



Grethe.Venaas@agrsci.dk

# Fermentation combined with enzyme supplementation – A strategy to improve both nutritional value of rapeseed cake and gastrointestinal health of pigs?

G. V. Jakobsen, B. B. Jensen, K.E. Bach Knudsen and N. Canibe

Aarhus University, Foulum, Department of Animal Science, P.O. Box 50, 8830 Tjele, Denmark

## Introduction

Due to its antimicrobial properties, fermented liquid feed (FLF) has gained interest in relation to the search for alternatives to antimicrobial growth promoters. During fermentation, a proliferation of lactic acid bacteria occurs. This results in an increased lactic acid concentration, lower pH and inhibited growth of *Enterobacteriaceae*.

Rapeseed is an important oil- and protein crop in Denmark. Rapeseed cake (a co-product from the biofuel production) contains protein of high nutritional quality. However, its use in animal feed is limited due to the content of anti-nutrients such as glucosinolates, phytate and dietary fibre.

### AIM

To investigate different enzymes for their ability to improve the nutritional value of rapeseed cake during liquid fermentation and at the same time keeping the antimicrobial properties of fermented liquid feed.

## Materials and Methods

**In vitro study:** Nine enzymes constituting carbohydrases, phytases and their combinations were tested *in vitro* at doses recommended by the manufacturers (Danisco Genencor and Novozymes A/S). Rapeseed cake and water were mixed (1:5.5 w/w) in bioreactors and one of the enzymes or enzyme combinations to be tested were added. The mixtures were incubated at 30°C. Back-slopping was performed, where 50% of the mixture was removed and replaced with the same amount of fresh feed, water and enzyme. Sampling was performed during the initial 48 hours and again after backslopping, from 168 to 192 hours.



**In vivo study:** Eight castrated male pigs of approx. 30 kg were used. The pigs were cannulated at the *distal ileum* and after a recovery period assigned to four diets according to a double latin square design.

**Control:** non fermented liquid feed standard diet  
**n-FLF:** non fermented liquid feed experimental diet  
**FLF:** fermented liquid feed experimental diet  
**FLF+enz:** fermented liquid feed + glucanase-xylanase (13.5 g enzyme/kg DM)

The experimental diets contained 20 % rapeseed cake. After an 11 day adaption period *ileal* and *fecal* sampling was performed.

## Results

One enzyme combination (glucanase+xylanase) had the clearest effect on fibre degradation - 46 % and 16 % reduction of total NSP (non-starch polysaccharide) in the two phases of fermentation, compared to the control (P ≤ 0.002) (Figure 1).

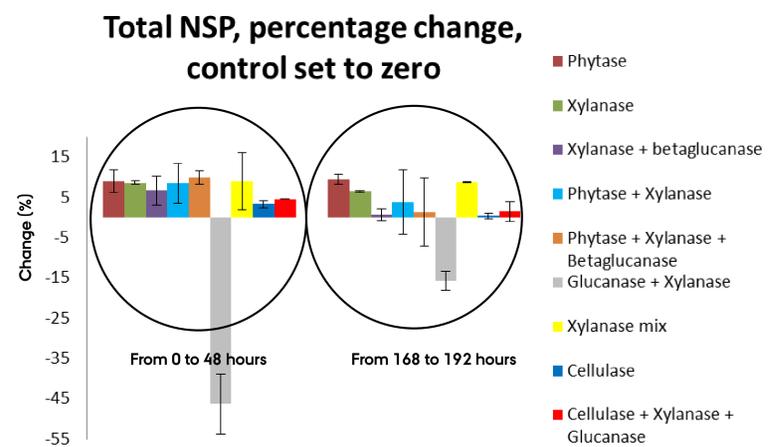


Fig.1. Change of total NSP (%) in the two phases of fermentation. Control set to zero.

Based on these findings the enzyme mixture glucanase + xylanase was chosen for an *in vivo* digestibility study on pigs. Results on microbial composition at *ileum* and in *feces* showed a tendency towards lower *Enterobacteriaceae* numbers at both sites (see table 1) of pigs fed FLF compared to those fed non-FLF. This indicated a beneficial effect of FLF on gastrointestinal health of the animals.

Table 1: Microbial counts (log cfu/g sample) in *ileum* and *fecal* samples from pigs fed four different diets. Superscripts in brackets tend to differ, with a p-value slightly above 0.05.

	Diet				SEM	P-value
	Control	n-FLF	FLF	FLF + enz		
<b>Ileum</b>						
<i>Enterobacteriaceae</i>	6.4 <sup>(a)</sup>	6.3 <sup>(a)</sup>	5.1 <sup>(b)</sup>	5.4 <sup>(b)</sup>	0.37	0.06
Lactic acid bacteria	8.8 <sup>a</sup>	8.3 <sup>a</sup>	7.5 <sup>b</sup>	7.7 <sup>b</sup>	0.19	<0.001
<b>Feces</b>						
<i>Enterobacteriaceae</i>	6.3 <sup>(a)</sup>	6.2 <sup>(a)</sup>	5.1 <sup>(b)</sup>	5.2 <sup>(b)</sup>	0.38	0.08
Lactic acid bacteria	9.2 <sup>a</sup>	9.4 <sup>a</sup>	8.5 <sup>b</sup>	8.4 <sup>b</sup>	0.17	0.001

Also a lower number of lactic acid bacteria was observed in the two fermented diets. This result has been obtained previously and is believed to be a result of a lower amount of substrate after the fermentation process.

## Conclusion

❖ The data from the *in vitro* screening study indicated that addition of enzymes to rapeseed cake during liquid fermentation can be a strategy to improve the nutritional quality of rapeseed cake to pigs. The mixture of glucanase + xylanase gave promising results.

❖ The microbial examination of *ilium* and *feces* from the *in vivo* digestibility study indicated that it is possible to keep the antimicrobial properties of fermented liquid feed using a rapeseed cake rich diet with enzyme addition.