

FROM COMMERCIAL RESULTS TO SCIENTIFIC EVIDENCE

Jens Legarth, CEO

HEALTHY ANIMALS ARE GOOD BUSINESS

Promoting health for animals and businesses.

Our fermented plant proteins modulate the gut microbiome of mothers and offspring.

Our high value proteins **increase earnings** for the farmer and reduce the need for antibiotics and ZnO.

Distributors in many countries.



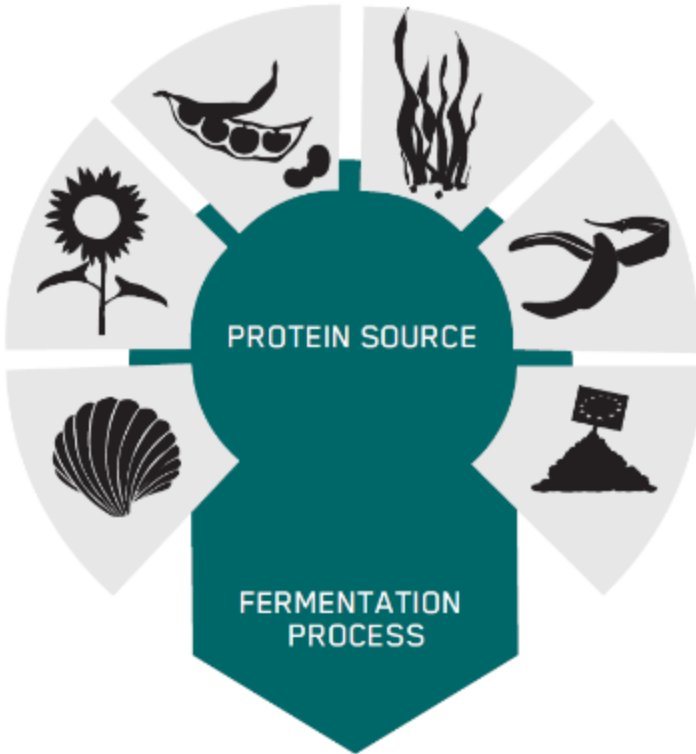
PROJECTS

Building bridges between science and the market

- Currently attending 10 projects (national/EU-level)
- Total budget of €36 million



ABOUT FERMENTATION

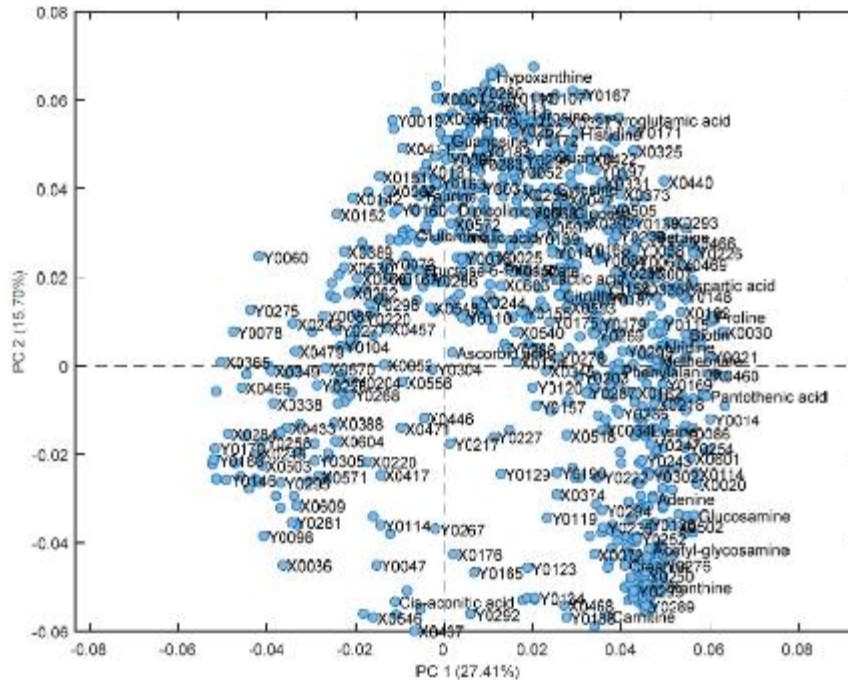


Designer protein

- › We can ferment most protein sources
- › We can convert indigestible sugar into lactic acid (5-10%)
- › Several advantages will pay for the drying cost
- › Higher protein, phosphorous, fibre and energy digestibility
- › Our dry products contain live bacteria and enzymes

ABOUT FERMENTATION

Compounds in fermented EP products

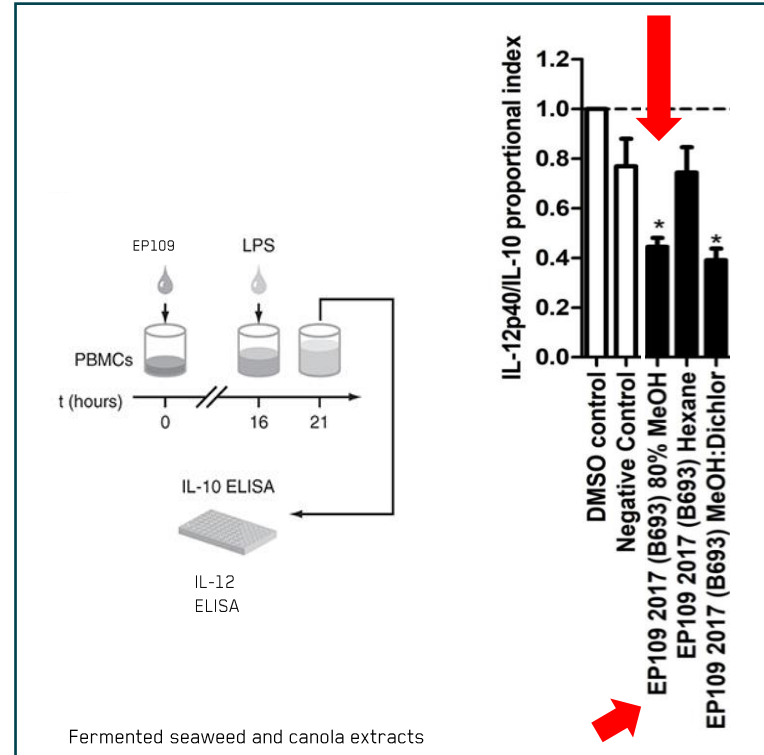


- Essential amino acids
- Polyunsaturated fatty acids
- Bioactive phenols
 - Anti-oxidants
 - Anti-microbial
 - Anti-inflammatory
- Vitamins from B-complex
- Phytate solubilization
 - Myo-inositol production
- Fiber solubilization
 - Sugars
 - Prebiotic oligosaccharides
- Lactic acid
- Bacterial biomarkers

IN VITRO ANTI-INFLAMMATORY EFFECT

Fermented seaweed and canola extracts

- Test of different fermented seaweed and canola extracts in peripheral blood mononuclear cells (PBMC) assay using 3 different human donors.
- Lipopolysaccharides (LPS) or endotoxins found of the outer membrane of Gram-negative bacteria was used to stimulate an inflammation in matured human PBMCs.
- Response was measured by inhibition of **IL12** proinflammatory cytokine and induction of anti-inflammatory cytokine **IL10** by extracts.



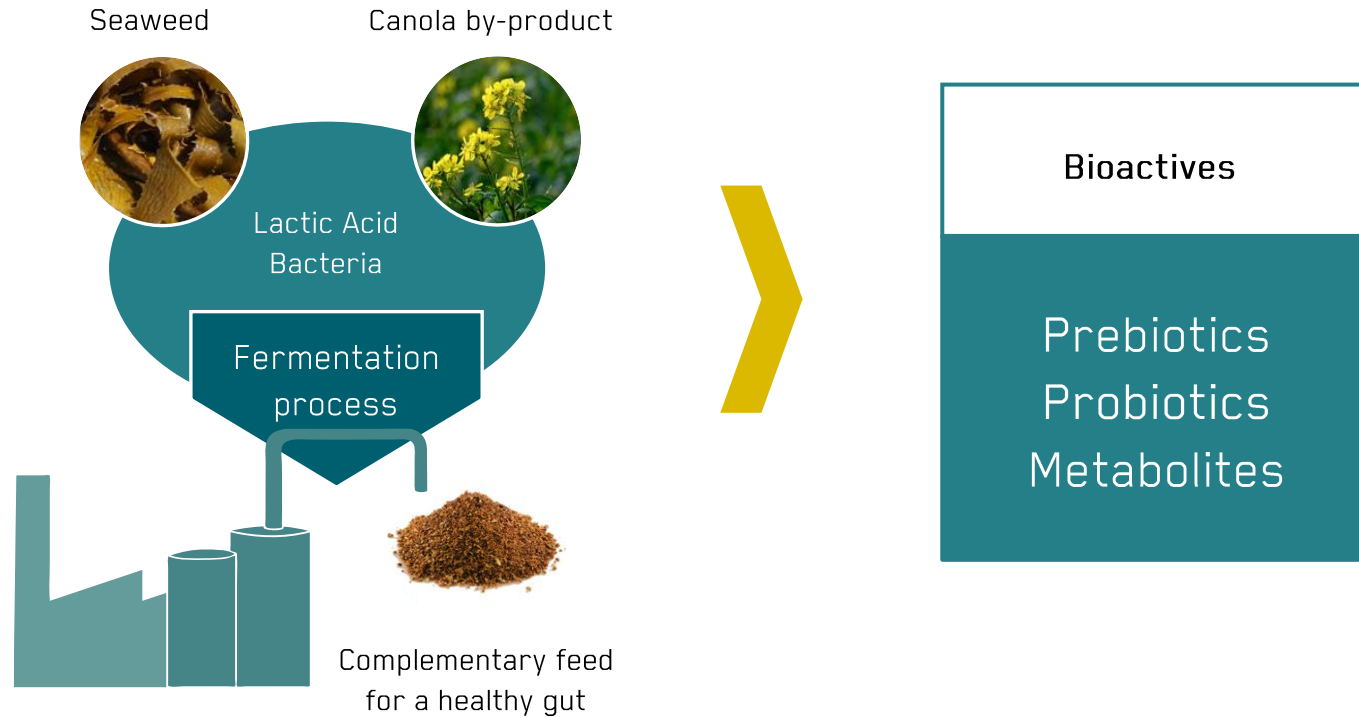
BACTERIA*

Table 2: Bacteria repression overview when using an EP-extract.

Strain	Type strain:	Concentration of bioactives (mg/mL) showing inhibition zone (indicated as +/-)			
EP-product	Pathogen	100 mg/mL	50 mg/mL	25 mg/mL	DMSO control
Gram positive					
Methicillin resistant <i>Staphylococcus aureus</i> (MRSA)	USA_300	-	-	+	++
Methicillin resistant <i>Staphylococcus aureus</i> (MRSA)	COL	-	-	-	++
<i>Staphylococcus carnosus</i>	15605T (CCUG)	-	-	-	++
<i>Clostridium perfringens</i>	19408 (CCUG)	-	-	-	++
<i>Streptococcus bovimastiditis</i>	69277T (CCUG)	-	-	++	++
Gram negative					
<i>Campylobacter jejuni</i>	29428 (CCUG)	-	-	-	++
<i>Campylobacter coli</i>	45147 (CCUG)	-	-	-	++
<i>E. coli</i>	11775 (CCUG)	-	-	-	++
<i>Vibrio parahaemolyticus</i>	27657 (DSM) / 67711 CCUG	-	-	+	++
<i>Salmonella enterica, supsp enterica</i>	46220 (CCUG)	-	++	++	++

- Symbol indicates no visible growth was observed in the MBC experiments.
- + Symbol indicates some visible growth and a small inhibition zone.
- ++ Symbols represents visible growth and no inhibition zone.

FERMENTATION & DRYING PROCESS

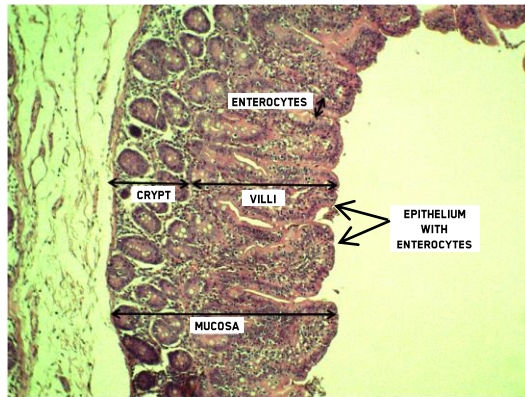


GUT DEVELOPMENT

Small intestine - Jejunum

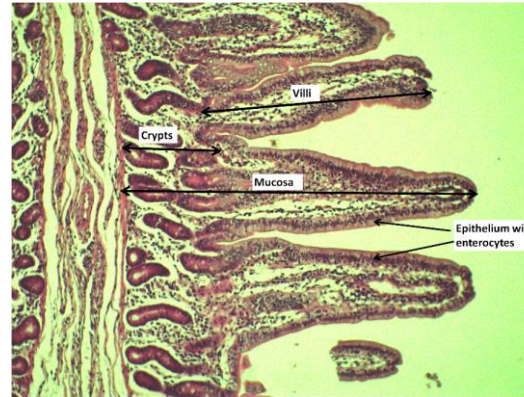
Magnification 10X – slide 2. *Mucosa including villi and crypts was analyzed under the microscope*

BASAL GROUP



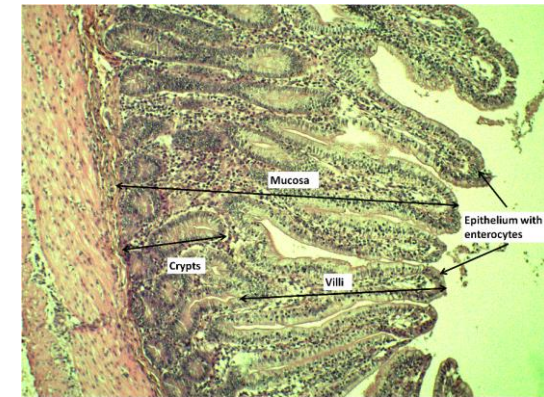
Thinner mucosa in jejunum in comparison with zinc and EP100i groups.
It is a sign of under- developed gut

ZINC GROUP



Loose mucosa in jejunum in comparison with basal and EP100i groups.
It is a sign of physiological stress, indicating a gut that is vulnerable to pathogen invasions and inflammation

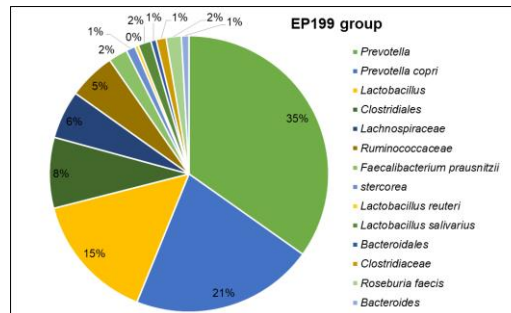
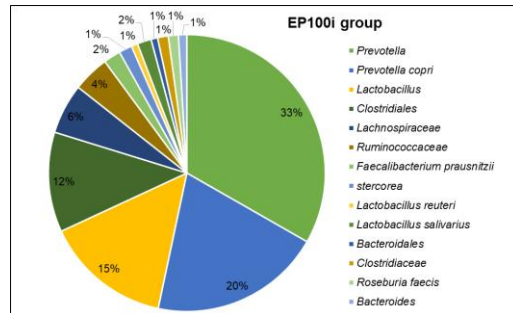
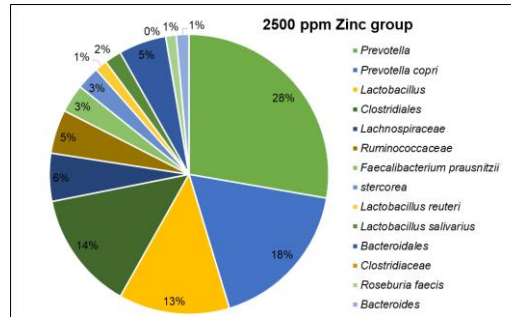
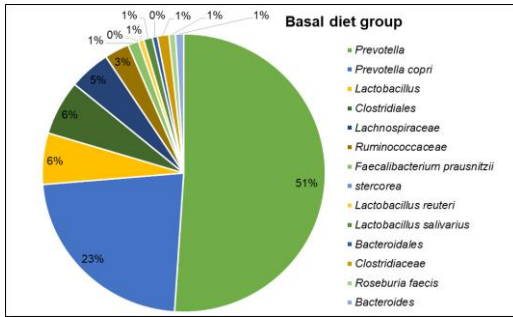
EP100i GROUP



Thick and packed mucosa in jejunum in comparison with basal and zinc groups.
It is a sign of a well-developed gut.

GUT DEVELOPMENT

Gut microbiome modulation. Increased bacterial evenness



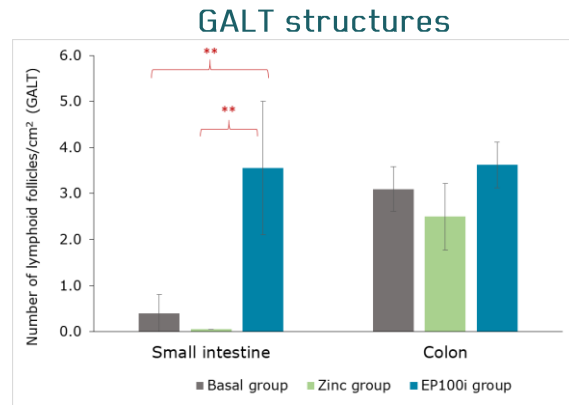
Increased number of recognized beneficial gut bacteria

	Basal diet	10 % EP199	10 % EP100i
<i>Lactobacillus spp.</i>	5.9	14.8	14.5
<i>Lachnospiraceae spp.</i>	4.7	5.6	5.7
<i>Ruminococcaceae spp.</i>	2.8	5.4	4.2
<i>Faecalibacterium prausnitzii</i>	1.2	2.2	2.0
<i>Lactobacillus salivarius</i>	1.0	1.5	1.6

Even dominance of several bacterial groups corresponds to improvement in animal health and performance as they improve gut-homeostasis

GUT DEVELOPMENT

Gut-immune system in small intestine and colon

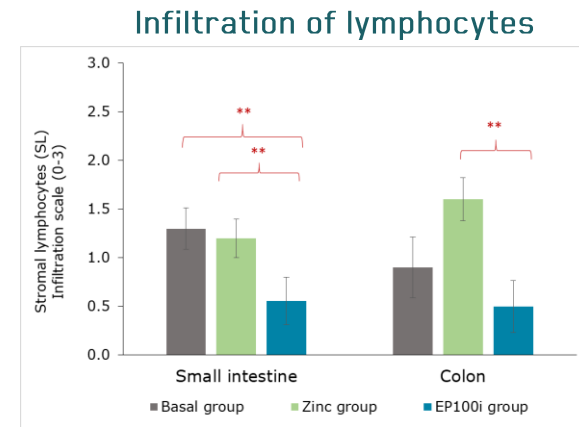
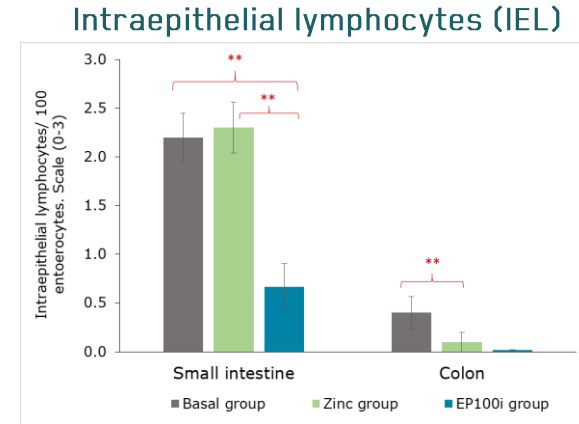


Gut-associated lymphoid tissue (GALT)

structures: The higher the number the more developed the immune system is

Intraepithelial lymphocytes (IEL) and infiltration of stromal lymphocytes (SL) are signs of inflammation:

The lower the number the less inflammation -> **Zinc: Early stage of inflammation**



PERFORMANCE

Impact of EP-products on weaner piglet performance

	TG1	TG2	TG4	
	Basal diet 2500 ppm zinc 10 % EP100i			
No. repetitions per TG	2	6	5	
Average pig/Pen	48	46	47	
Total no. pigs per TG	93	277	229	
Average weight at weaning (kg)	6,07	5,86	6,16	
				TG1:
ADWG (g)	272	285	304	P<0.05
AWG (kg)	17,1	18,0	19,2	
FI/pig (kg)	31,5	30,1	32,5	
FCR (kg feed:kg weight gain)	1,84	1,68	1,69	NS
Average body weight at D63 (kg)	23,0	23,9	25,5	
Average no. pig/pen at start of experiment	46,5	46,2	45,8	
Mortality (%)	12,0	5,2	4,1	NS

FEEDING CONCEPTS

Danish breeder and multiplier,
Søren Kjær Poulsen

HOW EP-PRODUCTS WORK



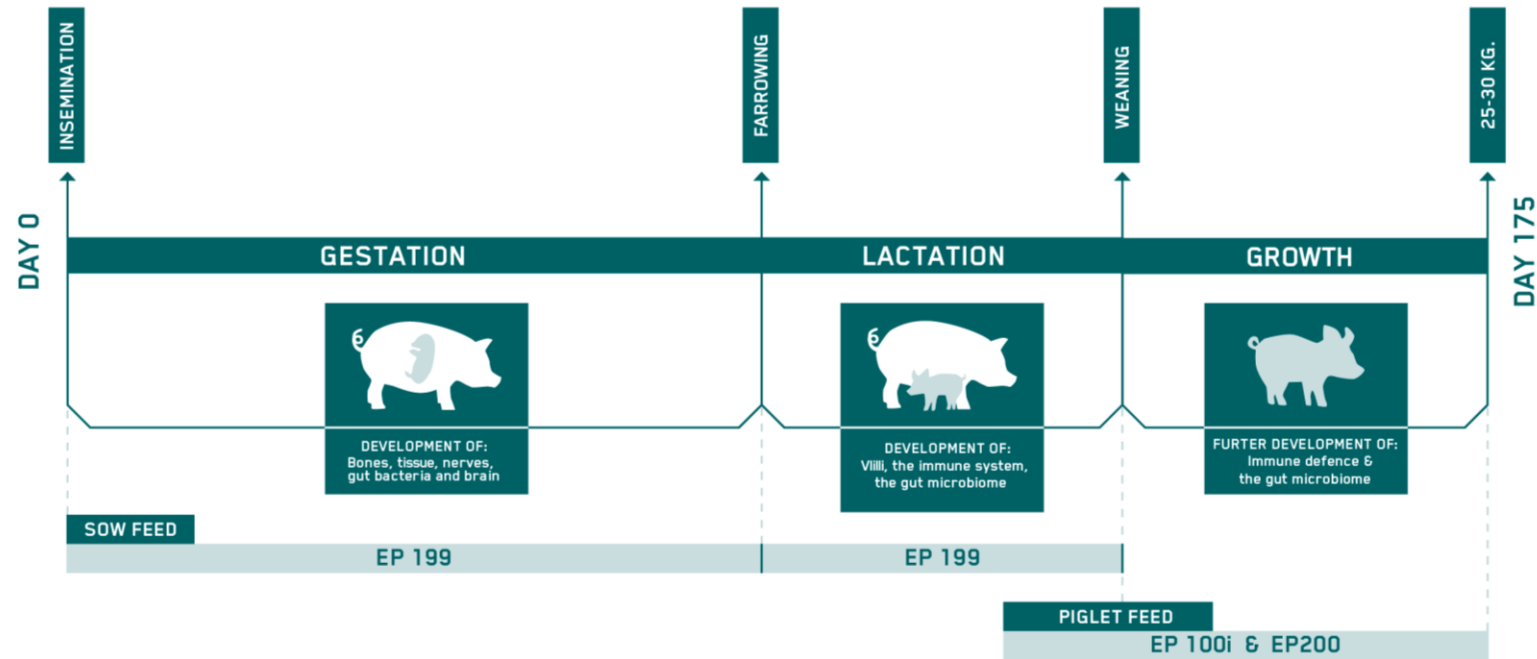
Try to improve overall health
through a single key

VS



Improve health by using
chords to create symbiotic
effects and harmony

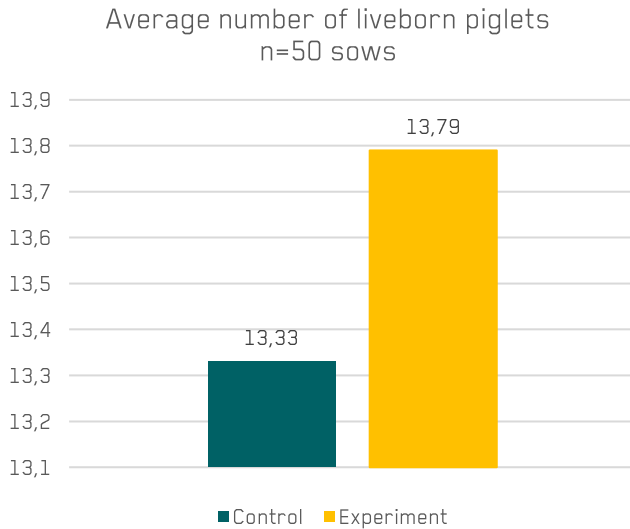
FEEDING CONCEPT: THE FIRST 175 DAYS



Research shows that up to 80 % of the piglet's DNA is founded during gestation and lactation.

COMMERCIAL FARM RESULTS

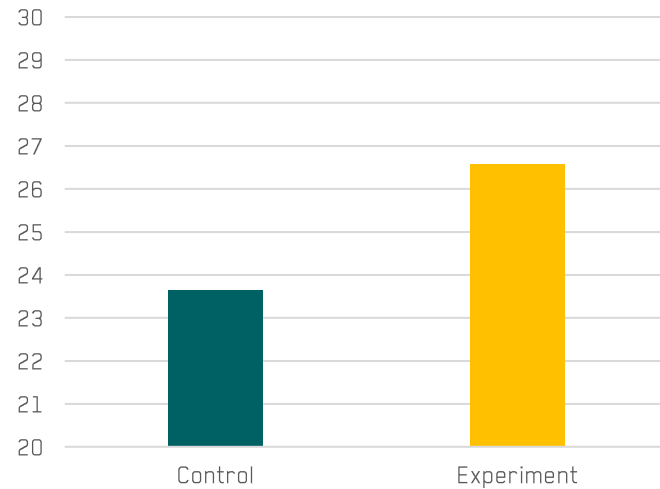
SOUTH AFRICA - TRIAL



On average, EP199-sows have 0,5 liveborn piglets more than the control group after just one cycle of EP199.

Below 20: Poor quality of colostrum
20-24: Borderline
25 to 29: Adequate
Over 30: Very good

Colostrum values (brix)

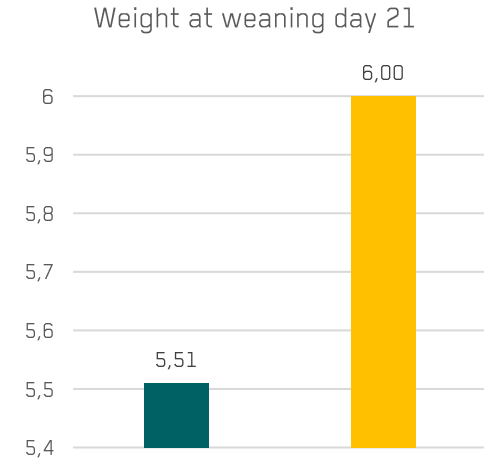
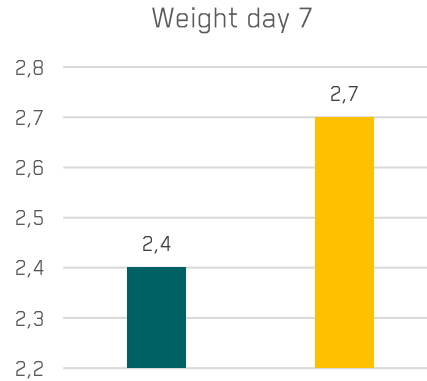
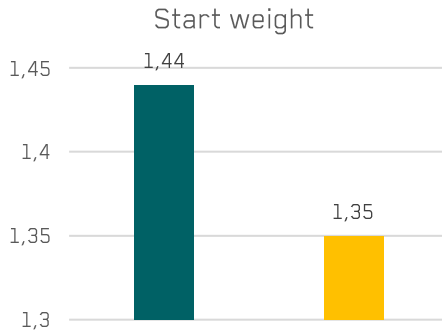


On average, EP199-sows increase the IgG content by 12% and moves the colostrum quality from a borderline quality to an adequate quality.

COMMERCIAL FARM RESULTS

SOUTH AFRICA - TRIAL

On average, EP199-sows increase piglet weight gain by more than 0,5 kilos at weaning even though the sows have half a piglet more to raise.



Control



EP199

Unpublished data. Distribution is prohibited.

MATERNAL FEEDING

Improved bone structure and mechanical properties of offspring₁

Bone properties of samples from knees of 77 days old piglet.

BASAL GEOMETRIC PROPERTIES	Unit	Group control	FFEED	P-value
Bone length	mm	143	156	0.006
Bone weight	g	149	156	0.036
GROWTH PLATE CARTILAGE	Unit	Group control	FFEED	P-value
Total cartilage thickness	µm	1369	2100	<0.001
ARTICULAR CARTILAGE	Unit	Group control	FFEED	P-value
Total cartilage thickness	µm	2067	3640	<0.001
STRUCTURAL PROPERTIES	Unit	Group control	FFEED	P-value
Yield load	kN	1.64	1.88	0.012
Ultimate load	kN	1.91	2.26	0.038
Stiffness	N/mm	830	1067	0.021
Elastic energy	J	1.74	1.81	0.022
Work to fracture	J	2.87	3.6	0.032
Bending moment	Nm	23.4	29.9	0.002

53%

76%

E. Tomaszewska et al., Dried fermented post-extraction rapeseed meal given to sows as an alternative protein source for soybean meal during pregnancy improves bone development of their offspring, livestock Science, Volume 224, June 2019, Pages 60-68.
<https://www.sciencedirect.com/science/article/abs/pii/S1871141319305165?via%3Dihub>

MATERNAL FEEDING

Influence on biochemical parameters in gilt and piglet blood

BLOOD	UNITS	CONTROL	FFEEED	P
LATE PREGNANCY n=6				
Haematocrit	%	38.77	41.12	P<0.05
Haemoglobin	mmol l ⁻¹	7.01	7.88	P<0.05
Red blood cells	10 ¹²	6.91	8.01	P<0.05
LATE LACTATION n=6				
Haematocrit	%	36.23	39.17	P<0.05
Haemoglobin	mmol l ⁻¹	4.56	5.98	P<0.05
Red blood cells	10 ¹²	6.23	6.96	P<0.05
OFFSPRING				
Haematocrit	%	36.24	39.96	P<0.05
Haemoglobin	mmol l ⁻¹	6.92	7.89	P<0.05
Red blood cells	10 ¹²	6.03	6.76	P<0.05

← 16% G

← 12% G

← 12% G

Iron binding
capacity increase
of more than 30%
for piglets

MATERNAL FEEDING

MINERALS IN BLOOD	GILTS				SOWS		
	Units	Control	FFEED	P	Control	FFEED	P
LATE PREGNANCY n=6							
Phosphorus	mmol l ⁻¹	1.42	1.70	P<0.05	1.38	1.88	P<0.01
Copper	μmol l ⁻¹	26.19	32.47	P<0.05	23.37	28.09	P<0.01
Iron	μmol l ⁻¹	15.38	21.06	P<0.05	13.88	19.18	P<0.01
LATE LACTATION n=6							
Phosphorus	mmol l ⁻¹	1.87	2.31	P<0.05	1.76	2.24	P<0.01
Magnesium	mmol l ⁻¹	1.04	1.27	P<0.05	1.03	1.28	P<0.01
Copper	μmol l ⁻¹	12.66	16.15	P<0.05	14.65	18.31	P<0.01
Iron	μmol l ⁻¹	18.78	25.43	P<0.05	23.54	29.17	P<0.01
Zinc	μmol l ⁻¹				14.18	17.78	P<0.01
OFFSPRING n=12							
Phosphorus	mmol l ⁻¹	1.97	2.42	P<0.05			
Calcium	μmol l ⁻¹	2.58	3.12	P<0.05	2.53	3.04	P<0.01
Copper	μmol l ⁻¹	20.66	26.91	P<0.05			
Zinc	μmol l ⁻¹	7.87	10.29	P<0.05			
Iron	μmol l ⁻¹	27.81	35.33	P<0.05	28.02	37.51	P<0.01

20% G, 36% S

37% G, 38% S

24% G, 27% S

35% G, 24% S

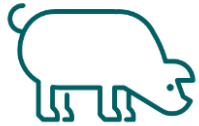
23% G

27% G, 34% S

Anna Czech et al., Biochemical and hematological blood parameters of sows and piglets fed a diet with a dried fermented rapeseed meal, Annals of Animal Science, 2019, DOI:10.2478/aoas-2019-0079. <https://sciendo.com/article/10.2478/aoas-2019-0079>

PRODUCTION RESULTS

INCREASE IN HEALTH, REPRODUCTION AND PRODUCTIVITY



2-3 PIGLETS MORE
WEANED PER SOW
PER YEAR



1,2 MORE
LIVEBORNS per
litter, uniform and
vital piglets.



2 PERCENTAGE
POINTS LOWER
MORTALITY rate and
less diarrhoea.



INCREASED MILKING ability
means **360 GRAMS MORE**
weight per piglet before
weaning.



12% LESS SOW FEED
PER PRODUCED
PIGLET



50 - 100 EUR MORE PROFIT
PER SOW PER YEAR

Porcine veterinarian preparing to
take DNA-fecal samples



DOCUMENTING THE FEEDING CONCEPT

GUT HEALTH DATABASE

GUT HEALTH DATABASE

Test before and after feeding with EP199

FAECES DNA-TEST

Together with Copenhagen University we do 16S RNA gene tests of bacteria in the gut microflora to map changes before and after supplementing.

BLOOD TEST

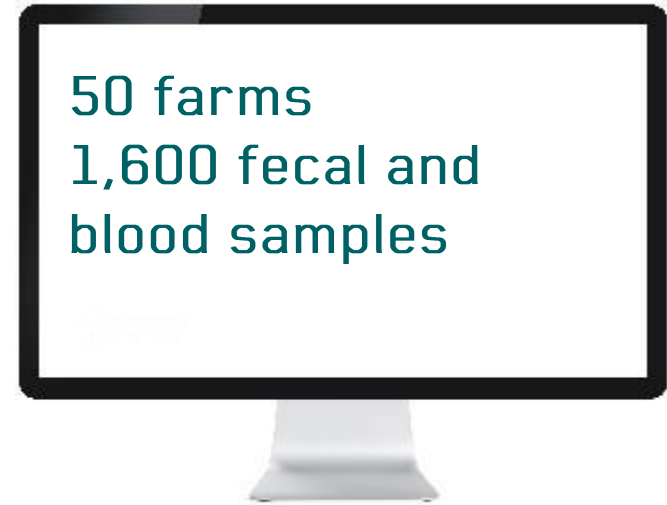
Test of biomarkers for inflammation before and after feeding

MILK TEST

Test of antibodies in colostrum before and after feeding

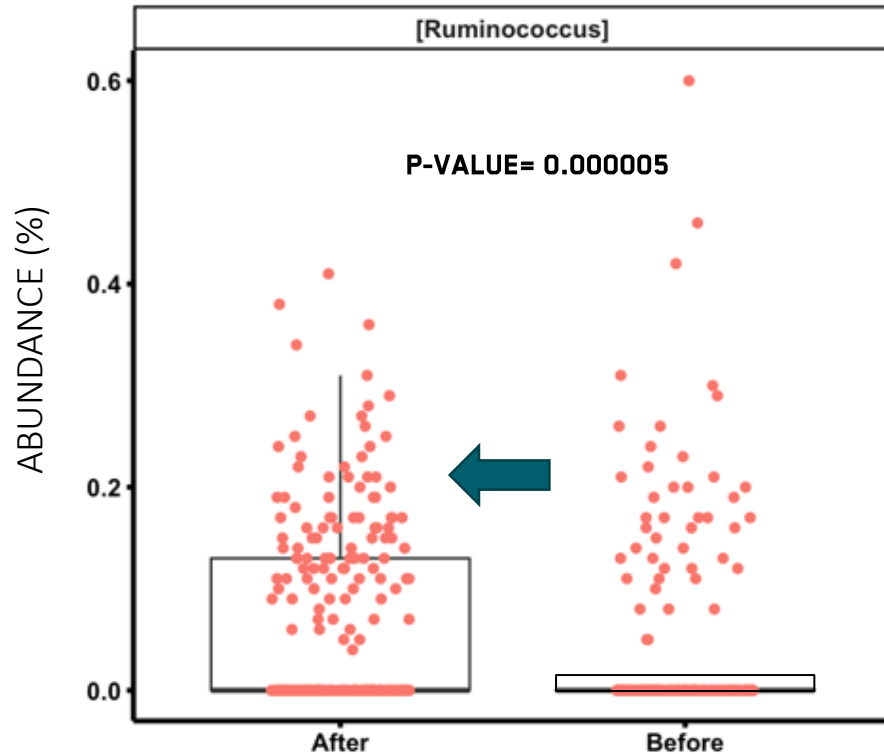
DATA

E-control, optimization, use of medication (antibiotics, zinc)



PROMOTION OF BENEFICIAL BACTERIA

19 FARMS, 456 SOWS, 608 SAMPLES



Significantly more SCFA-producing and/or fiber-degrading bacteria detected in high-reproductive performance farms.

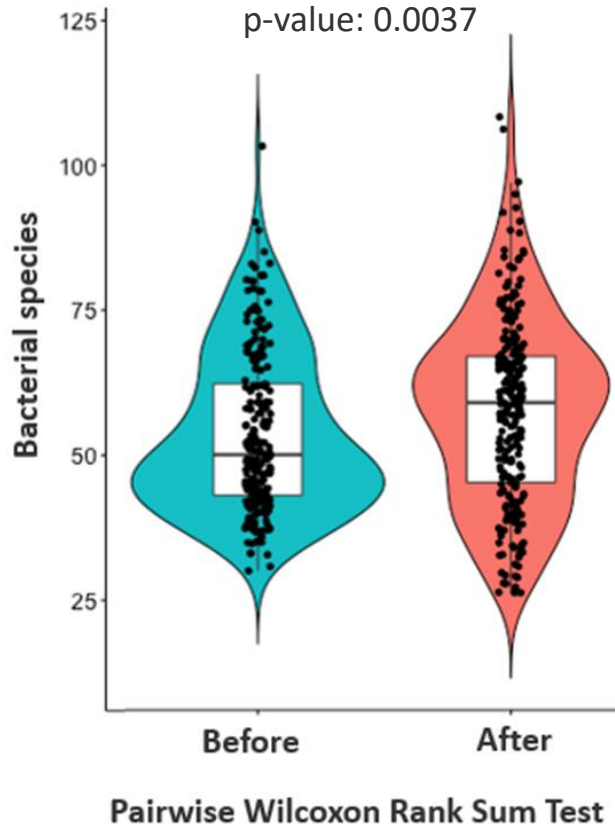
Increase in Ruminococcus correlates positively with:

- Farrowing performance
- Body weight
- Daily weight gain

GUT MICROFLORA DIVERSITY

19 farms, 456 sows, 608 samples

Alpha Diversity- All Farms



Significantly increased bacteria diversity (red) 20% increase.

Start of diseases is associated with gut dysbiosis and loss of microbial diversity in the gut microbiota.

Intervention to increase bacterial diversity would result in restoring gut dysbiosis and increasing health

Copenhagen University, March 2021: Comparing 16s rRNA faecal samples from 456 sows/gilts/young females before and after supplementing the fermented rapeseed and seaweed-protein EP199.



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