



Clostridial Necrotic Enteritis and Cholangiohepatitis — Magne Kaldhusdal, Ph.D.¹

“Biography”

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Clostridial Necrotic Enteritis and Cholangiohepatitis

“Abstract”

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Introduction

From time to time clostridial enteritis turns up as a big problem in various regions. The reasons for these problems are complex, but resistance against in-feed antibacterial and anticoccidial compounds has been one of the factors involved. However, in general these compounds have been very successful in preventing necrotic enteritis, and still are. So why do we see an increasing interest in this disease complex now? One major reason is restrictions on the use of in-feed antibacterial and anticoccidial compounds that have limited the use of these preventive measures, and more restrictions are expected to come. Another reason may be that after 40 years with more or less unpredictable and sometimes severe problems, the broiler (and turkey) industry wants to find out more about how to control the disease.

This paper presents some basic information and evaluations that hopefully are useful in the understanding of this important disease complex.

Etiology

Clostridial enteritis and (cholangio)hepatitis is caused by *Clostridium perfringens* (CP). Toxin types A and C of this bacterium have been associated with disease outbreaks. The typical gut lesions have been reproduced experimentally by intraduodenal infusion of toxins produced by CP type A. The alpha toxin was most likely the factor causing the gut lesions (Al-Sheikhly and Truscott, 1977b). CP toxin type A is ubiquitous, spore forming, extremely prolific, and toxigenic. These attributes enables the bacterium to be present at almost any time and place with poultry, and then proliferate and produce toxins when the circumstances are favourable.

Hosts

Necrotic enteritis has been reported in many bird species. Among chickens the disease is by far most common in broilers, but outbreaks in layer pullets and adult layer strain chickens have also been reported.

Economical significance

Lovland and Kaldhusdal (2001) studied the association between *Clostridium perfringens* infection and production performance in commercial broiler flocks during a 2.5 years time period when clinical and subclinical necrotic enteritis was frequently seen. They found that the farmer's profit on average was reduced by 33 % when comparing flocks with high and low levels of the disease. Impaired feed conversion, reduced live weight at slaughter and increased condemnation percentage were major causes of production losses associated with *Clostridium perfringens* infection. Subclinical clostridial enteritis has also been associated with impaired feed conversion and retarded growth in a pen trial (Kaldhusdal and Hofshagen, 1992).

Clinical finding

The clinical appearance of clostridial enteritis may vary a lot, from no other 'symptoms' than impaired performance to a sudden and high mortality. The disease can affect birds at almost any age, but in our experience the disease most commonly occurs in broilers two weeks of age or older. Age pattern of the disease may vary, depending on factors like immunity and management, including types and programs of in-feed additives.

Pathogenesis and predisposing factors

The pathogenesis of the natural disease is far from well understood. In our experience (Kaldhusdal *et al.*, unpublished data) the pathogenic bacterium is usually present in numbers sufficient to induce subclinical disease, if no antibacterial compounds are added to the feed. However, the appearance of clinical disease even in flocks offered feed with antibacterial compounds, suggests that a number of interacting, known and unknown factors influence the disease process significantly. As with other clostridial diseases, feed composition and feeding practises are assumed to affect disease incidence (Kaldhusdal, 1999). Coccidial infections are assumed to be a predisposing factor in many cases. These organisms invade and damage intact mucosal gut tissue, which is likely to favour the establishment of a non-invasive bacterium like *Clostridium perfringens*. But necrotic enteritis no doubt can appear in the absence of coccidia.

At the molecular level, one or several factors triggering the alpha toxin production of *Clostridium perfringens* may be essential. Once the bacteria produce significant amounts of toxins, these toxins eventually induce gut tissue necrosis, favouring proliferation of *Clostridium perfringens* and further tissue damage within the mucosa.

The development of liver lesions is poorly understood. Both bacteria and toxins may be transported through a damaged gut mucosa to the liver by the blood. An ascending infection via the bile tree, and inflammatory processes obstructing the bile flow, are other possibilities.

Pathology

The characteristic lesions are found in the mucosa of the small intestine. Mild lesions appear as small (in some cases barely visible to the naked eye) ulcers or light yellow spots on the surface of the mucosa, usually in the anterior part of the gut. More severe lesions may show membranes covering the entire mucosa of large segments of the small intestine, in some cases even the colorectum and the caecal tonsils.

Two types of liver lesions have been described (Onderka *et al.*, 1990, Løvland and Kaldhusdal, 1999). An inflammation affecting the bile tree (cholangiohepatitis) is the most common lesion type. Intrahepatic parts of the bile tree are most frequently affected, but gall bladder and extrahepatic bile duct changes may also be found. Another liver lesion type associated with CP infection is multifocal hepatitis, histologically characterized by fibrinoid necroses with or without an inflammatory response.

Experimental disease models

Numerous workers have reproduced clostridial necrotic enteritis experimentally using a challenge model (Al-Sheikhly and Truscott, 1977ab, Prescott *et al.*, 1978, Brennan *et al.*, 2001). We have recently developed a necrotic enteritis model (Kaldhusdal *et al.*, unpublished) based on the assumption that a spontaneous CP infection sufficient to induce a natural, mild clostridial necrotic enteritis usually takes place, providing the appropriate predisposing factors are present. The average frequency of individuals with specific lesions in the eight first experiments using this model was 20.9 %. The model has already been used to test the effect of vaccines, litter types and feed additives (unpublished data) on clostridial enteritis.

Immunity

Few studies on the immunity against *Clostridium perfringens* infection in poultry have been published. Heier *et al.* (2001) found that the level of maternal antibodies to CP alpha toxin in day-old broiler flocks varied considerably and was influenced by parent flock age. Unpublished data from our own laboratory (Kaldhusdal *et al.*) indicate that levels of specific maternal antibodies can be increased substantially by vaccination of the parent hens. Further, this increase in maternal antibodies has been associated with reduced levels of spontaneous necrotic enteritis. Lovland *et al.* (unpublished data) have also found increased levels of specific serum antibodies in flocks with a previous experience of CP infection.

Diagnosis

Clinical disease is best diagnosed by examination for gross intestinal lesions in birds that have died with the disease. Gross lesions are usually distinct from those of coccidiosis. In cases of doubt, direct microscopy of gut smears showing few or none coccidia will help establish the diagnosis. The presence of birds with cholangiohepatitis among bird from such flocks is another helpful indicator, although this is relatively seldom seen. If many coccidia are found in gut smears, and the gross lesions are considered ambiguous, histological and microbiological examinations may be required for a definitive diagnosis. In some cases of mixed infections, it may be impossible to verify which one is of primary importance.

Subclinical disease can best be diagnosed by random sampling of live birds in flocks with vague symptoms or suboptimal performance. Gut lesions must be looked for immediately following euthanasia of the birds. The gross lesions of the intestinal mucosa are specific, but some experience is required to detect and recognize especially the most moderate lesions.

Bacteriology is most useful if quantitative examinations are done. Gut contents or faeces should contain at least 1 million (often 100 and even 1000 millions in individual specimens) CP per gram if CP-associated necrotic enteritis is likely to be the disease cause.

Monitoring

Broiler flocks may be monitored for level of CP infection if reliable records of specific liver lesions at slaughter are available (Løvland and Kaldhusdal, 1999).

Unpublished data from our laboratory (Lovland *et al.*) suggest that broiler flocks may be monitored by examination of blood samples for CP specific antibodies. This method is based on the ELISA technique, and may possibly be established in laboratories using ELISA serology. However, further work is required before the method can be used on a routine basis.

Control

At present clostridial enteritis and hepatitis is best controlled by use of in-feed antibacterial compounds.

Natural gut flora products have been shown to reduce the disease frequency and *Clostridium perfringens* counts in experiments (Elwinger *et al* 1992, Hofacre *et al.*, 1998, Craven *et al.* 1999) and in a farm with recurring problems (Kaldhusdal *et al.*, 2001).

Vaccination may become a viable alternative in the future.

Conclusion

- 1) Clostridial necrotic enteritis cannot be prevented by means of good hygiene alone.
- 2) Clostridial enteritis exists in subclinical as well as clinical form. Both forms affect productivity significantly.
- 3) The diagnosis of clinical and subclinical necrotic enteritis requires different procedures.
- 4) Preliminary findings at our laboratory suggest that clostridial necrotic enteritis may be monitored using an ELISA assay on blood samples from broiler flocks.

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Clostridial Necrotic Enteritis and Cholangiohepatitis

“Slide Presentation”

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Figure G-1.

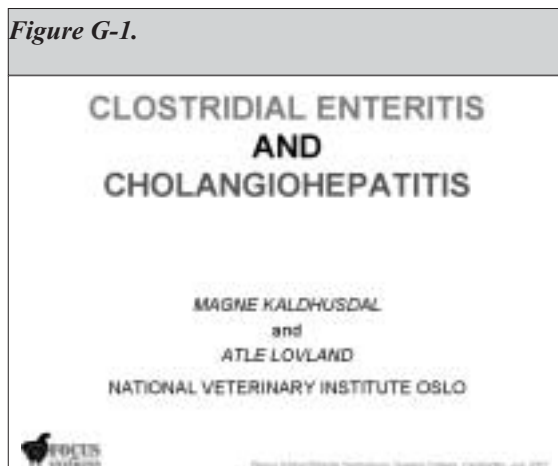


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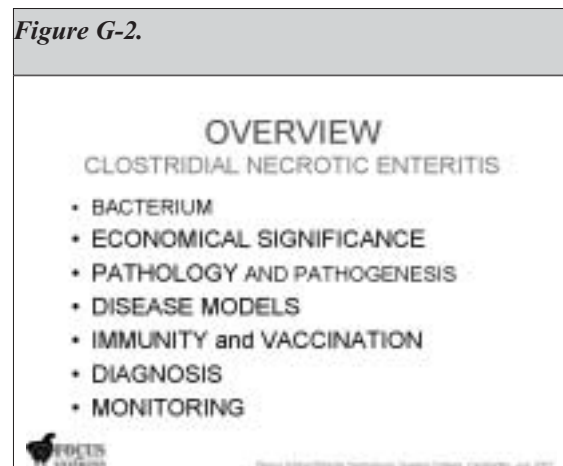


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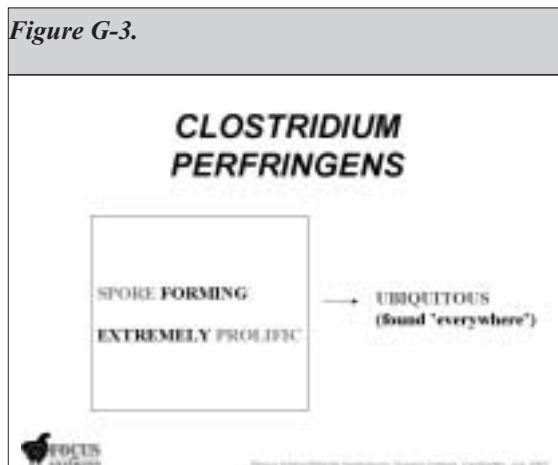


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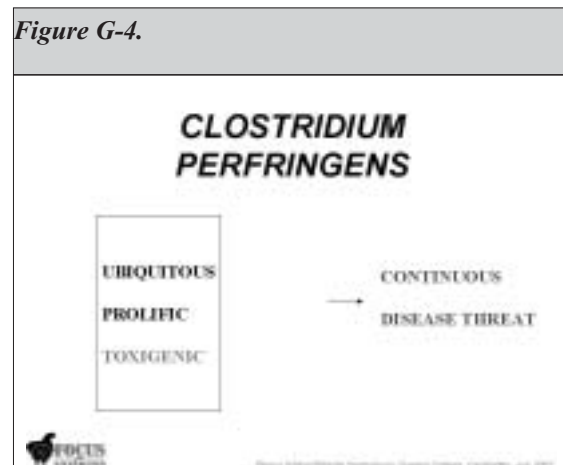



Figure G-5.

200 COMMERCIAL FLOCKS:
**HIGH vs LOW %
 CLOSTRIDIAL
 ENTERITIS & HEPATITIS**

OUTCOME	HIGH/LOW	DIFFERENCE	(P-value)
LIVE WEIGHT (g)	145,71485	28	(0,005)
FCR (kg feed/kg bird)	1,89718,826	71	(<0,001)
MORTALITY (%)	4,530,71	0,82	(<0,001)
CONDENMED (%)	1,360,34	1,02	(<0,001)
PROFIT (low=100)	86,7/100	33,3%	(<0,001)

(Lofrend and Rathusdal, 2001)



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Figure G-6.

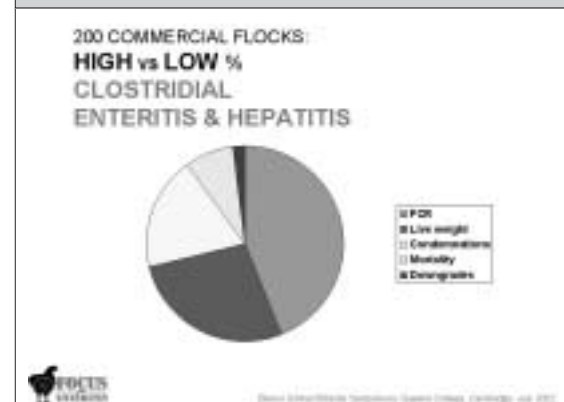



Figure G-7.

**DEFINITION OF
 CLOSTRIDIAL ENTERITIS**

- LESIONS
 - GUT NECROSES?
- TOXIN TYPE
 - ALPHA TOXIN
 - ENTEROTOXIN
- CLOSTRIDIAL
 COUNT > 1 mio/g



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Figure G-8.

**HYPOTHETICAL
 PATHOGENESIS**

EXTRUSION
 ZONE

TOXINS

NECROSIS




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Figure G-9.

**CLINICAL FORMS OF
 CLOSTRIDIUM PERFRINGENS DISEASE**

- NON-NECROTIC CLOSTRIDIAL ENTERITIS?
- SUBCLINICAL NECROTIC ENTERITIS
- CLINICAL NECROTIC ENTERITIS
- CLOSTRIDIAL HEPATITIS



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Figure G-10.


**PATHOLOGY
 SUBCLINICAL
 NECROTIC ENTERITIS**

EUTHANATIZED BIRDS

(ANTERIOR) SMALL INTESTINE

MUCOSAL LESIONS

SMALL MEMBRANES AND ULCERS



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Figure G-11.


PATHOLOGY
CLINICAL
CLOSTRIDIAL ENTERITIS

BIRDS DEAD WITH THE DISEASE
(ENTIRE) SMALL INTESTINE (C/COLORECTUM, CAECAL TORSION)

MUCOSAL GUT LESIONS

DEHYDRATION

DARK LIVER



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Figure G-12.

PATHOLOGY
CLOSTRIDIAL HEPATITIS

USUALLY DETECTED AT SLAUGHTER

CHOLANGIOHEPATITIS
GALL BLADDER
BILE DUCTS
TERMINAL BILE TREE

MULTIFOCAL HEPATITIS




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Figure G-13.

NECROTIC ENTERITIS
ANTIBODY RESPONSE IN BROILERS
(SICLAND ET AL, UNPUBLISHED)

COMMERCIAL BROILER FLOCKS
POSITIVE CORRELATION LIVER LESIONS VS ANTIBODY RESPONSE
ANTIBODY RESPONSE IN FLOCK WITH CLINICAL NECROTIC ENTERITIS

SPONTANEOUS DISEASE MODEL (PEN TRIALS)
HIGHER FREQUENCY OF RESPONDERS THAN IN COMMERCIAL FLOCKS



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Figure G-14.

DIAGNOSING
CLOSTRIDIAL
NECROTIC ENTERITIS


CLINICAL DISEASE FORM

EXAMINE BIRDS DEAD WITH DISEASE

GROSS INTESTINAL LESIONS USUALLY DIAGNOSTIC

GUT SMEAR MICROSCOPY USEFUL

HISTOPATHOLOGY AND/OR BACTERIOLOGY
MAY BE INDICATED



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Figure G-15.

DIAGNOSING
CLOSTRIDIAL
NECROTIC ENTERITIS


SUBCLINICAL DISEASE FORM

RANDOM SAMPLING OF LIVE BIRDS

EXAMINE THE GUT IMMEDIATELY
FOLLOWING EUTHANASIA

GROSS INTESTINAL LESIONS DIAGNOSTIC

GUT SMEAR MICROSCOPY USEFUL



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Figure G-16.

MONITORING
CLOSTRIDIAL ENTERITIS

GUT SCORING

LIVER LESIONS AT SLAUGHTER

SEROLOGY: PROMISING ELISA



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Figure G-17.


DISEASE MODELS
CLOSTRIDIAL ENTERITIS

CHALLENGE MODELS

INDIVIDUAL ORAL INOCULATION (Paris, 1962)

INOCULATION OF FEED (Lang and Truscot, 1970)

INTRADUODENAL INFUSION
SURGICAL (Truscot and Al-Shakhly, 1977)
NON-SURGICAL (Wainman et al., 1994)



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Figure G-18.


DISEASE MODELS
CLOSTRIDIAL ENTERITIS

A MODEL BASED ON SPONTANEOUS INFECTION

ASSUMPTION:
THE ORGANISM IS ALWAYS THERE!

PREDISPOSING FACTORS
FEEDING, CHICKENS, PARENTS, ENVIRONMENT

DETECTION PROCEDURE
TIMING, SPECIFIC LESIONS

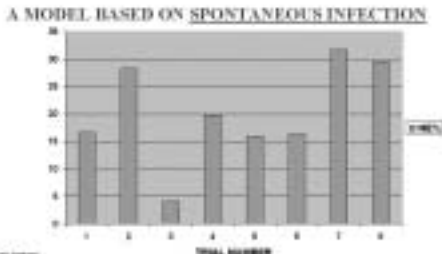


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Figure G-19.

DISEASE MODELS
CLOSTRIDIAL ENTERITIS

A MODEL BASED ON SPONTANEOUS INFECTION



Category	Total Number
1	15
2	35
3	5
4	20
5	15
6	15
7	35
8	35
9	15



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Figure G-20.

CONTROL
CLOSTRIDIAL ENTERITIS

IN-FEED ANTIBACTERIAL SUBSTANCES

NATURAL GUT FLORA PRODUCTS

VACCINATION

FEEDING

MANAGEMENT



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Figure G-21.

CONCLUSION
CLOSTRIDIAL ENTERITIS


CLOSTRIDIAL ENTERITIS CANNOT BE PREVENTED BY HYGIENE ALONE

SUBCLINICAL CLOSTRIDIAL ENTERITIS IMPAIRS PRODUCTIVITY

SUBCLINICAL ENTERITIS A DIAGNOSTIC CHALLENGE!

CLOSTRIDIAL ENTERITIS MONITORED BY BLOOD TESTING!

NO GOOD ALTERNATIVES TO IN-FEED SUBSTANCES YET



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