Available online at www.sciencedirect.com

ScienceDirect

journal homepage: http://www.elsevier.com/locate/repbio

Review

Mechanisms linking bacterial infections of the bovine endometrium to disease and infertility☆

Luísa Cunha Carneiro[a,b], James Graham Cronin[a], Iain Martin Sheldon[a,*]

[a] Institute of Life Science, Swansea University Medical School, Singleton Park, Swansea SA2 8PP, United Kingdom
[b] Faculty of Agricultural and Veterinary Science, Universidade Estadual Paulista, Jaboticabal, Brazil

ARTICLE INFO

Article history:
Received 14 July 2015
Received in revised form
24 November 2015
Accepted 5 December 2015
Available online 23 December 2015

Keywords:
Cow
Endometritis
Cytolysis
Innate immunity
Uterus

ABSTRACT

Bacterial infections of the endometrium after parturition commonly cause metritis and endometritis in dairy cattle, and these diseases are important because they compromise animal welfare and incur economic costs, as well as delaying or preventing conception. Here we highlight that uterine infections cause infertility, discuss which bacteria cause uterine disease, and review the evidence for mechanisms of inflammation and tissue damage in the endometrium. Bacteria cultured from the uterus of diseased animals include *Escherichia coli*, *Trueperella pyogenes*, and several anaerobic species, but their causative role in disease is challenged by the discovery of many other bacteria in the uterine disease microbiome. Irrespective of the species of bacteria, endometrial cell inflammatory responses to infection initially depend on innate immunity, with Toll-like receptors binding pathogen-associated molecular patterns, such as lipopolysaccharide and bacterial lipopeptides. In addition to tissue damage associated with parturition and inflammation, endometrial cell death is caused by a cholesterol-dependent cytolysin secreted by *T. pyogenes*, called pyolysin, which forms pores in plasma membranes of endometrial cells. However, endometrial cells surprisingly do not sense damage-associated molecular patterns, but a combination of infections followed by cell damage leads to release of the intracellular cytokine interleukin (IL)-1 alpha from endometrial cells, which then acts to scale inflammatory responses. To develop strategies to limit the impact of uterine disease on fertility, future work should focus on determining which bacteria and virulence factors cause endometritis, and understanding how the host response to infection is regulated in the endometrium.

☆ This paper has been given as a lecture at the International Conference on Biology and Pathology of Reproduction in Domestic Animals, "Endometritis as a cause of infertility in domestic animals", held on September 28th–30th, 2015 Gdansk, Poland.

* *Corresponding author*. Tel.: +44 1792 602709.
E-mail address: i.m.sheldon@swansea.ac.uk (I.M. Sheldon).

http://dx.doi.org/10.1016/j.repbio.2015.12.002

1. Introduction

Bacterial infections of the uterus cause disease and infertility in dairy cattle, particularly after parturition, when they lead to metritis and endometritis in up to 40% of animals [1]. These postpartum uterine diseases are important because they compromise animal welfare, and incur costs for treatment, reduced milk production, and replacement of infertile animals; €1.4 billion every year in the EU alone [2]. Here we address the question of how infections of the endometrium are linked to disease and infertility. We highlight the impact of uterine disease on fertility, then discuss which bacteria cause disease, and finally assimilate recent evidence about the mechanisms of inflammation and tissue damage in the endometrium.

2. Bacterial infections of the uterus cause infertility

In a meta-analysis of more than 10,000 animals, postpartum metritis increased the time to first insemination by 7.2 days, reduced conception rate to first insemination by 20%, and increased the calving to conception interval by 18.6 days [3]. Similarly, clinical endometritis increased the interval to first insemination by 11 days, and delayed conception by 32 days, compared with animals that did not have endometritis [4]. Cows with clinical endometritis between 20 and 33 days post partum were also 1.7 times more likely to be culled for reproductive failure than cows without endometritis [5]. Furthermore, infertility could be due to the endometrial inflammation in postpartum cattle infected with *Trueperella pyogenes* [6]. Together, these observations linking bacterial infections of the uterus with infertility provide an impetus to discover the underlying mechanisms. Evidence for how uterine disease impacts ovarian and neuroendocrine function have been reviewed recently [7]. So, the objective of the present review is to focus on the uterus.

3. Microbial infections of the postpartum uterus

From a historical perspective, microbial disease of the uterus of cattle merited little comment 80 years ago, and endometritis was not considered a common problem. However, between 1960 and 2000, endometritis in cattle started to be the subject of investigations to understand the pathogenesis of the disease and to select the most effective treatments. In one study, 93% of the uteri obtained within 15 days of calving yielded bacteria on aerobic and anaerobic culture of endometrial swabs and tissue [8]. The proportion of uteri from which bacteria were isolated had declined to 78% by 16–30 days, 50% by 31–45 days, and 9% by 46–60 days postpartum. Similar proportions of animals yielded culturable bacteria in subsequent studies [9,10]. However, the situation is more complicated because the bacterial flora fluctuates during the first 7 weeks postpartum due to spontaneous contamination, clearance and recontamination [11]. Furthermore, which bacterial isolates are contaminants of the uterus and which are pathogens is open to debate. Uterine infection was most commonly associated with the presence of *Escherichia coli*, *T. pyogenes*, *Fusobacterium necrophorum*, and *Prevotella* or *Bacteroides* species in studies spanning from the 1960s to the 1990s [8,10–12]. In the last 15 years, studies using aerobic and anaerobic culture methods provided similar evidence to the earlier investigations [9,13–15]. These bacteria were identified by standard culture techniques and are classified into pathogens, potential pathogens and opportunist contaminants (Table 1). In particular, *T. pyogenes* is linked to the severity of endometrial pathology and clinical disease [10,14,16]. Furthermore, *T. pyogenes*, *F. necrophorum* and *Prevotella* species can act synergistically to increase the likelihood of endometritis and the severity of disease [17,18]. Associations between uterine disease and bacteria that are not readily cultured by standard techniques emerged recently as researchers started to use biochemical, molecular and sequencing techniques [19–23]. These studies have provided

Table 1 – Categorization of bacteria, isolated by aerobic and anaerobic culture of uterine swabs, based on their potential pathogenicity [8–14,17,18]. Categories: (1) pathogens known to cause endometrial lesions; (2) potential uterine pathogens; and (3) bacteria not recognized as uterine pathogens that are likely contaminants of the uterine lumen.

Pathogens	Potential pathogens	Contaminants
Escherichia coli	*Acinetobacter* spp.	*Aerococcus viridans*
Trueperella pyogenes	*Bacillus licheniformis*	*Clostridium butyricum*
Prevotella spp.	*Enterococcus faecalis*	*Clostridium perfringens*
	Haemophilus somnus	
Fusobacterium necrophorum	*Mannhiemia haemolytica*	*Corynebacterium* spp.
Fusobacterium nucleatum	*Pasteurella multocida*	*Enterobacter aerogenes*
	Peptostreptococcus spp.	*Klebsiella pneumoniae*
	Staphylococcus aureus (coagulase +)	*Micrococcus* spp.
	Streptococcus uberis	*Providencia rettgeri*
	Bacteroidetes species	*Providencia stuartii*
	Firmicutes species	*Proteus* spp.
	Fusobacteria species	*Proprionobacterium granulosa*
		Staphylococcus species
		α-haemolyic Stretococci
		Streptococcus acidominimus