First report of *Clostridium difficile* in Austrian piglets with diarrhea



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Introduction

The objective of the study was to analyze different segments of the digestive tract and faeces of diarrheic juvenile pigs. The focus here is on bacterial results, see also Poster P75 (virologic findings).

Suckling and weaner pigs from 19 holdings in Styria (Fig.1) with severe problems of diarrhea were selected by veterinarians for analysis. One to three live piglets per holding (in total 55, Fig.2) and up to 6 faecal samples (in total 85) of other pigs were brought to the institute. Animals were euthanized and immediately processed for sampling. Samples of stomach, duodenum, jejunum, ielum, colon and rectum were obtained and prepared for bacteriological and histological analysis.

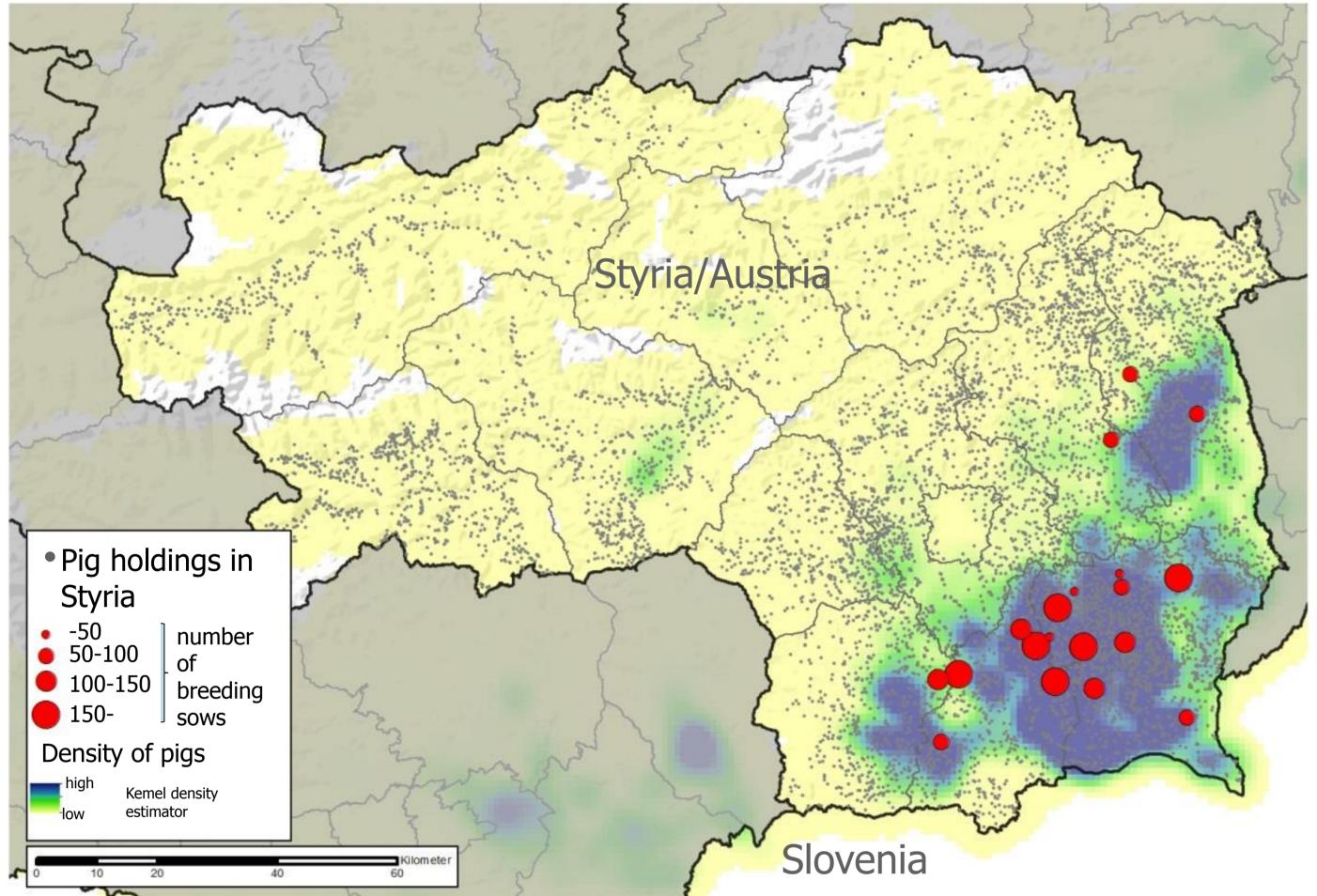


Fig.1: Samled pig holdings in Styria (in red)

Results

C. difficile was present in 5 holdings, 10 (18%) pigs and 10 (12%) swabs. Results of the subtyping are documented in table 1.

All isolated *C. perfringens* were carrying a toxin type A gen and were found in 18 holdings, 39 (71%) pigs and 61 (72%) swabs.

Hemolytic *E. coli* were detected in 13 holdings, 27 (49%) pigs and 29 (34%) swabs; non hemolytic *E. coli* were present in all holdings.

Brachyspira spp. were found in 7 holdings and 18 (33%) pigs, none of them was *B. hyodysenterieae* or *pilosicoli*, in two cases *B. intermedia / innocens* were identified.

5 Pigs (9%) of 3 holdings were infected with Eimeriidae, and 1 (2%) pig with *Trichuris suis*.

Histological analyses helped to clarify the etiological relevance of the detected microbials. Tissue lesions typical for bacterial infections were apical necrosis of intestinal villi, ulcerations, crypt abscesses, neutrophil granulocytes and detritus in the activated lymphoglandular complexes. Bacterial induced lesions were more often found in neonatal pigs.

Material and methods

Bacteriological examination of ileum, colon and faecal swabs included aerobic culture on COS agar (Biomerieux) and McConkey agar for detection of *Escherichia coli*, anaerobic culture on COS agar with Neomycin for detection of *Clostridium perfringens*, phase contrast microscopy for detection of *Brachysira spp.*, and direct anaerobic culture as well as culture after enrichment, with and without alcohol shock, on CLO agar (Biomerieux) to find *C. difficile*. *C. perfringens* and *C. difficile* were further typed with PCR toxin typing and ribotyping. Positive samples on *Brachyspira spp.* were further investigated via PCR (Adiavet TM Brachy, Adiagen).

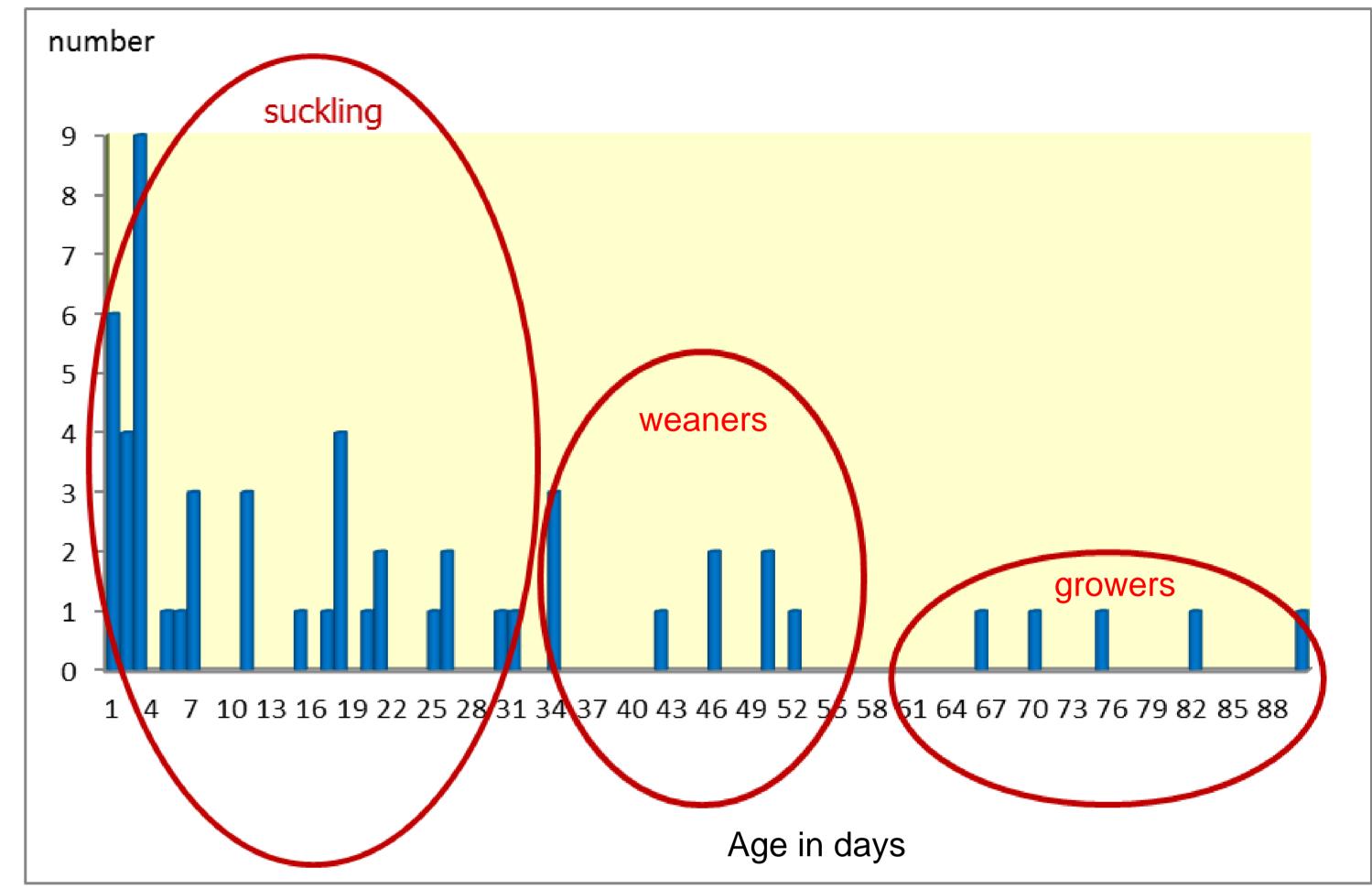


Fig.2: Age distribution of sampled pigs

Cl diff	1 s.i.	1 l.i.	2 s.i.	2 l.i.	3 s.i.	3 l.i.	4 s	5 s	6 s	7 s	8 s	9 s
Farm 2	neg	neg	neg	pos								
Ribotyp				Al-12								
F. 11	neg	neg	pos	neg	pos	neg	pos	neg	neg	neg	neg	neg
Ribotyp			RT005		Al-12		RT005					
F. 12	pos	pos	neg	neg	pos	neg	neg	neg	neg	pos	pos	neg
Ribotyp	R 078	AI-12			R078					R078	R078	
F. 16	neg	neg	neg	pos	neg	pos	neg	pos	neg	pos	neg	pos
Ribotyp				AI-12		598		241		AI-12		AI-12
F. 18	neg	pos	neg	pos	neg	pos	pos	pos	pos	pos		
Ribotyp		598		598		598	598	598	598	598		

Table 1: *C. difficile* results, abbrevations: Cl diff = *Clostridium difficile*, F. = holding, Ribotyp = ribosomale typing result, s.i. = small intestine, l.i. = large intestine, s = swab (n=85), n=85, n=85

Summary

Seven holdings showed bacterial infections with two or more type of bacteria, three holdings were additionally infected with parasites and most holdings had coinfections with viruses.

In humans *Cl. difficile* is known as nosocomial agent and causes diarrhea and pseudomembranous colitis.

The role of *C.difficile* in diarrhea in pigs is not yet fully revealed. In our findings there was no clear connection between isolation of this pathogen and histological findings.

