properties of mucus may start to act against it, potentially yielding secretions that are excessively watery (seen as too much uterine fluid) or excessively viscous (inspissated secretions which cannot be voided). Different virulence mechanisms of bacteria, and different responses to infection, could potentially push the process in either direction leading to differences in clinical signs, and perhaps different approaches to appropriate therapy.

7. Clinical relevance

Discussion of the clinical relevance of mucociliary clearance in the mare is dangerous due to a lack of information. No management recommendations can be made based on available information. However, it is perhaps worth reviewing the potential impact of current therapies on mucociliary clearance, and how improved knowledge of mucociliary clearance could potentially lead to improved prevention of uterine infections.

7.1. Therapy

Current therapies for uterine infections would appear to potentially support mucociliary clearance mechanisms. Therapies that remove accumulations of intrauterine fluid might restore the uterine topography necessary for mucociliary clearance. Oxytocin's induction of uterine contractions would help reduce spaces between the uterine folds. Alternatively, uterine lavage, by removing inflammatory debris and inspissated secretions, might be useful in restoring normal mucus flow after periods of accumulation of viscid mucus. The impact of antibiotics on mucociliary function is difficult to predict, but available evidence suggests that intrauterine antiseptics, and certain antibiotics may prove harmful to mucociliary clearance [42,52]. Interestingly, the use of dilute solutions of vinegar, commonly used to treat uterine fungal infections, might be expected to reduce ciliary activity, due to their lowering of pH. It may eventually prove necessary to balance mucociliary disruption with microbicidal efficacy for some intrauterine treatments. All of the foregoing is purely speculative, but it nevertheless illustrates how considerations of mucociliary clearance could broadly impact therapy for uterine infections.

7.2. Prevention

Better understanding of uterine pathology might help us to identify those risk factors which render mares susceptible to uterine infections. Accurately identifying mares with mucociliary pathology in a clinical setting might allow us to retrospectively determine what leads to such dysfunction, and thereby prevent it, or be better prepared to manage it at breeding. This might be accomplished through endometrial biopsy, perhaps making greater use of electron microscopy. However, to be validated, such approaches would require a parallel clinical assessment of mucociliary function. While mucus transport rate may be measured in other systems, such as the frog soft palate [24], such a technique has yet to be developed for clinical use in the mare. The cause and effect link between pathology and mucociliary dysfunction still needs to be established, and much work will be required to do so.

7.3. Mucus and mucosal immunity

Attempts to boost uterine mucosal immunity might prove to be useful adjuncts to mucociliary function. Secretory IgA, produced by plasma cells lining the endometrium, may bind uterine pathogens, prevent their attachment to the epithelium, and trap them in the mucus blanket [57]. Secretory IgA does not require complement, so it remains effective in the uterine lumen [32]. The mucosal immune response is of particular value in the uterus because it tends to be non-bactericidal, relying instead on mechanisms of physical clearance. Such a response may help provide a non-toxic environment for the early equine embryo. In contrast, bactericidal mechanisms, for example phagocytosis and complement mediated cell lysis, probably present a toxic environment to the embryo in mares which fail to clear uterine inflammation, such as those with delayed uterine clearance. Available data suggests that mucosal vaccination of the uterus may be helpful in removing intrauterine streptococci, but clinical efficacy in large studies has yet to be attempted [58].

8. Conclusion—the mystery of the resistant mare

How the healthy mare can clear bacteria, semen, and inflammatory debris of breeding so rapidly, and then "culture" an embryo a short time later, remains a mystery to this author. Mucociliary activity may, or may not, play a central role. Nevertheless, the process underlying this feat is worth understanding for several reasons. First, it provides a useful model for the prevention of opportunistic infections. By unravelling the process in the mare, we may shed additional light on mucosal infections in general. In addition, it may lead to a better understanding of bacterial pathogenesis, as the differences between genital commensals and uterine pathogens become clearer. And, perhaps most importantly, it may help extend the productive life of our brood mares.

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