Watery Mouth Disease in Neonatal Lambs: a Systematic Literature Review.

Abstract

Watery mouth disease is a considered to be a significant cause of neonatal mortality in lambs. The clinical signs are strongly associated with an endotoxaemia produced as a result of the lysis of gram-negative bacteria. It has been associated with mass antibiosis to neonatal lambs at birth, a practice which is now untenable. It can be prevented in many cases through the timely administration of good quality colostrum and a hygienic birth environment. However, despite this, much remains unknown about the specific aetiopathogenesis. Alternative strategies for prevention, treatment and control are required, particularly when colostrum quality is poor, or delivery is absent, and where unhygienic conditions predominate.

Introduction

The mortality and morbidity of neonatal lambs is a cause of poor welfare for affected cases and results in the reduced production and economic profitability of farms. Worldwide, on average, 15% of lambs die in the neonatal period with this rate remaining unchanged for the past 40 years (Dwyer and others 2016). Following birth, the neonate enters a new and hostile environment compared to that *in utero* and in particular must meet the challenge presented by infectious organisms which very rapidly become important causes of mortality for neonatal lambs. In recent surveys, infectious disease accounted for 10% of neonatal deaths in Wales, UK (HCC 2011) and 36% in Norway (Holmoy and others 2016), with both surveys reporting the majority of deaths within 48

hours of birth. Arguably, the most important infectious disease syndrome reported in this very early period is an endotoxaemia associated with undefined pathotypes of *Escherichia coli,* colloquially known as watery mouth disease (WM) on account of one of the clinical signs seen – drooling saliva from the mouth.

Although WM has been recognised since at least the 1920s (Eales 1987) surveys have suggested in intensively farmed sheep it can account for of up to a quarter of all neonatal deaths (King and Hodgson 1991; Scott 2007). Together with this, WM is cited as the biggest reason for the widespread prophylactic and metaphylactic use of antibiotics, in large numbers of flocks in some regions (Davies and others 2017; King and Hodgson 1991). This practice is clearly untenable, in particular in light of the integrated global challenges of improving food security, maintaining and improving animal welfare and reducing the use of antimicrobials in farmed species (O'Neill 2016; WHO 2015). Watery mouth disease has for too long been neglected from a research perspective, and alternatives to mass antibiosis are urgently required.

The aim of this review is to summarise the current knowledge of WM in lambs and clarify the evidence base for each aspect considered.

Materials and Methods

Due to the small numbers of research papers published on WM, a broad systematic approach was used to try and identify as many peripheral studies, letters and reports as possible (**Figure *1***). Three English language databases were utilised: PubMed, Web of Science and Scopus over the time frame 1980-2019 inclusive. These databases were searched using predefined search terms (**Table *1***). The citations were primarily

screened for eligibility by title and abstract and the full text used for any ambiguous citations.

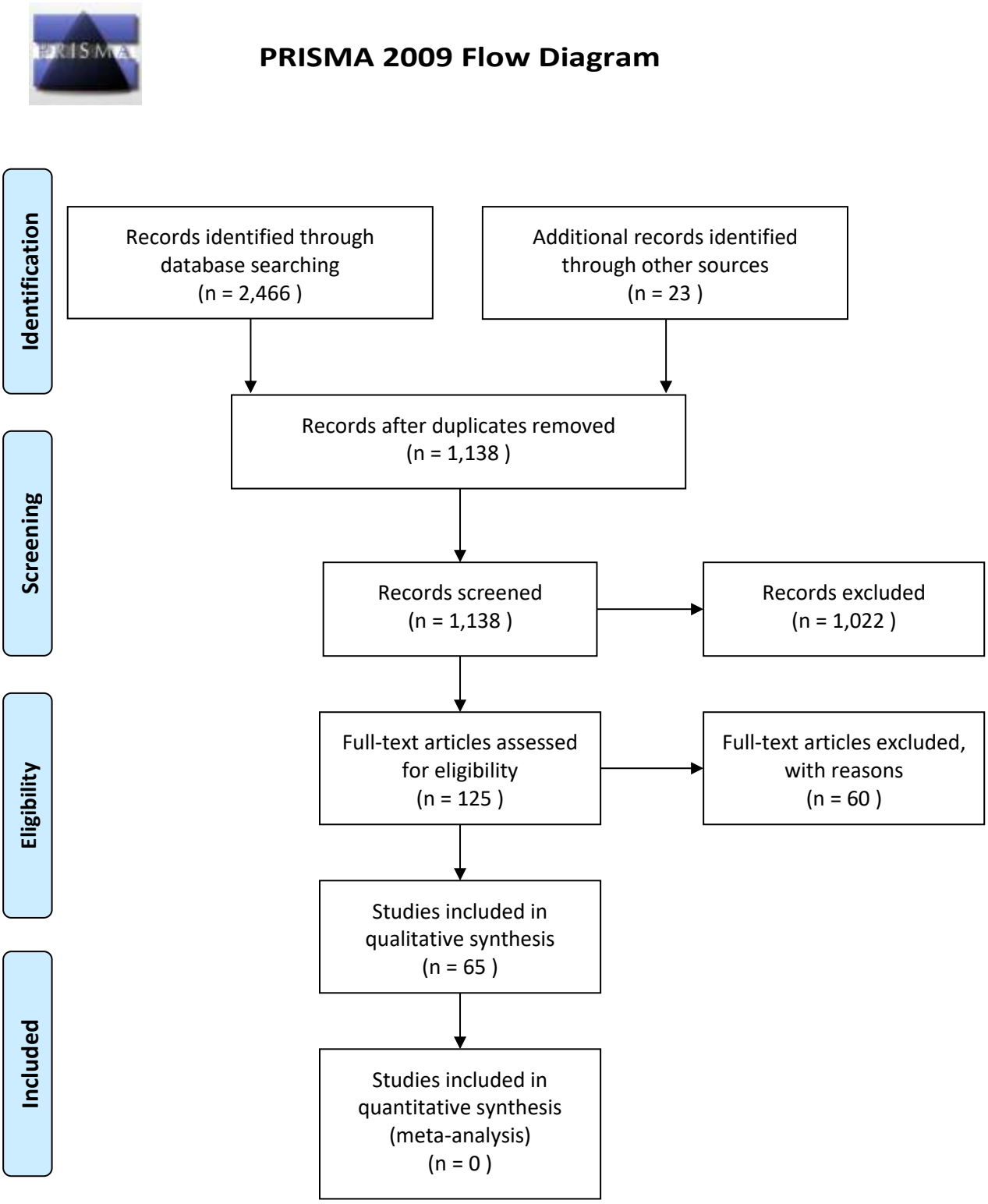

Figure 1: Prisma flow diagram indicating the number of citations at each stage of the systematic review process. From: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(7): e1000097. doi:10.1371/journal.pmed1000097. For more information, visit www.prisma-statement.org.

Database and search terms used	N citations
PubMed – 29/11/2019	
"watery mouth"[Title/Abstract]	14
(endotoxic[Title/Abstract] AND lambs[Title/Abstract]	2
(endotoxaemia[Title/Abstract]) AND lambs[Title/Abstract]	5
(endotoxin[Title/Abstract]) AND lambs[Title/Abstract]	66
(((Escherichia coli[Title/Abstract]) OR E. coli[Title/Abstract])) AND Lambs[Title/Abstract]	201
Web of Science - 29/11/2019	
(**TS**="watery mouth") *AND* **LANGUAGE:** (English))	21
(TS=(endotoxic AND lambs) *AND* **LANGUAGE:** (English))	6
(**TS=**(endotoxaemia AND lambs) *AND* **LANGUAGE:** (English))	8
(**TS**=(endotoxin AND lambs) *AND* **LANGUAGE:** (English))	209
(**TS**=((Escherichia coli OR E. coli) AND lambs)) *AND* **LANGUAGE:** (English))	839
Scopus – 29/11/2019	
TITLE-ABS-KEY ("watery mouth")	20
TITLE-ABS-KEY (endotoxic AND lambs)	4
TITLE-ABS-KEY (endotoxaemia AND lambs)	6
TITLE-ABS-KEY (endotoxin AND lambs)	95
TITLE-ABS-KEY ((Escherichia coli OR E. coli) AND lambs)	970

Table 1: Databases and search terms used in the systematic review process, together with the number of citations identified from each individual search.

Citations were excluded for the following reasons:

- If duplicated.
- If not written in English and a suitable translation was unavailable.
- If considered irrelevant in terms of the disease process or body system considered e.g. excluded if the study considered solely the respiratory system.
- If solely considered pre-term lambs or lambs older than 7 days.
- If primarily investigating the LamB protein in *E. coli.*

Following the initial screening process a further 60 full-text articles were examined and removed. Throughout this process a small number of additional articles (n=23) not

identified through the original database searches were found by searching the reference lists of each citation and included if considered relevant.

Results

Clinical Features

Twenty four citations were obtained that described clinical features of WM (Table 2) and of these 15 specified a case definition. Increased salivation/mucus around the mouth was the only feature described by all 24 publications, 17 described cases as having reduced appetite or ceasing to suck, 17 described cases as having a distended abdomen/abdominal tympany and 15 described cases as dull or depressed. Other features were less frequently reported although in some citations were considered as key features of the disease e.g. retained meconium or reduced gut motility (Mitchell and Linklater 1983).

In summary, the disease usually manifests in lambs <72 hours of age usually presenting as dull/depressed with reduced sucking/inappetence followed by the development of increased salivation/mucus around the mouth and hence the name 'watery mouth' (**Figure *2***, a) and b)). This feature has also led to the synonymous colloquialisms: slavers/slavery mouth (King 1986; King and Hodgson 1991); two French reviews use the colloquialisms 'agneaux baveurs' (slobbery lambs) and 'bouche baveuse' (slobbery mouth) (Millemann and others 2003; Poncelet 2004). Abdominal tympany appears later on in the disease process (**Figure *2***, image c)) and several authors describe that gentle shaking of these lambs elicits a splashing sound hence a further colloquialisation 'rattle belly' (Eales 1987; Haig 1981; King 1986). Scouring is unusual and often constipation or

retained meconium may be present (Eales and others 1986). Untreated cases die within six to 24 hours showing a terminal hypoglycaemia, hypothermia and lacticacidaemia (Collins and others 1985; Hodgson and others 1989b).

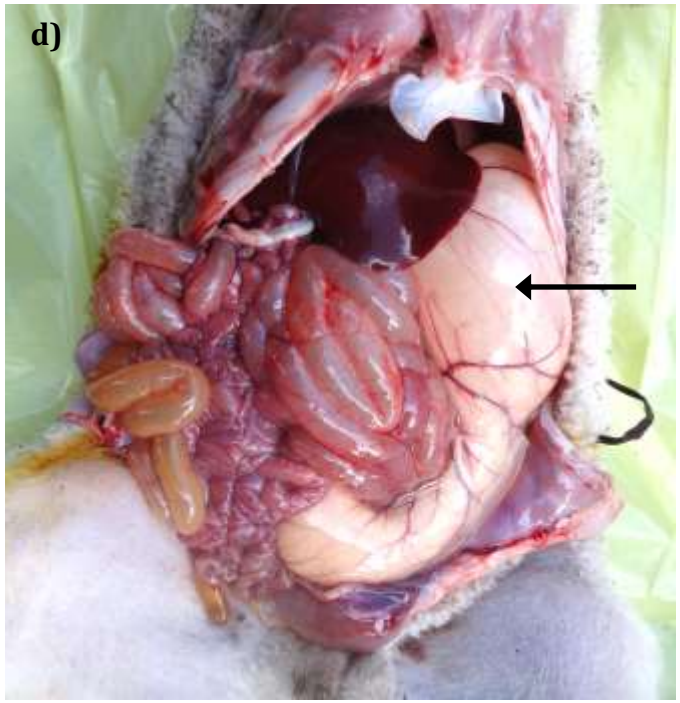

Figure 2: Clinical signs associated with watery mouth disease; a) and b) two lambs with signs of increased salivation around the mouth; c) a lamb with right sided abdominal distension associated with abdominal tympany; d) post mortem dissection clearly showing the dilation of the abomasum.

Clinical feature recorded in cases	Citation	N citations	Used as part of a specific case definition	N citations
Increased salivation/mucus around mouth	Christodoulopoulos (2008); Christodoulopoulos and others (2013); Collins (1981); Collins and others (1985); Eales (1987); Eales and others (1986); Eales and others (1985); Findlay (1973); Haig (1981); Hodgson (1993); Hodgson and others (1995); Hodgson and others (1999); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Linklater (1989); Millemann and others (2003); Mitchell and Linklater (1983); Orr (1995); Poncelet (2004); Scott (1988); Scott and Gessert (1996); Vaid (1991)	24	Christodoulopoulos (2008); Christodoulopoulos and others (2013); Eales (1987); Eales and others (1986); Eales and others (1985); Findlay (1973); Hodgson and others (1995); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Mitchell and Linklater (1983); Scott (1988); Scott and Gessert (1996); Vaid (1991)	15
Stopped/reduced sucking/inappetence/anorexia	Eales (1987); Eales and others (1986); Eales and others (1985); Findlay (1973); Hodgson (1993); Hodgson and others (1995); Hodgson and others (1999); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Linklater (1989); Mitchell and Linklater (1983); Sargison and others (1997); Scott (1988); Scott and Gessert (1996); Vaid (1991)	17	Eales (1987); Eales and others (1986); Eales and others (1985); Findlay (1973); Hodgson and others (1995); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Mitchell and Linklater (1983); Scott (1988); Scott and Gessert (1996); Vaid (1991)	13
Distension of the abdomen or abdominal tympany	Christodoulopoulos and others (2013); Collins and others (1985); Eales (1987); Eales and others (1986); Findlay (1973); Haig (1981); Hodgson (1993); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Linklater (1989); Mitchell and Linklater (1983); Sargison and others (1997); Scott (1988); Scott and Gessert (1996); Vaid (1991)	17	Christodoulopoulos and others (2013); Findlay (1973); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); Mitchell and Linklater (1983); Scott (1988); Scott and Gessert (1996); Vaid (1991)	9
Dull/depressed	Eales (1987); Eales and others (1986); Eales and others (1985); Haig (1981); Hodgson (1993); Hodgson and others (1995); Hodgson and others (1999); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Linklater (1989); Mitchell and Linklater (1983); Sargison and others (1997); Vaid (1991)	15	Eales (1987); Eales and others (1986); Eales and others (1985); Goelst and others (1992); Hodgson and others (1995); Hodgson and others (1989a); Hodgson and others (1989b); King (1986); King and Hodgson (1991); Vaid (1991)	10
Usually in lambs <72 hours old	Christodoulopoulos (2008); Christodoulopoulos and others (2013); Collins and others (1985); Eales (1987); Haig (1981); Hodgson (1993); King and Hodgson (1991); Linklater (1989); Poncelet (2004)	9	Christodoulopoulos (2008); Christodoulopoulos and others (2013); Eales (1987); King and Hodgson (1991)	4
Retained meconium/constipation	Collins (1981); Findlay (1973); Gilmour and others (1985); Haig (1981); King and Hodgson (1991); Linklater (1989); Mitchell and Linklater (1983); Scott (1988); Winkler (1981)	9	Mitchell and Linklater (1983); Scott (1988)	2
Reduced gut motility/gut stasis	Eales and others (1985); Hodgson (1993); Hodgson and others (1989a); Hodgson and others (1989b); Linklater (1989); Mitchell and Linklater (1983); Vaid (1991)	7	Hodgson and others (1989a); Hodgson and others (1989b); Mitchell and Linklater (1983); Vaid (1991)	4
Unable/reluctant to stand	Collins and others (1985); Findlay (1973); Hodgson and others (1999); King and Hodgson (1991); Sargison and others (1997); Scott (1988)	6	Findlay (1973); Scott (1988)	2
Lethargy	Christodoulopoulos and others (2013); Collins and others (1985); Linklater (1989); Scott (1988); Scott and Gessert (1996)	5	Christodoulopoulos and others (2013); Scott (1988); Scott and Gessert (1996)	3
Mostly normothermic	Collins and others (1985); Eales (1987); Eales and others (1986); Linklater (1989); Sargison and others (1997)	5		
Mostly not scouring	Collins and others (1985); Eales and others (1986); Gilmour and others (1985); King and Hodgson (1991); Linklater (1989)	5		
Increased lacrimation	Hodgson (1993); Hodgson and others (1999); King and Hodgson (1991)	3		
Usually scouring	Hodgson and others (1999)	1		
Congested mucous membranes	Sargison and others (1997)	1		

Table 2: Specific **c**linical signs of clinical cases with watery mouth as described or reported by publications, together with those that described the clinical feature as part of a specific case definition.

Differential diagnoses

Many cachectic lambs will salivate in extremis (Linklater 1989) but usually less so than those with WM, so in the terminal stages of disease diagnosis may be more difficult. However, in earlier stages of the disease and by observing the progression of individual cases diagnosis presents few difficulties. Diagnosis may be clarified post mortem where abomasal distension with gas and saliva together with a possible retained meconium may be observed (Carson 2019).

Differential diagnoses include colibacillosis (neonatal lamb diarrhoea) and lamb dysentery (Mitchell and Linklater 1983; Sargison 2008b), 'salivary abomasum disease' (SAD) (Christodoulopoulos and others 2013) and 'lamb D-lactic acidosis syndrome' (LDLAS) (also known as 'drunken lamb syndrome' (DLS)) (Angell and others 2013). These diseases can usually be differentiated clinically from WM by careful observation of the progression of the disease and the clinical signs seen.

Lamb dysentery manifests as the sudden death of young lambs, usually up to 10 days of age, and more slowly developing cases may produce profuse volumes of blood stained diarrhoea, although death may occur too rapidly for this to be observed (Sargison 2008b). At post mortem these lambs classically have obvious haemorrhages in the small intestine (**Figure *3***) and epsilon toxin may be identified.

Lambs affected with SAD tend to be older than those with WM (mean age 7.23 days (SD 3.14)), and do not present with clinical or biochemical signs of endotoxic shock (Christodoulopoulos and others 2013). It has been suggested that SAD is

possibly equivalent to WM but presents differently due to the increased age of the lambs (Christodoulopoulos 2008). To the authors' knowledge there has only been one study of SAD to-date and more research is needed to identify the key similarities and differences between SAD and WM.

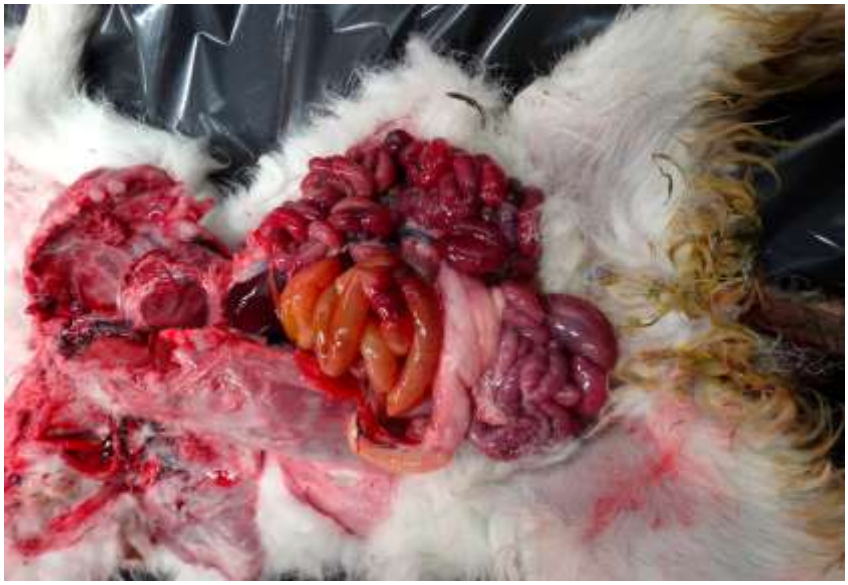

Figure 3: Haemorrhagic fluid distending the jejunum of a lamb with lamb dysentery. Diarrhoea evident staining the perineum.

Some clinical signs are shared between lambs with WM and those with LDLAS, however those with LDLAS also tend to be older - approximately 2 weeks of age, and do not present with hyper-salivation. Enteritis was noted in 2/10 cases of LDLAS together with nephrotic changes in all 10 cases, however endotoxaemia was not a feature of the biochemical values (Angell and others 2013). Furthermore, whilst acidosis is a feature of LDLAS and possibly SAD, an alkalosis has been observed in some cases of WM (Scott and Gessert 1996) until the latter stages when a metabolic acidosis results (Hodgson and others 1989b).

Biochemical and haematological data

The mean biochemical and haematological data reported by Eales and others (1986) revealed no abnormalities, and similar findings were demonstrated by Scott and Gessert (1996). However, in an experimental model, plasma lactate, and urea concentrations increased significantly in cases compared to controls towards the *terminal stages* of disease and plasma glucose, total protein (mostly the globulin component) and the number of white blood cells decreased significantly in cases compared to controls (Hodgson and others 1995; Hodgson and others 1989b).

Pathology

Only one systematic study of pathological changes in lambs with WM was identified - Gilmour and others (1985) - and the results from these 38 lambs are summarised in

Table ***3***. Enteritis was a feature of the disease in 25 out of 38 cases (66%), although the causal nature cannot be ascertained due to the cross-sectional nature of the study. Retained meconium was only present in 12 (32%) and the changes seen in the liver, lung and nervous system appear incidental compared to the changes observed in the alimentary tract. In this study there was no correlation between WM and low colostrum absorption, although immunoglobulins were not assessed by methods other than histology. In the study by Collins and others (1985) lower values of immunoglobulin G may have been associated with an increase in *mortality*, however there was wide variation within the small sample (n=23). In a natural experimental model of infection 9/18 lambs developed WM

and these were reported to have inflammation of the gastrointestinal tract (also distended with gas), pale kidneys and muscle, a dehydrated carcass and enlarged mesenteric lymph nodes (Hodgson and others 1999). In a single case from New Zealand, the abomasum was pale, gas filled and contained a small milk clot and clear mucoid fluid. The small intestine was also pale and gas filled and contained sparse contents. The bladder serosa was congested and the pericardium contained 10ml of serous fluid (Sargison and others 1997). Similarly, Shaw (1981) reports that the abomasum usually contains an excessive quantity of clear mucous, which may or may not be mixed with the colostrum/milk and there may be a retained meconium.

In comparison, abomasal haemorrhages, abomasal bloat and the presence of excess saliva in the abomasum were a feature in cases of SAD and some also had nephrosis (Christodoulopoulos 2008), but neither abomasal haemorrhages or nephrosis were reported in the WM cases by Gilmour and others (1985). In LDLAS the pathological picture was also different to WM with nephrosis a feature in all 10 cases and mild enteritis only identified in 3/10 (Angell and others 2013).

Body region/organ system and pathological change observed	Number of lambs (Percentage of lambs*)
External examination	
External evidence of diarrhoea	11 (29)
Rumen	
pH (n=37)	Mean 5.8 (SE 0.15)
Milk present	1 (3)
Watery content	37 (97)
Abomasum	
pH (n=38)	Mean 3.9 (SE 0.15)
Milk/clots present	20 (53)
No milk present	18 (47)
Inflammatory changes present in mucosa	1 (3)
Bacteria isolated	
Small intestine	
Empty	1 (3)
Liquid	37 (97)
Inflammatory changes present in mucosa (n=25)	12 (48)
Histological evidence of colostrum absorption	29 (76)
Large intestine	
Empty	1 (3)
Liquid	16 (42)
Firm	9 (24)
Meconium present	12 (32)
Inflammatory changes present in mucosa (n=25)	13 (52)
Inflammatory changes present in lambs with retained meconium (n=12)	8 (67)
Liver	
Hepatitis associated with ascending umbilical infection	2 (5)
Fatty degeneration	4 (11)
Lung	
Suppurative broncho-pneumonia	1 (3)
Mild interstitial pneumonia	9 (24)
Retention of some fetal characteristics	4 (11)
Nervous system	
Mild non-suppurative meningoencephalitis	1 (3)
Perivascular lymphocyte cuffs in the coeliacomesenteric ganglion	2 (5)

Table 3: Summary of pathological changes observed in 38 lambs with WM as reported by Gilmour and others (1985). *N=38 unless otherwise specified.

Bacteriology

In an observational study, Gilmour and others (1985) reported a bacteraemia at the point of euthanasia, with *E. coli* identified in 14/20 (70%) lambs and culture of abomasal and intestinal samples revealed coliform, clostridia and streptococcus species to be most prevalent. In another observational study, positive bacterial cultures from blood samples obtained from 71 cases (38%) demonstrated coliforms (31%), *Staphylococcus* spp. (30%) and *Bacillus* spp. (20%) (Eales and others 1986). A pure growth of *E. coli* (without the K99 antigen) was also identified from the small intestine, liver and pericardial fluid in a single case from New Zealand (Sargison and others 1997).

In experimental studies, colostrum-deprived lambs reared in a contaminated environment were shown to frequently develop a bacteraemia within two - twelve hours of birth and a bacteraemia was a persistent feature of the lambs with WM compared to controls, with all the isolates identified as coliforms (Hodgson and others 1999; Hodgson and others 1992). A bacteraemia was also demonstrated experimentally in gnotobiotic lambs experimentally inoculated by mouth using an *E. coli* strain isolated from a clinical case (Hodgson and others 1995). However, this bacteraemia was more intermittent and of lower concentration (cfu/ml) in lambs given human endotoxin-core hyperimmune IgG prior to inoculation compared to those given a saline control.

In comparison, *E. coli* were only cultured from 6/37 abomasal samples from cases of SAD (Christodoulopoulos and others 2013) and were found to be prevalent in cases of LDLAS (Angell and others 2013).

Aetiology and Pathogenesis

The specific aetiology of watery mouth is unknown and indeed there may not be a specific aetiological agent. The pathogenesis so far has only partially been unravelled and there remain key gaps in our knowledge, however three key features appear pertinent and are likely to play a role in the pathogenesis: 1) the ingestion of *E. coli* from a contaminated environment, 2) the production of endotoxin and 3) a delay in abomasal emptying.

Biochemical analysis has shown in both observational and experimental studies that endotoxins – possibly released as a result of lysis of large numbers of gram negative bacteria – play a role in the pathogenesis and are responsible for the majority of the clinical signs (Hodgson 1993; Hodgson and others 1995; Hodgson and others 1989a; Hodgson and others 1989b; Sosa and others 1994). Experimental work has shown that *E. coli* serotypes not currently considered 'pathogenic' may cause the disease when orally inoculated into gnotobiotic lambs, however there have been no comprehensive bacterial surveys of the *E. coli* serotypes associated with watery mouth and it is currently unclear whether any *E. coli* can cause the symptoms seen, given the right conditions, or whether certain pathotypes are more commonly associated with disease.

The effects of endotoxin are well studied and have been reviewed extensively by Hodgson (2006). It can cause hypoglycaemia, reduce body temperature, depress appetite and induce gut stasis and lead to death via multiple critical organ failure

(Hodgson 1993). Specifically endotoxin has been shown to have the following effects on neonatal lambs:

1. *Effects on body temperature*

In three lambs aged 4-11 days, *E. coli* derived endotoxin was shown ($P<0.05$) to accelerate the rate of decline in summit metabolism – that is lambs found it harder to keep warm - compared to control lambs (Alexander 1970). In another study, endotoxin derived from *E. coli* induced a smaller rise in temperature in lambs compared to ewes when administered intravenously (Coceani and others 1995) and additionally failed to elicit a significant change in temperature compared to saline treated control lambs (Goelst and others 1992). These findings may explain why many lambs clinically appear normothermic with WM (Collins and others 1985; Eales and others 1986).

2. *Effects on the cardiovascular system*

Sosa and others (1994) administered very large doses of *E. coli* derived endotoxin (0.5mg/kg) to neonatal lambs (aged 0-3 days) and studied the effects on ventricular contractility. The mean arterial blood pressure decreased as a result of a decline in myocardial contractility. The adverse effects were shown to be stabilised with saline boluses resulting in survival of these lambs compared to those that did not receive saline. However, this study used much larger[1] doses of

[1] Working on a circulating blood volume of 60ml/kg (SWENSON, M. J. (1993) Physiological Properties and Cellular and Chemical Constituents of Blood. In Dukes' Physiology of Domestic Animals. 11 edn. Eds M. J. SWENSON, W. O. REECE. USA, Cornell University Press. p 45) and a weight of 4kg for a new born lamb, then a lamb may have an approximate circulatory volume of 60 x 4 = 240ml of blood. A dose of 0.5mg/kg is 4 x 0.5 = 2mg, therefore 2mg/240ml = 0.008mg/ml or 8,000ng/ml. This is approximately 33 times more than was observed in an experimental infection.

endotoxin compared to the concentration observed (240ng/ml) as a result of experimental infection with orally inoculated *E. coli* (Hodgson and others 1989a) and therefore these effects of endotoxin are likely to be much less in a natural infection. In a similar study Bech-Jansen and others (1972) did not show any change in arterial blood pressure, however the follow-up in that study was only for 30 minutes, which when compared to the study by Sosa and others (1994) was not long enough to detect an effect. Feng and others (2010) showed that cerebral blood flow could be reduced following the administration of *E. coli* derived endotoxin, however again they used much higher doses (10 times) than may be detected in cases of watery mouth. However, even a minor dysregulation of the cerebral blood flow could exacerbate clinical signs, accelerate decline and reduce the chances of recovery.

There is good evidence for the role of endotoxins in WM. The administration of human IgG polyclonal antibody to endotoxin core prior to the intravenous infusion of *E. coli* was successful in preventing clinical, biochemical and haematological signs of WM in 5 lambs, compared to 3 control lambs infused with human plasma albumin (Hodgson and others 1995). In that study, none of the five lambs pretreated with the polyclonal antibody developed signs of WM, but two out of three control lambs did.

Delayed abomasal emptying has been observed clinically (Collins and others 1985) and this was confirmed in an experimental study comparing 34 cases with WM with 68 healthy controls (Eales and others 1985). In this study, contrast radiography with barium sulphate was used to compare the rate of abomasal

emptying. Delayed abomasal emptying was observed in healthy lambs aged 24-48 hours, however gut tone and motility were depressed to a greater extent in the lambs with WM ($P<0.05$). More recently, the acetaminophen absorption test was also used experimentally to investigate the effects of an infusion of endotoxin (2μg/kg) in lambs aged 1-3 weeks (Mozaffari and others 2018). In that study, the mean C_{MAX}, T_{MAX} and AUC were lower ($P<0.05$) for the five treatment lambs compared to the five control lambs, suggesting delayed abomasal emptying as a result of the endotoxin.

An aetiopathogenic hypothesis

The integration of this evidence has led to the development of the following aetiopathogenic hypothesis:

The abomasal contents of the newborn lamb is relatively neutral and the gut has relatively reduced motility - which may serve normally to assist in the uptake of colostrum derived immunoglobulins (Eales and others 1985; Leek 1993). Paradoxically, this may also allow the survival and growth of bacteria ingested from the environment, especially in the absence of colostrum, and this may be compounded by the effect of bacterial endotoxin. The normal pinocytotic mechanisms, which exist for the first 24 hours of life to transport large molecules from the gut into the systemic circulation, may allow access by bacteria and result in a bacteraemia (Hodgson and others 1989a; Hodgson and others 1992). Some bacteria may express factors that opportunistically assist in this process e.g. the aerobactin-mediated iron transport system (Der Vartanian and others 1992).

Lysis *en-masse* of gram-negative bacteria may lead to large volumes of endotoxin resulting in an endotoxaemia and its clinical sequelae (

Figure ***4***) (Hodgson 1993; Hodgson and others 1989b; King and Hodgson 1991; Mozaffari and others 2018; Sosa and others 1994).

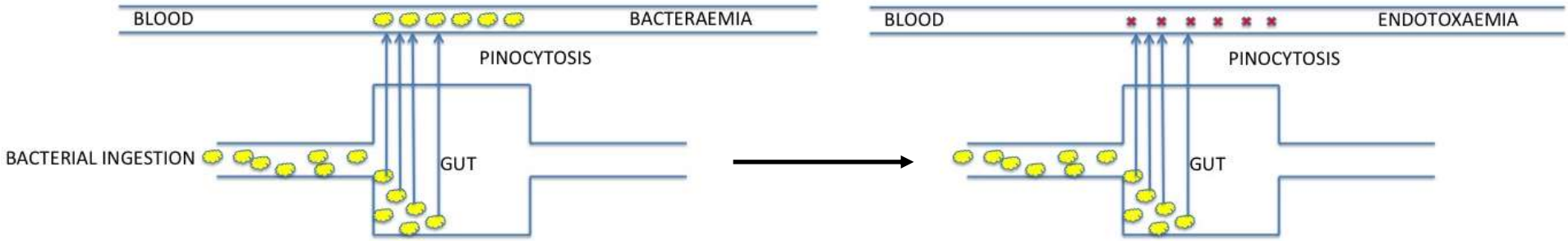

Figure 4: A schematic diagram illustrating a proposed pathogenic hypothesis of the development of WMD. The clinical signs and subsequent sequelae are then strongly associated experimentally and observationally with the endotoxaemia.

Epidemiology

Frequency and distribution

Watery mouth disease has been reported widely in the UK, but we are also aware of reports from France (including a similar condition in neonatal goat kids) (Poncelet 2004; Savey 1986), Spain (Garcia de Jalon and others 1990), Canada (Hodgson 1993), New Zealand (Orr 1995; Sargison and others 1997) and Norway (Phythian, C.J., 2016, personal communication). It seems likely that it could occur anywhere where sheep are produced under intensive conditions.

Accurate up-to-date data on prevalence or incidence rates at regional or national level are unavailable, however personal experience and numerous discussions with practitioners would suggest that WM is widespread, with large variations between farms and within farms from year to year. Unpublished pilot work with one large UK veterinary practice in 2015 revealed that 64.6% of 895 farmers used some form of prophylactic/metaphylactic antibiotic medication to prevent or mitigate the losses caused by WM, implying that this proportion either currently see or have seen cases historically. In addition, Davies and others (2017) reported 47% (95%CI: 41-62%) of 207 flocks (convenience sample) located in England and Wales used licensed oral antibiotics for treatment/prophylaxis of colibacillosis, with a further 2 flocks prescribed oral antibiotic tablets.

Historically, Collins and others (1985) reported an incidence rate on six farms of up to 3.1% and the mortality of affected cases as 41% - although mortality was observed to worsen with age of onset with those aged more than 48 hours when infected being less likely to recover. Eales and others (1986) reported an

incidence rate of up to 24% of lambs born and mortality of affected cases up to 83%, however in this study there was large variation between the 11 farms with those farms with greater contact with the investigators (Woodhouselee, Glencorse and Longyester) reporting a higher incidence rate and lower mortality of affected cases compared to those with less contact (farms A-H). The initial data reported by Scott (1988) suggests that in the control groups across eight farms, the incidence rate was reported to be up to 43% of lambs born at which point prophylactic therapy was instigated, and mortality of affected cases up to 25%, despite treatment. Finally, Hodgson (1993) reported that from a survey of 170 farmers owning 100,000 ewes WM was responsible for 23% of deaths among housed lambs.

Factors associated with watery mouth

Risk factors for watery mouth include increased litter size – triplets being at greater risk than twins (Collins and others 1985) and lambs from multiple litters more likely than from single litters ($P<0.01$) (Eales and others 1986). Also, Eales and others (1986) reported smaller lambs to be slightly more at risk ($p<0.05$) although this difference was not observed in cases when compared to their healthy twin. Additionally lambs born to ewes of poor condition were more at risk although there may have been some confounding by, or interaction with age, which was also associated - lambs were more at risk when born to ewes aged one or five years but at lower risk when born to ewes aged three years. Early castration (within the first 60 minutes of life) has also been shown to increase the risk of developing WM possibly as a result of delayed ingestion of colostrum (Collins and others 1985). Furthermore, incidence rates may vary considerably

from year to year which may suggest that combinations of environmental and management factors may be important (Collins 1981; Eales and others 1986).

It has been suggested that these factors combined lead to reduced colostrum intake increasing the risk of acquiring WM (Eales 1987; King and Hodgson 1991). In experimental studies adequate colostrum intake reduces the risk of WMD (Hodgson and others 1999; Hodgson and others 1992), however both Collins and others (1985) and Eales and others (1986) showed no association with colostrum derived IgG in lamb serum. Watery mouth disease has been reported to be more common in lambs born to housed ewes, although it can occur in ewes lambed outside (Collins 1981; King and Hodgson 1991; Sargison 2008a), and it is hypothesised that wet and insanitary conditions lead to an increased risk of ingesting the pathogen by the lamb. This is corroborated by data from Scott (1988), who reported increased incidence over the lambing period in eight flocks, and is also supported by data from Binns and others (2002) who showed that after adjusting for other confounders those farmers who failed to bed down the mothering pens used at lambing daily reported higher mortality rates compared to those that did.

Clearly, the epidemiology of WM is poorly understood in terms of the frequency, distribution and determinants of disease, indeed we are limited mainly to one admirable yet flawed investigation from 1986. The scant available evidence and clinical experience would suggest the disease is widespread and endemic in a majority of flocks. A sound understanding of the epidemiology of WM is required

to enable its prevention and control and facilitate appropriate targeted treatment as opposed to the current situation of unsustainable mass antibiosis.

Treatment

There have been no well-designed intervention studies and the few reports available have been aimed at targeting the perceived aetiopathogenesis. There are two case reports investigating the response to empirical treatment: in Eales and others (1986) cases of WM were treated with antibiotics administered enterally (neomycin and streptomycin) and parenterally (amoxicillin), glucose/electrolyte solution administered by stomach tube, together with not feeding milk but leaving the lamb to suck the ewe if and when it recovered. This was an uncontrolled study but was reported to be successful in 89% of cases. A further report investigated the treatment response following amoxicillin plus clavulanic acid injection together with intravenous flunixin meglumine and an oral rehydration solution. This protocol was successful in 21 of 23 cases (91.3%) (Scott and Gessert 1996). Other anecdotes include the use of oral flunixin meglumine in isolation (Mitchell 1992), paracetamol (acetaminophen) (Nixon 1991), warm water enemas (Robinson 1981; Shaw 1981), laxatives (Shaw 1981; Watt. A. 1980) and metoclopramide in attempt to restore or increase gut motility (Mitchell and Linklater 1983; Scott 1988).

It is possible that antibiotic treatment of already bacteraemic lambs may result in a more severe endotoxaemia due to the increased release of endotoxin as a result of bacterial death. In two studies: Shenep and others (1985); Shenep and Mogan (1984) a rabbit model was used to show that plasma endotoxin concentration

could increase to much greater levels following the administration of an antibiotic compared to a placebo, but this could vary depending on the antibiotic used. Clearly, well designed intervention studies are necessary in order to develop an evidence base for treated cases with WM, including supportive therapy aimed at treating the endotoxaemia and also a particular focus on the selection of particular antibiotics and timing and route of administration.

Prevention and Control

Hodgson and others (1999) clearly demonstrated that lambs fed either colostrum or ewe milk replacer with the addition of oral spectinomycin as the first feed were prevented from developing WM in a natural infection model compared to lambs just fed ewe milk replacer. In that study, 9/18 lambs developed WM when fed just milk replacer but no lambs from the other two groups developed disease. The use of this antibiotic was justified on the basis of its poor absorption from the gut and its bacteriostatic activity, in an attempt to reduce the liberation of endotoxin *en-masse* from lysed bacteria (Shenep and others 1985; Shenep and Mogan 1984).

On a single farm Eales and others (1986) examined two preventative protocols. In the first they administered a combination of neomycin and streptomycin orally (n=58) and in the second amoxicillin orally (n=46), with both interventions given within 15 minutes of birth. Both interventions were pseudo-randomly allocated and compared to lambs receiving no treatment (n=60 and n=63 respectively). Both interventions were shown to prevent WM compared to the control lambs ($P<0.001$ and $P<0.05$ respectively), however the study is weakened by the lack of blinding, the possibility of bias during randomisation and the lack of control

substance/placebo in the control lambs. Further analysis using the t test suggested that whilst the four farms that used antibiotics prophylactically had lower incidence rates compared to those that did not (mean 1.1% (SD 0.47) of lambs born, P=0.03), they had relatively high mortality rates in affected cases (mean 59.5% (SD 30.0) of cases P<0.001).

Metoclopramide has been used prophylactically as a preventative in a placebo controlled study of eight farms. On four of these farms there was a reduced incidence compared with the placebo, however in one farm the association was reversed. In the other three flocks the incidence in both groups was too low for comparisons to be made (Scott 1988).

On the basis of these small studies, veterinary advice has included recommending adequate colostrum intake, reducing exposure to infection by ensuring clean dry bedding and cleaning and disinfecting lambing pens between occupants, and administering oral antibiotics prophylactively/metaphylactically within 15 minutes of birth. Much of this advice remains sound although the routine prescribing of antibiotics for prophylactic/metaphylactic use is now clearly unsuitable and irresponsible (Sheep Veterinary Society 2017).

Vaccination

A vaccine would be an attractive option, however the onset of disease is so rapid in clinical cases that a vaccine administered to lambs at birth is unlikely to be efficacious. Vaccinating the ewe would be possible to boost immunoglobulins in colostrum if specific *E. coli* were associated with disease, as has been shown for

some pathogenic *E. coli* (Acres and others 1979; Altmann 1983; Cameron and Fuls 1970; Gregory and others 1983; Morris and others 1980; Pugh and Wells 1985; Sojka and others 1978; Wray and others 1983), however this could be unsuccessful if lambs still fail to get colostrum.

Alternative strategies for prevention

Sodium chlorate was shown to reduce the number of generic *E. coli* in the colon of treated lambs compared to controls, although the effect seen was weak (P=0.06) possibly due to the low power of the study. However, this option is unlikely to be pursued due to the risks of toxicity (Taylor and others 2012).

Two other intervention strategies worthy of investigation include the use of probiotic preparations (Bilková and others 2013; Zhakupova and others 2017) and the use of bacteriophages (Johnson and others 2008; Smith and Huggins 1983). Currently the normal development of the ovine gastrointestinal microbiome is unknown, however fostering the development of a healthy microbiome may serve to outcompete pathogenic species or the overgrowth of less desirable commensal species. This has been attempted with some success in experimentally reducing the growth of *E. coli* with the K99 antigen and may warrant further investigation (Bomba and others 1997; Timmerman and others 2004). Studies of the microbiome would also serve to inform whether the use of bacteriophages may be worthy of exploration. Several studies again looking specifically at known pathogenic *E. coli* have shown promising results in neonatal ruminants including lambs (Johnson and others 2008; Smith and Huggins 1983). In order to be successful, specific *E. coli* associated with WM would need to be

identified in order to investigate possible phage candidates as targeted therapeutics.

Conclusions

The clinical picture of WM appears well described and the pathophysiology has been elegantly demonstrated to be attributed to the production of endotoxin, probably as a result of undefined, previously considered apathogenic *E .coli*. The reasons for these agents becoming established and causing disease in lambs are unknown although early ingestion from an insanitary environment is plausible. However, the pathology and epidemiology are poorly understood in turn hampering prevention and therapeutic investigation. We believe WM to be widespread, endemic and largely ignored due to the routine use of prophylactic and metaphylactic antibiosis, a state of affairs which needs to be addressed immediately. Given the need to improve animal welfare, improve food security, develop farm profitability and reduce antibiotic use in food producing animals, there is clearly an urgent need to assess the current incidence of WM, develop a rigorous understanding of the farm, animal and environmental determinants of disease and develop appropriate and effective non-antibiotic approaches to disease prevention and control. In addition, further investigation using newer technologies of the developing microbial communities within the neonatal gastrointestinal tract may help determine if there are specific pathogens associated or if disease actually occurs due to a deviation from normal physiology or the developmental microbiology. From an animal welfare perspective, well designed treatment studies are warranted in order to treat sick cases appropriately and effectively.

References

ACRES, S. D., ISAACSON, R. E., BABIUK, L. A. & KAPITANY, R. A. (1979) Immunization of calves against enterotoxigenic colibacillosis by vaccinating dams with purified K99 antigen and whole cell bacterins. Infect Immun 25, 121-126

ALEXANDER, G. (1970) Summit metabolism in young lambs. Effect of hormones and drugs that affect mobilization and utilization of substrate for cold-induced thermogenesis. Biol Neonate 15, 37-48

ALTMANN, K. (1983) Passive immunisation of neonatal lambs against infection with enteropathogenic *Escherichia coli* via colostrum of ewes immunised with crude and purified K99 pili. Res Vet Sci 35, 234-239

ANGELL, J. W., JONES, G., GROVE-WHITE, D. H., JONES, E., HIGGINS, R. J. & OTTER, A. (2013) A prospective on farm cohort study investigating the epidemiology and pathophysiology of drunken lamb syndrome. Vet Rec 172, 154

BECH-JANSEN, P., BRINKMAN, C. R., 3RD, JOHNSON, G. H. & ASSALI, N. S. (1972) Circulatory shock in pregnant sheep. II. Effects of endotoxin on fetal and neonatal circulation. Am J Obstet Gynecol 113, 37-43

BILKOVÁ, A., DUBNIČKOVÁ, M. & SEPOVÁ HANA, K. (2013) Antimicrobial susceptibility and immunomodulatory properties of lamb isolate of lactobacillus mucosae, new probiotic candidate. Acta Facultatis Pharmaceuticae Universitatis Comenianae 60, 1

BINNS, S. H., COX, I. J., RIZVI, S. & GREEN, L. E. (2002) Risk factors for lamb mortality on UK sheep farms. Prev Vet Med 52, 287-303

BOMBA, A., KRAVJANSKÝ, I., KAŠTEL, R., HERICH, R., JUHÁSOVÁ, Z., ČÍŽEK, M. & KAPITANČÍK, B. (1997) Inhibitory effects of Lactobacillus casei upon the adhesion of enterotoxigenic Escherichia coli K99 to the intestinal mucosa in gnotobiotic lambs. Small Ruminant Research 23, 199-206

CAMERON, C. M. & FULS, W. J. (1970) Immune response to dead and live Escherichia coli vaccines and colostral transfer of immunity to calves and lambs. Onderstepoort J Vet Res 37, 157-163

CARSON, A. (2019) Hypogammaglobulinaemia and other causes of death in neonatal lambs. Vet Rec 184, 17-18

CHRISTODOULOPOULOS, G. (2008) Salivary abomasum disease in young lambs. Veterinary Record 162, 732

CHRISTODOULOPOULOS, G., SCOTT, P. R., JEHL, N., FILIOUSSIS, G. & SMITH, S. H. (2013) Clinical, microbiological and histological findings in lambs affected by 'salivary abomasum disease'. Vet Rec 172, 100

COCEANI, F., BISHAI, I., ENGELBERTS, D., HOUSE, R. V. & ADAMSON, S. L. (1995) Response of newborn and adult sheep to pyrogens: relation between fever and brain eicosanoid changes. Brain Res 700, 191-204

COLLINS, R. (1981) Lamb survival. Veterinary Record 109, 43-44

COLLINS, R. O., EALES, F. A. & SMALL, J. (1985) Observations on watery mouth in newborn lambs. Br Vet J 141, 135-140

DAVIES, P., REMNANT, J. G., GREEN, M. J., GASCOIGNE, E., GIBBON, N., HYDE, R., PORTEOUS, J. R., SCHUBERT, K., LOVATT, F. & CORBISHLEY, A. (2017) Quantitative analysis of antibiotic usage in British sheep flocks. Vet Rec 181, 511

DER VARTANIAN, M., JAFFEUX, B., CONTREPOIS, M., CHAVAROT, M., GIRARDEAU, J. P., BERTIN, Y. & MARTIN, C. (1992) Role of aerobactin in systemic spread of an

opportunistic strain of Escherichia coli from the intestinal tract of gnotobiotic lambs. Infection and Immunity 60, 2800-2807
DWYER, C. M., CONINGTON, J., CORBIERE, F., HOLMOY, I. H., MURI, K., NOWAK, R., ROOKE, J., VIPOND, J. & GAUTIER, J.-M. (2016) Invited review: Improving neonatal survival in small ruminants: science into practice. Animal 10, 449-459
EALES, A. (1987) Watery mouth. In Practice 9, 12-17
EALES, F. A., SMALL, J., GILMOUR, J. S., DONACHIE, W., FITZSIMONS, J. & DINGWALL, W. S. (1986) A field study of watery mouth: clinical, epidemiological, biochemical, haematological and bacteriological observations. Vet Rec 119, 543-547
EALES, F. A., SMALL, J., MURRAY, L. & MCBEAN, A. (1985) Abomasal size and emptying time in healthy lambs and in lambs affected by watery mouth. Vet Rec 117, 332-335
FENG, S. Y., SAMARASINGHE, T., PHILLIPS, D. J., ALEXIOU, T., HOLLIS, J. H., YU, V. Y. & WALKER, A. M. (2010) Acute and chronic effects of endotoxin on cerebral circulation in lambs. Am J Physiol Regul Integr Comp Physiol 298, R760-766
FINDLAY, C. (1973) Serum immune globulin levels in lambs under a week old. Veterinary Record 92, 530-532
GARCIA DE JALON, J. A., DE LAS HERAS, M., FERRER, L. M. & SANCHO, F. (1990) Sindrome de la boca mojada en corderos (Watery mouth syndrome in lambs). Medicina Veterinaria 7, 505-509
GILMOUR, J. S., DONACHIE, W. & EALES, F. A. (1985) Pathological and microbiological findings in 38 lambs with watery mouth. Vet Rec 117, 335-337
GOELST, K., MITCHELL, D. & LABURN, H. (1992) Fever responses in newborn lambs. Pflugers Arch 421, 299-301
GREGORY, D. W., CARDELLA, M. A. & MYERS, L. L. (1983) Lamb model in the study of immunity to enteropathogenic Escherichia coli infections. Am J Vet Res 44, 2073-2077
HAIG, G. (1981) Lamb survival. Veterinary Record 108, 195
HCC (2011) Making Every Lamb Count. Hybu Cig Cymru/Meat Promotion Wales. pp 2-3
HODGSON, J. C. (1993) Watery mouth disease in newborn lambs. Veterinary Annual 33, 102-106
HODGSON, J. C. (2006) Endotoxin and mammalian host responses during experimental disease. J Comp Pathol 135, 157-175
HODGSON, J. C., BARCLAY, G. R., HAY, L. A., MOON, G. M. & POXTON, I. R. (1995) Prophylactic use of human endotoxin-core hyperimmune gammaglobulin to prevent endotoxaemia in colostrum-deprived, gnotobiotic lambs challenged orally with Escherichia coli. FEMS Immunol Med Microbiol 11, 171-180
HODGSON, J. C., BREBNER, J. & MCKENDRICK, I. J. (1999) Efficacy of a single dose of oral antibiotic given within two hours of birth in preventing watery mouth disease and illthrift in colostrum-deficient lambs. Vet Rec 145, 67-71
HODGSON, J. C., KING, T., MOON, G., DONACHIE, W. & QUIRIE, M. (1989a) Host responses during infection in newborn lambs. FEMS Microbiol Immunol 1, 311-312
HODGSON, J. C., KING, T. J., HAY, L. A. & ELSTON, D. A. (1989b) Biochemical and haematological evidence of endotoxic shock in gnotobiotic lambs with watery mouth disease. Res Vet Sci 47, 119-124

HODGSON, J. C., MOON, G. M., HAY, L. A. & QUIRIE, M. (1992) Effectiveness of substitute colostrum in preventing disease in newborn lambs. BSAP Occasional Publication 15, 163-165
HOLMOY, I. H., WAAGE, S., GRANQUIST, E. G., L'ABEE-LUND, T. M., ERSDAL, C., HEKTOEN, L. & SORBY, R. (2016) Early neonatal lamb mortality: postmortem findings. Animal, 1-11
JOHNSON, R. P., GYLES, C. L., HUFF, W. E., OJHA, S., HUFF, G. R., RATH, N. C. & DONOGHUE, A. M. (2008) Bacteriophages for prophylaxis and therapy in cattle, poultry and pigs. Anim Health Res Rev 9, 201-215
KING, T. (1986) Pathogenesis and control of watery mouth in lambs. The Veterinary Record : Journal of the British Veterinary Association 119, 211-211
KING, T. & HODGSON, C. (1991) Watery mouth in lambs. In Practice 13, 23-24
LEEK, B. F. (1993) Digestion in the Ruminant Stomach. In Dukes' Physiology of Domestic Animals. Eds M. J. SWENSON, W. O. REECE. London, Uk, Cornell University Press. pp 397-416
LINKLATER, K. A. (1989) Watery mouth in lambs. Veterinary Annual 29, 88-92
MILLEMANN, Y., ADJOU, K., MAILLARD, R., POLACK, B. & CHARTIER, C. (2003) Les diarrhées néonatales des agneaux et des chevreaux (Neonatal diarrhoea in lambs and kids). Le Point Veterinaire 233, 22-29
MITCHELL, G. & LINKLATER, K. (1983) Differential diagnosis of scouring in lambs. In Practice 5, 4-12
MITCHELL, K. W. (1992) Treatment of 'watery mouth'. Vet Rec 130, 384
MORRIS, J. A., WRAY, C. & SOJKA, W. J. (1980) Passive protection of lambs against enteropathogenic Escherichia coli: role of antibodies in serum and colostrum of dams vaccinated with K99 antigen. J Med Microbiol 13, 265-271
MOZAFFARI, A. A., JARAKANIBAYAT, S., VOSOUGH, D., ESKANDARZADE, N. & TAJIC, J. (2018) The effect of the endotoxemia on the abomasal emptying rate in suckling lambs as shown by acetaminophen absorption test. Comparative Clinical Pathology 27, 283-287
NIXON, S. R. (1991) Treatment of watery mouth in lambs. Vet Rec 129, 58
O'NEILL, J. (2016) Tackling Drug-Resistant Infections Globally: Final Report and Recommendations., Review on Antimicrobial Resistance
ORR, M. B. (1995) Animal Health Laboratory Network review of diagnostic cases - October to December 1994, Ministry for Primary Industries
PONCELET, J. L. (2004) Les Colibacilloses. In Journees nationales des GTV, Societe Nationale des Groupements Techniques Veterinaires
PUGH, C. A. & WELLS, P. W. (1985) Protection of lambs against enteric colibacillosis by vaccination of ewes. Res Vet Sci 38, 255-258
ROBINSON, J. (1981) Lamb survival. Veterinary Record 108, 130-131
SARGISON, N. D. (2008a) The Lambing Percentage. In Sheep Flock Health: A planned approach. . Oxford, UK, Blackwell Publishing Ltd.
SARGISON, N. D. (2008b) The Lambing Percentage. In Sheep Flock Health: A planned approach. Oxford, UK, Blackwell Publishing Ltd.
SARGISON, N. D., WEST, D. M., PARTON, K. H., HUNTER, J. E. & LUMSDEN, J. S. (1997) A case of "watery mouth" in a New Zealand Romney lamb. N Z Vet J 45, 67-68
SAVEY, M. (1986) La maladie ovine du 'watery mouth', un modele pour comprendre le 'chevreau mou'? Pointe Veterinaire 17, 738-740

SCOTT, P. R. (1988) The possible use of metoclopramide to prevent watery mouth in lambs in commercial flocks. Br Vet J 144, 570-572
SCOTT, P. R. (2007) Neonatal Lamb Diseases. In Sheep Medicine. London, UK, Manson Publishing Ltd. pp 89-91
SCOTT, P. R. & GESSERT, M. E. (1996) Watery mouth disease in lambs: biochemical parameters before and after treatment. Vet Rec 139, 117-118
SHAW, W. B. (1981) Lamb survival. Vet Rec 108, 265
SHEEP VETERINARY SOCIETY (2017) Sheep Veterinary Society Policy on Responsible Use of Antimicrobials in SheepAccess, 2017
SHENEP, J. L., BARTON, R. P. & MOGAN, K. A. (1985) Role of antibiotic class in the rate of liberation of endotoxin during therapy for experimental gram-negative bacterial sepsis. J Infect Dis 151, 1012-1018
SHENEP, J. L. & MOGAN, K. A. (1984) Kinetics of endotoxin release during antibiotic therapy for experimental gram-negative bacterial sepsis. J Infect Dis 150, 380-388
SMITH, H. W. & HUGGINS, M. B. (1983) Effectiveness of phages in treating experimental Escherichia coli diarrhoea in calves, piglets and lambs. J Gen Microbiol 129, 2659-2675
SOJKA, W. J., WRAY, C. & MORRIS, J. A. (1978) Passive protection of lambs against experimental enteric colibacillosis by colostral transfer of antibodies from K99-vaccinated ewes. J Med Microbiol 11, 493-499
SOSA, G., MILSTEIN, J. M. & BENNETT, S. H. (1994) Escherichia coli endotoxin depresses left ventricular contractility in neonatal lambs. Pediatr Res 35, 62-67
SWENSON, M. J. (1993) Physiological Properties and Cellular and Chemical Constituents of Blood. In Dukes' Physiology of Domestic Animals. 11 edn. Eds M. J. SWENSON, W. O. REECE. USA, Cornell University Press. p 45
TAYLOR, J. B., DUNGAN, R. S. & LEWIS, G. S. (2012) Sodium chlorate reduces the presence of Escherichia coli in feces of lambs and ewes managed in shed-lambing systems. J Anim Sci 90, 381-386
TIMMERMAN, H. M., KONING, C. J., MULDER, L., ROMBOUTS, F. M. & BEYNEN, A. C. (2004) Monostrain, multistrain and multispecies probiotics--A comparison of functionality and efficacy. Int J Food Microbiol 96, 219-233
VAID, J. (1991) Bacteraemia in man and animals: an overview. Vet Res Commun 15, 341-362
WATT. A. (1980) Neontal losses in lambs. In Practice 2, 5-9
WHO (2015) Global Action Plan on Antimicrobial Resistance. Geneva, Switzerland, World Health Organization
WINKLER, C. (1981) Lamb survival. Veterinary Record 108, 342
WRAY, C., SOJKA, W. J. & MORRIS, J. A. (1983) Use of "E. coli" K99 vaccines in ewes to passively protect lambs against experimental colibacillosis. Dev Biol Stand 53, 177-181
ZHAKUPOVA, A. A., BIYASHEV, K. B., BIYASHEV, B. K., ERMAGAMBETOVA, S. E. & NURGOZHAYEVA, G. M. (2017) Performance Test of the Drug "Enterocol". Journal of Pharmaceutical Sciences and Research 9, 1941-1942