44. Immune modulating influence of Echinacea purpurea as an intermittent feed additive in layers and fattening pigs (Der Einfluss eines intermittierenden Einsatzes von Echinacea purpurea als Futterzusatz auf verschiedene Immunparameter von Legehennen und Mastschweinen). Barbara M. Böhmer*, H. Salisch, Brigitte R. Paulicks and F. X. Roth – Freising-Weihenstephan/Poing

Echinacea is used widespread as an herbal paramunity-inducer to reduce various infections in humans. The recommended application is a prophylactic use for several weeks. On the other hand, it was shown that e.g. phagocytosis is depressed by an intake duration exceeding 5 days. The aim of the present trials was to test if a repeated short-time application of Echinacea juice as feed additive is sufficient to stimulate the immune response of layers and fattening pigs.
Methods: In a trial with 72 white layers (LSL) two Echinacea juices (Berghof-Kräuter, Heilsbronn) as feed additives were tested for their influence on feed intake, performance, blood count, phagocytosis of granulocytes, and antibody titer (ND, Newcastle Disease) following vaccination. The dosage of the juice was adjusted on the basis of recommendations for humans at 0.25 ml/kg $BW^{0.75}$. Echinacea juice was pressed juiced of aerial parts of the plant preserved with either ethanol (20%) or by fermentation. The contents of cichory acid and alkamids in the ethanolic and fermented juice were analyzed at 2.1, 1.9 mg/ml, and 27.7, 2.7 µg/ml, respectively. The application scheme followed a repeated 14 days regimen with either 5 days application and 9 days without application or 2 days application and 12 days without application. Additionally, blood count and phagocytosis of granulocytes (luminescence assay) were determined in fattening pigs (80–100 kg) after Echinacea application (2 x 5 days). Blood was taken immediately following an application phase.
Results: Feed intake and performance of the hens were not impacted. Significant differences were found in the number of lymphocytes, the phagocytosis, and the antibody titer. The number of lymphocytes was highest in the group with 5 days ethanolic juice. Phagocytosis was reduced in both groups receiving ethanolic juice for 2 and 5 days. The ND antibody titers (1 (a) and 2 (b) weeks after vaccination) were highest in the groups receiving the fermented juice for 2 days. The phagocytosis in the fattening pigs showed a significant increase in both supplemented groups. The number of lymphocytes was also increased significantly in the groups provided with Echinacea.

Table: Effect of Echinacea supplementation on selected blood parameters of layers and fattening pigs

	Form of juice	**0**	**Ethanolic**	**Fermented**	**Ethanolic**	**Fermented**
	Duration (d)	**0**	**5**	**5**	**2**	**2**
Layers[1]	Lymphocytes	12165^{b}	14639^{a}	12320^{b}	10717^{b}	10435^{b}
	[2]ND titer (a)	428^{c}	525^{a}	520^{b}	486^{b}	527^{a}
	[2]ND titer (b)	488^{b}	465^{bc}	476^{bc}	454^{c}	533^{a}
	Luminescence (cpm)	0.42^{a}	0.26^{c}	0.37^{abc}	0.27^{bc}	0.39^{ab}
Pigs[3]	Lymphocytes	12570^{b}	15275^{a}	11218^{b}		
	Luminescence (cpm)	1.5^{b}	2.1^{a}	2.1^{a}		

$^{a\,b}$ significance according to [1]Duncan-test, [2]Wilcoxon and Wilcox-test, or [3]Student-Newman-Keuls-test (($P<0.05$)

Conclusion: It can be concluded, that a repeated short-time application of Echinacea juice as feed additive has effects on several immune parameters. The 2 days duration showed almost the same effects as the 5 days duration and seems to be sufficient for stimulation. The different effects of ethanolic and fermented juice are probably caused by the different concentration of the ingredients.

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45. Effects of alimentary tannin supplementations on nutrient digestibility and growth performance of weanling piglets (Einflüsse alimentärer Tanninzulagen auf Nährstoffverdaulichkeit und Wachstumsleistung von Absetzferkeln). Brigitte R. Paulicks*, G. Biagi and F. X. Roth – Freising-Weihenstephan/Bologna

Tannins are natural polyphenolic compounds with well-known antibacterial and antidiarrhoic properties favouring their inclusion in the feed of young piglets, which often suffer of digestion problems after weaning. On the other hand, it has also been demonstrated, particularly for feed-bound tannins, that they can bind nutrients and digestive enzymes, thus reducing the digestibility of the diet. In the present study the effects of tannin extract supplementations on the nutrient digestibility and the growth performance of weaned piglets were determined.

Methods: Forty-eight weaned piglets (Pit x DL; 8.2 kg BW) were divided into 4 groups (12 animals per group, housed in individual cages) for a 28 d trial. Treatments were a commercial diet (30 % corn, 27 % wheat, 17 % barley, 16 % soybean meal) with a) no addition (F0) or with b) 0.15% (F0.15), c) 0.30% (F0.30), or d) 0.60% (F0.60) of a chestnut wood extract containing 75% tannins (Farmatan®, Tanin Sevnica, Slovenia). Feed and water were provided ad libitum. Animal weights and feed consumption were recorded weekly. Apparent fecal nutrient digestibility was determined using marker technique (HCl-unsoluble ash) by collecting the feces for five days during the forth experimental week.

Results: Body weight, daily weight gain, and daily feed comsumption showed no significant differences between treatments averaging 19.8 kg, 406 g/d, and 565 g/d, resp. However, piglets of group F0.60 had about 5 % higher body weight, 13 % higher weight gain and 7 % higher feed consumption than piglets of group F0. Feed conversion ratio was significantly improved for F0.60 compared with the other treatments (1.32 versus 1.39). Digestibility of organic matter, crude protein, and N-free extracts decreased from 87.1, 83.7, and 92.5 %, resp. (F0) to 85.7, 80.7, and 91.6 %, resp. (means of F0.15, F0.30, and F0.60; $P < 0.01$). In contrast, digestibility of fiber, ether extracts, and phosphorus increased significantly ($P < 0.01$) with tannin supplementation from 46.2, 71.6, and 43.5 %, resp. (F0) to 49.1, 74.5, and 47.5 %, resp. (F0.60). There was no clear dose-response effect.

Table: Growth performance and digestibility of selected nutrients with graded tannin supplementation

Treatment	F0	F0.15	F0.30	F0.60	P <
Final body weight (kg)	19.3±2.3	19.4±2.2	19.7±2.4	20.7±2.5	n.s.
Weight gain (g/d)	395±47	400±55	410±75	445±58	n.s.
Feed consumption (g/d)	549±65	553±77	570±101	588±83	n.s.
FCR (kg feed/kg gain)	1.39^a±0.06	1.38^a±0.08	1.39^a±0.07	1.32^b±0.05	0.05
DOM (%)	87.1^a±1.0	85.3^b±0.9	85.8^b±1.6	86.1^b±1.2	0.01
DXP (%)	83.7^a±1.6	80.9^b±1.9	81.0^b±3.2	80.2^b±2.7	0.01
DP (%)	43.5^b±3.0	41.1^b±5.2	44.0^{ab}±5.3	47.5^a±4.1	0.01

^{ab} means with different superscripts differ significantly ($P < 0.05$; Duncan-test); n.s. $P > 0.1$

Conclusion: The present results show that tannins had depressive effects on the digestibility of several nutrients. However, growth performance and feed consumption were not impaired, and FCR was even improved by the highest tannin supplementation. As there were no cases of diarrhoea, no influence of tannins on health could be observed.

This study was supported by BAT (Bayerische Arbeitsgemeinschaft Tierernährung e.V., Freising-Weihenstephan).

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46. Effect of insoluble dietary fiber on performance, microbiological and morphological parameters and mRNA expression of inflammatory and cell cycle marker genes in the gut of weaning piglets used as animal model (Effekte von verschiedenen unlöslichen Ballaststoffen auf die zootechnischen Leistungen, mikrobiologische und morphologische Parameter, sowie auf die mRNA-Expression von inflammatorischen und Zellzyklus Markergenen im Verdauungstrakt des Modelltieres Absetzferkel). K. Schedle* and W. Windisch – Vienna

"Dietary fiber" comprises a variety of heterogeneous substances. Basically, total dietary fiber can be divided in a soluble and an insoluble fraction affecting functionality of the gastrointestinal tract including its microbial ecosystem and involved tissues in a different way. In the present study we investigated the effect of two insoluble dietary fiber sources differing in lignin contents, on performance and gastrointestinal physiology in weaning piglets used as animal model.

Methods: The study employed a total of 48 weaning piglets fed *ad libitum* a weaning diet (15.4 MJ ME/kg, 21.5% CP) during the first 9 days and then a starter diet (14.4 MJ ME/kg, 18.2% CP). Total dietary fiber content (TDF) was 14.6% and 13.3%, respectively. The two diets were modified by adding wheat bran or pollen from Chinese Masson pine (*pinus massoniana*). The pine pollen was used as representative of largely indigestible fiber with lignin as dominant fraction. The fiber sources were added to the diets as follows: (a) no addition (control), (b) 3.0% wheat bran, (c) 1.3% pine pollen, (d) 2.6% pine pollen. The amounts of added fiber sources were adjusted on their TDF contents and resulted in 16, 8, and 16 g added TDF per kg feed for diets b) to d), respectively. Animals were distributed according to litter, sex and initial body weight (approx. 8.5 kg) among the 4 diets a) to d) and were housed in one room with 8 pens (6 animals per pen, 2 pens per diet). Feed and water was provided *ad libitum* for 5 weeks. Individual body weight and pen feed intake was recorded. During the 5^{th} experimental week, individual fecal samples were collected. At days 36 and 37, each half of the animals were professionally slaughtered without starving in order to obtain samples of gut contents from ileum and colon for microbiological investigations. Tissue samples of stomach, jejunum, ileum, colon and mesenteric lymph nodes were collected and snap frozen for subsequent real time RT-PCR analysis. Another part of these samples was washed in sodium-chloride solution, stored in formalin until imbedding for morphological analysis.

Results: Insoluble fiber improved feed intake up to 42% and weight gain up to 23% but also raised feed conversion ratio. In chime of terminal ileum, fiber additions reduced colony counts of *E. coli* and Enterobacteriaceae. In the colon, ammonia contents were reduced by added insoluble fiber (-38%) as well as apparent digestibility of crude protein up to -5.5%. Both fiber sources increased villi length in jejunum (+10% on average) and ileum (+16% on average). Lymph follicle density in mesenterial lymph nodes tended to be increased by wheat bran but not by pine pollen. Wheat bran induced an up-regulation of NFkB in stomach and jejunum, as well as TNF alpha and TGF beta, and Caspase 3 in jejunum. No changes were observed in the ileum. Pine pollen induced down-regulation of NFkB, TNF alpha, TGF beta, Caspase 3, CDK 4 and IGF 1 in the colon as well as up-regulation of NFkB and TGF beta in mesenterial lymph nodes.

Conclusion: Results suggest that insoluble fiber may significantly affect microbiology, morphology and cell activity (proliferation, inflammation, cell cycle) along the entire digestive tract, depending on the nature of the fiber source. In total, additions of insoluble dietary fiber may significantly contribute to gastrointestinal stability and eubiosis with further benefits on overall body performance of weaning piglets.

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47. Influence of different application procedures of a probiotic *E. faecium* strain (direct oral dosing vs. supplementation in feed) on bacterial parameter in piglets (Vergleich verschiedener Applikationsarten eines probiotischen *E. faecium* Stamms (orale Dosierung vs. Futtersupplementierung) auf bakterielle Parameter in Ferkeln). Irina Gordeeva, O. Simon and W. Vahjen* – Berlin

Probiotics are supplemented to animal feeds in order to modify intestinal bacteria resulting in a beneficial outcome for the host animal. One probiotic mode of action is considered to be the interaction of probiotic cells with intestinal bacteria. This requires actively growing cells in adequate numbers in order to have an influence on the multitude of already existing bacterial interactions in the intestine. Probiotics are generally applied at concentrations of 10^9 viable cells/kg feed, but has been used much less frequently in drinking water. This study compares the probiotic application via feed (including supplemented mother sow feed) against the early, massive direct oral dosing of the probiotic strain.

Methods: In the direct dosing trial, one day old piglets of the trial groups were dosed daily with 10^9 viable cells of the probiotic strain *E. faecium* NCIMB10415 until the 31^{st} day of life. Piglets had access to a pre-starter diet supplemented with the probiotic strain (10^9 viable cells/ kg) from day 14 until weaning on day 28. After weaning, animals of the trial group were fed with a probiotic supplemented starter diet (10^9 viable cells/ kg). In the feeding trial, the mother sow feed was supplemented with the probiotic strain (10^9 viable cells/ kg) at the time of insemination and throughout the trial. Both were compared to control animals without probiotic. All piglets had access to a pre-starter diet from day 14 until weaning on day 28; after weaning, animals of the trial groups were fed with a probiotic supplemented starter diet (10^9 viable cells/ kg). In both trials, selected bacterial groups and species were monitored by real time PCR assays in samples from the stomach, jejunum and colon asc. Bacterial diversity in the intestine was recorded with a Denaturing Gradient Gel Electrophoresis assay using an eubacterial primer combination. Lactate was determined with an enzymatic assay, volatile fatty acids were determined by gas chromatography.

Results:

Despite the massive probiotic dosage of 10^9 cells per day, only minor differences in probiotic cell counts were noted between application procedures. Only on day 14 significantly increased probiotic cell numbers in the colon occurred in animals that were given the probiotic by direct oral dosage. However, compared to feed supplementation, direct oral dosing during the suckling period led to much higher ratios of the probiotic Enterococcus-strain within the total enterococci populations. Both trials showed no influence on lactobacilli, but *Escherichia* spp. were reduced in both trials. Compared to their respective controls, direct oral dosing led to relatively higher volatile fattty acid concentrations before weaning, but lower or similar amounts after weaning. The characterisation of bacterial diversity in the colon by DGGE analysis showed no lasting effect on species amount or diversity, but an increased similarity of bacterial populations among probiotic treated animals was noted. This effect was more pronounced throughout the suckling period in direct oral dosed animals.

Conclusions: This comparison showed that the probiotic strain reached a maximum colonization density of around 10^4 – 10^5 cells/g wet weight regardless of application. The increased percentage of the probiotic among all enterococci in direct oral dosed animals indicates a displacement of other enterococcal strains. Both applications procedures modified bacterial parameters in the intestine compared to their respective controls. The direct oral application showed increased bacterial metabolism compared to feed supplementation only during the suckling period.

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48. Effects of including organic acids or potassium diformate and grinding intensity (coarse/fine) of ingredients in the diet of weaned piglets experimentally infected with *E. coli* or *S. Derby* (Effekte von organischen Säuren oder Kaliumdiformiat im Futter sowie des Vermahlungsgrades (grob/fein) auf Keimgehalte im Chymus experimentell mit *E. coli* oder *S.* Derby infizierter Absetzferkel). Venja Taube*, Marion Neu, Yasmin Hassan, Jutta Verspohl and J. Kamphues – Hannover

Introduction: *E. coli* is responsible for high economic losses in pig production by causing fatal diseases whereas *Salmonella* is of interest as a zoonotic pathogen under aspects of food safety. The **aim** of the experiment was to examine whether and to what extent the addition of free acids (formic and propionic acid, fpa) or potassium diformate (pdf) and the structure of feed might affect the passage of orally applied *E. coli* and *Salmonella*, i. e. the efficacy of the stomach barrier function.

Methods: The experiments have been carried out with 80 pigs allotted into 4 groups in 4 consecutive trials (each trial: n = 20). Pigs were fed ad libitum, all diets were offered as pellets. Cereals (74 %) in the control diet were finely ground (2 mm screen; after pelleting: 5.3 % > 1.4 mm; 53.9 % < 0.4 mm), in test diets 0.9 % organic acids (75 % formic and 25 % propionic acid) or pdf (1.2 % in the diet) were used; in a 4th group (without organic acids) the diet was based on coarsely ground cereals (6 mm screen; after pelleting: 18.9 % > 1.4 mm; 48.1 % < 0.4 mm). 4-5 hours after oral application of 10 ml of a bouillon containing *E. coli* (7.8×10^8 to 7.1×10^{10} CFU/ml) or *S.* Derby (1.5×10^8 to 3.3×10^9 CFU/ml) pigs were sacrificed (after 35 days of experimental period; BW: 35.9 ± 8.2 kg) and samples of chyme from stomach, small intestine and caecum were obtained. Counts of *Salmonella* and *E. coli* were determined with classical culture methods.

Results: Adding free organic acids or pdf to the diet resulted in significantly lower counts of *E. coli* in the stomach after oral application 4-5 h before. Lower counts of *Salmonella* were found in the stomach and also in the small intestine and the caecum. Comparing the fine and the coarse diet (both without acids) the counts of applied bacteria in the chyme did not differ significantly.

diet	*E. coli* (CFU/g)			*Salmonella* (CFU/g)		
	stomach	small intestine	caecum	stomach	small intestine	caecum
fine	**3.8×10^7** *± 6.9×10^7*	**7.4×10^8** *± 8.5×10^8*	**1.8×10^9** *± 2.8×10^9*	**1.1×10^6** *± 8.6×10^6*	**1.5×10^7** *± 1.1×10^7*	**1.2×10^7** *± 7.1×10^6*
fine + pdf[1]	**4.1×10^5** *± 5.5×10^5*	**1.2×10^9** *± 3.5×10^8*	**3.7×10^8** *± 4.0×10^8*	**6.2×10^4** *± 4.7×10^4*	**7.5×10^4** *± 3.2×10^4*	**9.1×10^5** *± 6.4×10^4*
fine + fpa[2]	**9.3×10^5** *± 4.0×10^8*	**7.9×10^8** *± 1.9×10^9*	**6.1×10^8** *± 9.4×10^8*	**1.4×10^5** *± 1.9×10^5*	**6.4×10^5** *± 4.9×10^4*	**1.6×10^6** *± 1.2×10^6*
coarse	**2.4×10^7** *± 1.9×10^7*	**2.4×10^{10}** *± 5.5×10^{10}*	**1.3×10^{10}** *± 3.5×10^{10}*	**8.6×10^5** *± 6.2×10^5*	**1.4×10^7** *± 8.0×10^6*	**2.2×10^7** *± 2.7×10^7*

[1] pdf = 1.2 % **p**otassium **di**formate [2] fpa = 0.9 % free acids (75 % **f**ormic and 25 % **p**ropionic **a**cid)

Conclusion: fpa as well as pdf improved the efficiency of the stomach barrier. In more distal parts of the digestive tract their effect was less obvious concerning counts of *E. coli*, but also here counts of *Salmonella* were reduced markedly when diets contained pdf or fpa. The pelleted diet based on coarsely ground cereals without adding pdf or fpa failed to demonstrate positive effects concerning the passage of orally applied bacteria, but it has to be emphasized that the coarse diet was offered in a pelleted form (comparable amounts of fine particles) and not as meal. In further experiments it will be tested whether the process of pelleting per se affects the stomach barrier function, the passage of orally applied bacteria or - last but not least - the colonisation of intestinal mucosa.

Results achieved here allow to recommend organic acids or pdf as additives, when dietary measures against *E. coli* or *Salmonella* in pigs are required.

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