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Vitamins and Additives in the Nutrition of Man and Animal

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Interactions between vitamins and trace elements

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Bioavailable iron and vitamin A are mainly provided in the human diet by animal source foods. In the poorer populations of the developing world, consuming mainly plant-based diets, deficiencies of these micronutrients are common and could occur in the same individual. In addition, in areas where soil iodine is low, iodine deficiencies can also occur. Infants, children and pregnant and lactating women are most at-risk of deficiency because of their extra requirements for growth. About 250 million pre-school children are said to be vitamin A deficient based on low serum retinol values. Women of child-bearing age are especially at risk of iron deficiency because of monthly blood losses during menstruation. On the basis of low ferritin concentrations, it is estimated that about 2 billion people worldwide are iron deficient and that one billion of these iron deficient people are thought to suffer from iron deficiency anaemia, the most severe form of iron deficiency. Based on low urinary iodine levels 1.9 billion people are estimated to have inadequate iodine nutrition and 200 million suffer from goitre.

In this presentation, the influence of vitamin A deficiency on both, iron and iodine metabolism will be discussed. Low vitamin A status has long been known to promote anaemia although the mechanism is still unclear. Recent iron absorption studies have further complicated the issue as vitamin A added to iron-fortified foods has been shown to increase, decrease or have no effect on iron absorption depending on the subject. Recently, it has been also suggested that vitamin A supplements may have a negative impact on iodine metabolism in iodine deficient subjects. Finally the influence of iron deficiency on iodine metabolism will be discussed and food fortification studies demonstrating the beneficial effect of iron on iodine utilization will be presented.

The contribution of fortified foods to nutrient intake

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In general, micronutrient intakes in European countries appear to be adequate for most nutrients. However, a number of population subgroups are at higher risk of suboptimal intakes for folate, vitamin D, iodine, iron, and calcium. Dietary surveys indicate that food fortification can play a role in mitigating such risks for some nutrients.

Nowadays the interest in fortified food is constantly increasing among consumers as well as food producers. According to this, the range of fortified products in the market is expanding. Safety concerns are addressed in relation to the potentially increased level of nutrients with relatively low tolerable upper intake levels (e.g. iron).

The Institute of Nutritional Sciences thus carried out studies about fortified foods. An evaluation in supermarkets showed that 73% of nearly 500 different fortified products existing in the Austrian market were fortified with vitamin C. Vitamin C was thus the most frequently used nutrient although the vitamin C status can be regarded as good. Other vitamins and minerals were used less often. Most fortified foods were not adapted to the needs of specific target groups.

Food fortification is an effective tool for increasing the intake of some vitamins. The study population met e.g. 40% of the vitamin C requirements from fortified products.

Tab. 1: Average daily intake of micronutrients from fortified foods in % of the D-A-CH reference values, age group 25-51 years, n=1,700; (source: Austrian Nutrition Report 2003)

nutrient	in % of reference	nutrient	in % of reference	nutrient	in % of reference
Vitamin C	40	Vitamin B ₁₂	20	Copper	8
Vitamin B ₆	37	Vitamin E	17	Calcium	8
Niacin	29	Vitamin A	11	Magnesium	7
Vitamin B ₁	27	Folic acid	10	Zinc	4
Vitamin B ₂	23	Vitamin D	2	Iodine (excl. salt)	2*
Pantothenic acid	22	Iron	10	Fluoride	1

Nevertheless, fortification of foods mostly occurs arbitrary and special needs of vulnerable groups, such as elderly people, are rarely considered. The fortification with micronutrients, for which an increase of intake would be desirable (e.g. folic acid), is not sufficient. Other nutrients (e.g. niacin) are added in such a high amount which exceeds by far the required level of intake. However, the risk of an overdose of added nutrients is not given with the consumption habits of currently provided fortified foods.

Fatty acids in animal nutrition: fragile equilibrium between benefit and harm

(*Fettsäuren in der Tierernährung: empfindliches Gleichgewicht zwischen Nutzen und Schaden*)

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In the last decades, there was a permanent shift in perception of and in recommendations for and against certain fatty acids with respect to human health. Currently, low contents and proportions of medium-chain fatty acids and of most *trans* fatty acids, and high proportions of omega-3 (ω -3) fatty acids (respectively a low ω -6 : ω -3 ratio) and of conjugated linoleic acids (CLA) are recommended for human nutrition. There could be health advantages also of butyric acid and phytanic acid. Based on this, various attempts have been made in animal nutrition research to take into account these goals, although often not in a comprehensive way. Basically, two different approaches are chosen. The first is the preferential utilization of feeds which result in an improved fatty acids profile of the products. Grazing of dairy and beef cattle, for instance, is a strategy which nicely fits with animal welfare claims and is often associated with elevated contents of the beneficial fatty acids. The second approach is using desired (functional) fatty acids or precursors of them (e.g., linolenic acid for CLA in ruminants) as feed additives. A large number of experiments to enrich ω -3 fatty acids in eggs, meat and milk has been carried out, fewer of them to elevate CLA and phytanic acid. It has to be noted that even such strategic approaches often disturb the fragile equilibrium of maintaining a favorable fatty acid profile since other fatty acids are modified, too. An example is feeding (long-chain) polyunsaturated fatty acids to ruminants which does not only increase these fatty acids and CLA but also C18:1 *trans* fatty acids in the products. Furthermore, improving the health value of milk, meat and eggs is not the only goal of including fats and fatty acids in animal feed but, more frequently, the purpose is to improve the sensory quality of these products. This is particularly true with respect to firmness of pig carcass fat and of butter in summer, maintaining a white carcass fat and a long shelf life. Very often, these goals are contradictory to those of products considered to be especially healthy. Elements of a more holistic view in determining the ideal fat or fatty acid supplementation in ruminants, pigs and poultry are described.

Potentials of biotechnology to enrich valuable ingredients in crops under the special significance of vitamin E

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Vitamin E is the collective term for a group of eight related tocopherols and an essential component of human and animal diet. The lipid soluble tocopherols consist of a polar chromanol ring with a saturated C₁₆ prenyl side chain and comprise four homologous forms, namely α -, β -, γ - and δ -tocopherol, differing only in the number and position of methyl substituents on the chromanol head group, whereas the tocotrienols possess an unsaturated C₁₆ prenyl group but otherwise the same substitution pattern on the chromanol ring. These different tocopherol forms have varying antioxidative abilities in scavenging oxygen radicals and quenching singlet oxygen, with the α -forms having the highest biological activity. Vitamin E has presumably also a role in the preservation of membrane integrity by forming complexes with products of membrane lipid hydrolysis and in the regulation of transcription and post-translational processes.

The main features of the biosynthetic pathway of prenylquinones in plants have been elucidated several years ago using classical biochemical methods but the genes encoding the respective enzymes of this pathway have been cloned only during the past few years. The identification of the involved genes now facilitates the detailed characterisation of the pathway. This review will describe the characterisation of the key enzymes involved in tocopherol biosynthesis and their importance for the engineering of the tocopherol pathway by showing that the overexpression of the respective chimeric genes in transgenic crop plants increased the total content and changed the composition of the different tocopherols in the seed oil. The combined analysis of these transgenic plants facilitated new insights into the complex mechanisms, which regulate the metabolic flux of substrates and intermediates in the tocopherol pathway and represent new potent tools for future studies of the underlying regulatory mechanisms.

(1→3),(1→6)-beta-D-Glucan as feed additive

((1→3),(1→6)-beta-D-Glucan als Futtermittelzusatzstoff)

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Background: For many years, innate immunity has been considered to be of secondary importance in the hierarchy of immune functions. For the past few years interest in innate immunity has grown remarkably and much work has been done to strengthen its anti-infective potential (1). The purification of the binding protein for (1→3),(1→6)-beta-D-glucan (β -GI) and its biochemical characterization as pattern recognition receptor in macrophages and neutrophils promoted activities to use that polyglucoside as feed additive. The immunomodulatory action of (1→3),(1→6)-beta-D-glucan from various sources was chiefly studied on cellular systems and artificially infected laboratory animals.

Aim: This study is aimed to investigate the effect of β -GI-supplementation on performance and biological parameters of laying hens and broiler chickens living under conditions of contemporary farming.

Methods: The feed of 80 000 laying hens was supplemented with 1.5 kg per t of Leucogard[®] H, an autolytic cell wall product of *Saccharomyces cerevisiae* obtained from Fibona Health Products GmbH, Wiesbaden. 240 000 hens as control group obtained standard feed. In a further study 100 chicken broilers were fed with 1.5 kg per t Leucogard[®] H up to the 15th day of life. 100 chicken broilers as control group obtained standard feed. Blood was taken at various periods of feeding and haematological, immunological and biochemical parameters were investigated.

Results: In blood of laying hens β -GI increases the lymphocyte count, the concentration albumin, γ -Globulin, IgG and α_1 -Antitrypsin. Low values of bile acids indicate an improved liver function. The rate of mortality decreased and the rate of egg production increased. In broiler chickens β -GI feeding augments the content of lymphocytes, monocytes, beta-globulin, and gamma-globulin in blood (3,4).

Conclusion: β -GI given orally improves immune defence and alleviates challenges of microbial and other types of stress.

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Influence of juices rich in carotenoids on biomarkers of inflammatory status and oxidative stress

(Einfluss carotinoidreicher Säfte auf Entzündungsmarker und Biomarker des oxidativen Stresses)

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The intake of plant foods is associated with an increase of plasma levels of vitamins and antioxidants (carotenoids and phenolic compounds). However, only few studies have been carried out on their effects on the oxidative stress and inflammatory bio-markers.

Therefore, the effects of a tomato juice rich in carotenoids, mainly lycopene, and vitamin C on different biomarkers of oxidation and inflammation was investigated. 24 healthy volunteers (20 women, 4 men) with normal body weight participated in the 4-weeks-trial. After a two weeks depletion period, the participants were divided into two groups. The first group obtained 250 mL tomato juice without vitamin-C-fortification twice daily. The second group obtained the same amount of juice but with vitamin-C-fortification (total content 700 mg/L). Blood and urine were taken at the beginning and after two weeks each. Subjects continued their habitual diets during the study. As biomarkers for oxidative stress carbonyl proteins and thiobarbituric acid reactive sub-stances (TBARS) in serum and isoprostane (8-epi-PGF₂α) in urine were measured spectrophotometrically. In addition, as biomarkers of inflammation the concentrations of c-reactive protein, interleukin-1β and tumor necrosis factor (TNFα) in serum were also measured spectrophotometrically. To determine the antioxidative status, serum was investigated on accumulation of carotenoids and vitamin C as well as on ferric reducing antioxidant power (FRAP).

Results will show if there is any difference in behaviour of the parameters measured indicating if antioxidant activity of serum and concentration of the mentioned bio-markers is correlated with carotenoids or vitamin C concentration in serum.

Aspects of nutritional assessment of feeds from genetically modified plants (GMP) with output traits including feed and food additives

(Betrachtungen zur ernährungsphysiologischen Bewertung von Futtermitteln aus gentechnisch veränderten Pflanzen mit beeinflusstem Gehalten an „Zusatzstoffen“)

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The cultivation of genetically modified plants (GMP) increased from 1.7 million to 81 million ha from 1996 to 2004. Scientists and farmers, but also consumers, are asking for a nutritional assessment, including safety aspects, of feeds and food from those plants. Substantial equivalence was created as a framework for the compositional assessment of feeds and food from GMP of the so-called first generation (without substantial changes of composition or without output traits) and is widely accepted. Feeds with intended beneficial physiological properties like amino acids, fatty acids, minerals, vitamins and other substances (GMP with output traits or GMP of the second generation) may contribute to higher feed intake of animals and/or improved conversion of feed/nutrients into food of animal origin and lower excretion of nitrogen, phosphorus and other nutrients. Other types of studies are necessary for nutritional assessment of such feeds. Various experimental designs are recommended to demonstrate the efficiency of changes or of expressed nutrients/constituents:

- Bioavailability or conversion of nutrient precursors into nutrients (e.g., β-carotene)
- Digestibility/bioavailability of nutrients (e.g., amino acids, fatty acids, vitamins)
- Efficiency of substances which may improve nutrient digestibility/availability (e.g., enzymes)
- Utilization of substances with surplus effects (e.g., prebiotics)
- Improvement of sensoric properties/palatability of feed (e.g., essential oils, aromas) The paper will discuss proposals for experimental designs for assessing feeds from GMP of the second generation.

Dense gas technology for the preparation of innovative drug delivery systems

(Hochdruckverfahren zur Herstellung von neuen Wirkstoff-Träger-Systemen)

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Modern therapeutically used drugs are mainly lipophilic compounds. To act on the target structures, the drugs must be dissolved in physiological fluids and absorbed through entrance ports. Since the bioavailability of orally applied drugs depends on the velocity of dissolution and absorption, methods to increase the dissolution of the lipophilic substances are often necessary to reach significant blood levels. A well suitable way is the reduction of particle size to increase the dissolution velocity.

A promising method to improve the bioavailability of pharmaceutical agents is the reduction of particle size by dense gas technology. Former experiments show, that the Rapid Expansion of Supercritical Fluids (RESS) enables the formation of submicron particles of thermally labile drugs (e.g. Griseofulvin, Phytosterol, Ibuprofen). Depending on the process conditions conditions, particles could be produced with a mean diameter of 200 ± 50 nm. Dissolution experiments demonstrate that the RESS processing of Griseofulvin leads to a significantly better dissolution rate of the drug resulting in an improved bioavailability. There is, of course, the limitation that these submicron particles are very difficult to be included in solid dosage forms. To overcome this, two different processes were developed:

In the first process (CPD), the drug must be soluble and the porous carrier insoluble in supercritical CO_2 . The key idea behind CPD is to dissolve the solute of interest in a supercritical fluid, followed by permeation of the binary mixture into the pores and precipitation of the drug inside the pores, caused by a fast pressure drop. The experiments show, that high Ibuprofen loadings (> 80%) of the carrier (β -Cyclodextrin) and an improved dissolution behavior could be achieved. For the second process (CORESS) both, the drug and the biodegradable polymer are dissolved in supercritical CO_2 , followed by the rapid expansion of the ternary mixture. This leads to the simultaneous co-precipitation of the solutes, resulting in the encapsulation of Phytosterol in low-molecular-weight-polymeric matrices. The experimental results illustrate that the simultaneous co-precipitation of two solutes is a promising method to produce composite particles. Depending on the process conditions, the amount of polymer varies in case of Eudragit from ~ 10 to ~ 23 wt-% and from ~ 12 to ~ 43 wt-% in case of L-PLA and these particles appear as a drug core encapsulated in a polymer coating.

In the talk, the experimental setup and results will be discussed in detail.

Trans-11-18:1 is effectively $\Delta 9$ -desaturated compared with trans-12-18:1 in humans

(Trans-11-18:1 wird im Vergleich zu trans-12-18:1 im Menschen effektiv $\Delta 9$ -desaturiert)

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The aim of this human intervention study was to evaluate the $\Delta 9$ -desaturation of trans-11-18:1 (transvaccenic acid; $\Delta 9$) to cis-9,trans-11-18:2 (c9,t11 conjugated linoleic acids; CLA) and of trans-12-18:1 (t12) to cis-9,trans-12-18:2 after a short-term (7 d) and a long-term (42 d) supplementation period. The conversion rates (CR) of both trans-18:1 isomers were estimated by lipid analysis of serum and red blood cell membranes (RBCM). Subjects started with a 2-wk adaptation period without supplements. In the 42 d intervention period, the diet of the test-group (t-group, n=12) was supplemented with 3 g/d of $\Delta 9$ and 3 g/d of t12. The diet of the control-group (c-group, n=12) was supplemented with a CLA and TFA free control oil. Serum $\Delta 9$ and t12 levels of the t-group increased by 5- and 9-fold after 7 d, respectively and increased by 8- and 12-fold after 42 d, respectively when compared with the adaptation period ($P \leq 0.002$). The serum c9,t11-CLA levels increased by 1.7- and 2.0-fold after 7 d and 42 d, respectively ($P \leq 0.001$). After 42 d the t-group's RBCM c9,t11-CLA content was elevated by 20% ($P = 0.021$), whereas c-group's RBCM c9,t11-CLA content was decreased by 50% ($P = 0.002$). The t-group's $\Delta 9$ CR was calculated at 24% by serum and 19% by RBCM. No increase of c9,t12-18:2 was observed in serum as well as in RBCM and thus, no CR of t12 could be determined. In conclusion, the endogenous conversion of dietary $\Delta 9$ to c9,t11-CLA contributes approximately 1/4 to the total CLA reservoir in the human body and should be considered when determining the CLA supply.

Accumulation of non-esterified sitosterol different in particle size in the tissues of the guinea pig

(*Akkumulation von unverestertertem Sitosterol unterschiedlicher Partikelgrößen im Gewebe des Meerschweinchens*)

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The influence of a customary sitosterol and a submicron sitosterol supplemented food was tested on its sterol absorption and accumulation at the guinea pig model. 18 guinea pigs (Dunkin Hartley, female) were randomized to three groups with six animals each. Over two weeks they obtained either conventional food (control) or sitosterol supplemented food whereas there were two sizes of sterol particles: 10-90 μm (original) and 200-300 nm (submicron). The mean daily sitosterol uptake was 21 ± 7 mg (control), 154 ± 8 mg (original) and 127 ± 18 mg (submicron), respectively. Cholesterol and sitosterol were analysed in plasma, blood cells, tissues (liver, kidneys, gut segments), gut contents (jejunum, caecum, colon) and in faeces. Cholesterol concentrations in plasma, blood cells, kidneys, liver, jejunum (mucosa and serosa), caecum and colon remained unchanged. In the dry matter (DM) of faeces an elevated cholesterol concentration was verified at both supplement groups compared to control (control: $0,26 \pm 0,01$ mg/g DM, original: $0,32 \pm 0,02$ mg/g DM, submicron $0,31 \pm 0,02$ mg/g DM; $p < 0,05$). Besides kidney the sitosterol concentrations in all tested matrices have been significantly increased to control (plasma [$\mu\text{g/mL}$]: control $13,4 \pm 1,5$ vs. original $23,1 \pm 10,4$ vs. submicron $24,4 \pm 3,9$; serosa [mg/g DM]: control $0,52 \pm 0,2$ vs. original $1,16 \pm 0,59$ vs. submicron $1,17 \pm 0,43$; liver [mg/g DM]: control $0,09 \pm 0,01$ vs. original $0,17 \pm 0,03$ vs. submicron $0,19 \pm 0,03$; $p < 0,05$).

The particle size of the supplemented sitosterol had no influence on the sitosterol and cholesterol concentrations in the tissues and gut contents.

Enrichment of cows' milk with natural or synthetic vitamin E

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The content of vitamin E (α -tocopherol) in milk is very dependent on the content in the cows feed. Thus cows' grassing fresh green pasture has a significant higher content of α -tocopherol in milk than cows fed corn silage or a high proportion of concentrate. Numerous studies have shown a poor transfer of synthetic α -tocopherol from feed to cows' milk. Thus only a small enrichment of the milk has been observed even though the cows' were fed more than 3 g *all-rac*- α -tocopheryl acetate per day. The present presentation comprise results from three feeding experiments with cows showing the possibilities for enrichment of milk with α -tocopherol via the feed, as well as one experiment dealing with the metabolism of the different stereoisomers of α -tocopherol in cows' after an intramuscularly injection of *all-rac*- α -tocopheryl acetate and their secretion into cows' milk. The feeding experiments show that addition of natural α -tocopherol either together with oil or as RRR- α -tocopheryl acetate to the feed or directly via a high content in the roughage cause a significant increase of α -tocopherol in milk. The metabolism experiment showed a significant higher clearance in plasma of the 2S forms of α -tocopherol compared to the natural RRR- α -tocopherol with the mixed R/S isomers in between. The synthetic isomers disappeared faster from plasma than from milk, apparently because they were transferred to the liver, from where they were metabolised or passive and slowly released back to the blood stream, from where they were secreted into the milk.

Vitamin D metabolites: Physiological role in bone and pancreas and therapeutic potential for bone diseases and diabetes

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It is well established that the vitamin D hormone $1\alpha,25\text{-dihydroxyvitamin D}_3$ has an essential role for mineral metabolism. Vitamin D analogs act through the vitamin D receptor (VDR), which is a member of the nuclear receptor superfamily. Research conducted within the last years has conclusively shown that vitamin D metabolites lack an essential physiological function in bone, and that the development of rickets in the course of vitamin D deficiency is solely caused by impaired intestinal calcium absorption and subsequent secondary hyperparathyroidism. It is, on the other hand, well established that pharmacological administration of vitamin D analogs can have pronounced bone anabolic effects, and there is good evidence from a variety of studies that at least some of the bone anabolic properties of vitamin D analogs are independent of the untoward calcemic effects. In addition, recent experiments have suggested that the suppression of bone resorption seen under treatment with vitamin D analogs depends on the route of administration, and is mediated both by a direct antiresorptive effect, and by indirect effects via hypercalcemia and suppression of parathyroid hormone secretion. An important goal for future research aimed at the development of bone specific vitamin D analogs is to gain further insight into the molecular pathways involved in the direct bone anabolic effects of this compound class.

Apart from its essential role in mineral metabolism, the vitamin D hormone may have important functions in other organ systems. For example, it is well known that pancreatic islets show strong expression of the VDR. We have recently shown that gene-targeted mice with a functionally inactive mutant VDR show impaired oral glucose tolerance and reduced insulin secretory capacity, independent of changes in calcium homeostasis. Additional evidence linking the VDR to the endocrine functions of the pancreas came from epidemiological studies showing that VDR restriction site polymorphisms are associated with the genetic susceptibility to type 1 diabetes in different populations, and that vitamin D supplementation in early childhood is associated with a reduced risk for type 1 diabetes. In addition, recent studies have indicated that peripheral insulin sensitivity is regulated by the vitamin D status. Therefore, vitamin D analogs with reduced calcemic activity may have a future role in the prevention and therapy of diabetes.

On the mode of action of probiotics

(Zur Wirkungsweise von Probiotika)

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Probiotics are viable forms of micro-organisms, which are used in animal nutrition as feed additives. At present 19 preparations containing one strain of bacteria or yeast or a combination of two bacterial strains are authorized for the use as feed additives in the EU. Their approval is based on experiments showing their beneficial effects for the target animal species. However, as revealed by publications on this subject the effects on performance data are not consistent and rarely significant. On the other hand the majority of publications show especially in piglets and calves significant reductions on the incidence or frequency of diarrhoea due to the addition of a probiotic micro-organism to the feed. Because of the different origin of micro-organisms used as probiotics, including e.g. *Enterococcus* spp., spores of *Bacillus* spp. or *Saccharomyces cerevisiae*, one may suppose that the mode of action may differ considerably.

This contribution will summarize the results of an integrative analysis on the mode of action of probiotics in pigs sponsored by the German Research Foundation (DFG – project FOR 438). Two probiotics were studied systematically, i.e. *Enterococcus faecium* NCIMB 10415 and *Bacillus cereus* var. *toyoi*. Both did not influence the performance of sows or piglets significantly but reduced incidence of diarrhoea after weaning significantly. Bacterial communities of sows and piglets were modified consistently. The frequency of several *E. coli* pathogens and pathogenic genes was reduced by both probiotics. It was shown that both probiotics were directly transferred from the sow to the piglets. During the first weeks of life both probiotics modified the population of intraepithelial lymphocytes, however, in a different way. *E. faecium* seems to act mainly via the modification of the intestinal flora, while *B. cereus* seems to stimulate the immune system probably in a direct way. The interaction between micro-organisms and the host animal is very complex and specific for the micro-organism (probably even strain) used as feed additive.

Free radicals, antioxidants and aging

(*Freie Radikale, Antioxidantien und Altern*)

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A wide spectrum of beneficial activity to human health has been advocated for antioxidant nutrients. More recently the ability of antioxidants to affect gene expression and cell response has been reported, providing a novel mechanistic perspective on their biological activity. An imbalance in the oxidant antioxidant homeostasis may be detrimental to the organism and contribute to the pathogenesis of age-related diseases. The efficacy of different antioxidants (e.g., polyphenols and vitamin E) to influence positively the molecular mechanism implicated in age-related diseases including neurodegeneration (e.g. Alzheimer disease) is becoming increasingly important. Differential changes in the expression of several groups of genes might be a key point underlying the complex behaviour of antioxidants *in vivo*. Therefore determining a global picture of the effect of antioxidants on gene expression through transcriptomic techniques should provide a better understanding of their action on the molecular level. Gene chips technology in combination with proteomics opens up new avenues in discovering redox-sensitive genes, transcription factors and signal-transduction pathways. This approach may lead to better insights into the role of dietary antioxidants in healthy aging, thereby offering a novel strategy in nutrition research.

Encapsulation of vitamins and ingredients: Demands and technological options

(*Verkapselung von Vitaminen und Zusatzstoffen: Bedarf und technologische Möglichkeiten*)

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During the last years growing markets for human convenience food as well as the nutritional needs in modern animal production led to an increasing demand for products based on encapsulated vitamins and other ingredients.

Beneficial effects such as a higher shelf-life due to protection against environmental effects, e.g. air, light, and especially a controlled release of the substances inside the intestinal tract of both humans and animals can be achieved by encapsulation.

From the technological point of view two major challenges has to be faced for the manufacture of suitable encapsulated products. First, depending on the kind of protection needed and the conditions under which the encapsulated substances should be released in a controlled way, e.g. temperature, acidic or neutral pH, a suitable encapsulation matrix has to be chosen under the limited number of substances allowed for nutritional uses. Second, the encapsulation technology should be able to manufacture the product in the desired concentration inside the chosen matrix, the desired form, size distribution and stability as well as for low costs.

Different encapsulation strategies mainly based on controlled release demands and technologies for product manufacture will be presented and their pros and cons will be discussed.

Additional consumption and diet's supplementation participation in vitamins intake during alimentation of young female students of the Military University of Technology in Warsaw

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Women, candidates for professional soldiers, have been studying in the Polish military universities for many years. Carried out inquiry showed that some of women did not eat all the meals served during the day. In such situation mass nutrition might not meet the nutrition requirements in full and participation of additionally bought food may significantly affect the fulfillment of the obligatory in Poland norms for particular nutritive elements, including vitamins. The aim of the work was estimation of additional consumption and supplementation participation in the nutrition norms for vitamins fulfillment (on the safe level) for female students of the military university. A poll among 84 female students from the Military University of Technology (MUT) in Warsaw, young women aged 21-25, burdened with intense physical activities, was conducted in 2004. To calculate the vitamins content in the additionally eaten food the software „DIETETYK 2”, issued by the National Food and Nutrition Institute in Warsaw, was used. The average age of examined women was 22.9 ± 1.0. Up to 92.9% ate meals in the canteen and 7.1% ate at home. All persons eating in the canteen bought additional food products. The supplementation was used by 67.9% of female students eating in the canteen. Significant individual differentiation in vitamins intake both with the diet's supplementation and additional food consumption was found. As it results from carried out examination additional consumption, including supplementation, delivered vitamins in amounts exceeding the recommended in Poland norms. The average daily intake of vitamins A, E, B₁₂ and C amounted as follows: 708.1 ±1104.3 µg, 13.0 ±28.1 mg, 3.0 ±11.5 µg, 124.3 ±129.4 mg, what made 118.0%, 162.5%, 150.0% and 207.2% of recommended norm. Besides, additional consumption and supplementation significantly affected the fulfillment of the Polish requirements for such vitamins as: folic acid, B₁, B₂ and B₆. Intake of these vitamins made 54.7-83.5% of recommended norm. Carried out examinations revealed significant influence of additional consumption and diet's supplementation with vitamins on fulfillment of nutrition recommendations for vitamins for young female students of MUT.

Does dietary intake of rosehip puree affect levels of carotenoids, vitamin E and vitamin C in human plasma?

(Beeinflusst die Aufnahme von Hagebuttenmark die Gehalte an Carotinoiden, Vitamin E und Vitamin C im menschlichen Plasma?)

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During the last years, the use of some wild fruits as raw materials for a lot of products increased. Rosehips as fruits of wild roses are used to produce mark, jams and juices. Rosehips have traditionally been used as a vitamin supplement or for health food production, as the fruits are a rich source of ascorbic acid. Studies have also shown that rosehip extract inhibits lipid oxidation in vitro and reduces the chemotaxis and chemoluminescence of leucocytes. Extracts also possess antioxidative capacity as well as antimutagenic effects. The anti-inflammatory properties of rosehips are useful as a natural treatment in patients with osteoarthritis. Until now it is not clear which compounds are responsible for these effects.

In addition to ascorbic acid, rosehips deliver other antioxidants such as carotenoids, tocopherols/tocotrienols and phenolics. Fruits of wild roses showed remarkable contents of lycopene with an unexpected isomer pattern. Lycopene is an acyclic carotenoid which is found only in few foods such as tomatoes and tomato products. Recent epidemiological studies showed that supplementation of diets rich in lycopene was reversely connected with the risk of many chronic diseases, such as cancer and heart diseases. All these properties can only be effective in vivo if lycopene and the other ingredients are available to the human organism.

The purpose of the study was to determine the effect of dietary intake of rosehip puree on plasma levels of carotenoids, vitamin E and vitamin C. After a two weeks depletion period, the five participants ingested 38 g rosehip puree combined with 2.5 g sun flower oil twice a day for four weeks. All subjects avoided food rich in lycopene during the six weeks of the study. Fasting blood samples were withdrawn from the participants prior the study and thereafter weekly. The blood samples were centrifuged and aliquots of plasma were analysed on concentrations of carotenoids and vitamin E by high performance liquid chromatography. Vitamin C levels were analysed spectrophotometrically.

Mechanisms of the insulin sensitising effect of supranutritional selenate doses in type II diabetic dbdb mice

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To examine the mechanisms behind the insulin sensitising effect of high supranutritional selenate doses in a type II diabetic animal model 21 young female dbdb mice were randomly assigned to 3 experimental groups of 7 animals each. Group 0 was fed a selenium deficient diet for 8 weeks while the animals of groups Se IV (selenite) and Se VI (selenate) were supplemented with high supranutritional doses of selenite and selenate in addition to the diet.

- Selenate treatment of the mice increased whole body insulin sensitivity significantly in comparison to selenium deficiency and selenite treatment.
- Selenate administration to the mice (0.58 ± 0.15 U/mg protein) inhibited the activity of cellular protein tyrosine phosphatases (PTPs) in comparison to selenium deficient (0.93 ± 0.23) and selenite treated mice (1.20 ± 0.36).
- The application of supranutritional selenate doses resulted in a 2 to 3-fold increase in the expression of the peroxisome proliferator-activated receptor gamma (PPAR) accompanied by significant changes in total liver fat content and in the portions of triglycerides, phospholipides and cholesterol in comparison to both other groups.

The results of our study in dbdb mice showed that two efficient mechanisms, discussed as important factors for an increase in insulin sensitivity, were affected by the administration of high supranutritional selenate doses and therefore these mechanisms may have mediated the insulin sensitising effect of selenate.

Effects of vitamin E and vitamin C on cellular and humoral immune parameters of dogs

(*Effekte von Vitamin E und Vitamin C auf das zelluläre und humorale Immunsystem des Hundes*)

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Vitamin C is a water soluble substance, and is believed to be one of the most important antioxidants in extracellular fluids. Vitamin E in contrast is fat soluble, and is the major antioxidant in cellular membranes, protecting polyunsaturated fatty acids against oxidation. The objective of the present study was to investigate the effect of both vitamins on the cell-mediated and humoral immune response of adult dogs.

A total of 15 beagle dogs (1 to 7 years old) were split in three groups and obtained one capsule filled with 60 mg vitamin E and respective 0, 30 or 60 mg vitamin C per day for a period of 35 days. Each supplementation was administered to all three groups alternately after 7 days wash-out. At the beginning and at the end of each period blood was sampled to assess haematology, basic serum biochemistry, differential blood count, lymphocyte proliferative response to three different mitogens (ConA, PHA and PWM), lymphocyte subset distribution (CD4, CD8, CD21 and CD5) and IgA and IgG concentrations in the plasma. Data were evaluated by analysis of variance ($p \leq 0.05$).

Neither general health nor haematological or biochemical parameters were affected by vitamin supplementation; all measured parameters were in the physiological ranges. The differential blood count and the expression of cell surface markers of lymphocytes also did not change during the experimental period. Lymphocyte proliferation significantly increased after treatment with 60 mg vitamin E and 30 mg vitamin C. For the other two groups, a significant decrease of lymphocyte proliferation was observed. The IgA-concentration in the plasma was not altered while the IgG-concentration increased after administration of vitamin E and vitamin C in both dosages.

The current findings, however, indicate that vitamin E and vitamin C exert an effect on the immune system of adult dogs. Further investigations are necessary to understand the mode of action.

Influence of feeding various amounts of minerals and vitamins to first lactating cows on some blood parameters

(Einfluss der Fütterung unterschiedlicher Mengen an Mineralstoffen und Vitaminen bei erstlaktierenden Kühen auf ausgewählte Blutparameter)

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By monitoring the status of health of cows on farm level many undersupplies of minerals and vitamins in their blood were reported.

Methods: In this experiment 30 first lactating cows were split into two feeding groups, all receiving a maize-grass-silage *ad libitum* at the rate of 60:40 (DM-base) and concentrate depending on their milk yield. Group 1 was fed a concentrate with mineral and vitamin level according to the recommendations of the GfE (2001), whereas group 2 was offered twice as much. From all animals blood was taken from the *vena jugularis* one week before calving and one, four, eight, sixteen and thirty-six weeks after calving. The concentrations of calcium, magnesium, phosphorus, copper, zinc, β -carotin and of the vitamins A and E in the blood serum were analysed and compared with present reference values. **Results:** During lactation the progression of the mean values of phosphorus, zinc and Vitamin E varied between the feeding groups ($p < 0.05$). The other parameters did not vary significantly between the groups. In comparison to reference values, lower concentrations of copper, zinc, β -carotin and vitamin E were found especially at the beginning of lactation. After 16 weeks concentration of β -carotin and after 4 weeks concentration of vitamin E were in the reference interval. The concentrations of phosphorus and zinc were lower in group 1 during the whole lactation, whereby only zinc lied under reference values. Most time of lactation excepting one week after calving concentrations of copper were lower than the reference values in both feeding groups.

Conclusions: In comparison with present reference values many cows showed low blood concentrations of copper, zinc, β -carotin and vitamin E in spite of recommended feeding, but no observed clinical deficiency syndromes. More studies under consideration of further indicator samples (liver, hair etc.) are necessary to assess reference values.

How does sample concentration affect measurement of antioxidant capacity?

(Wie beeinflusst die Probenkonzentration die Messung der antioxidativen Kapazität?)

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Several investigations have shown a concentration-dependent antioxidant activity of ascorbic acid in peroxyl radical trapping tests, while others demonstrated that the antioxidant efficiency of ascorbate in the FRAP assay is not affected by concentration. Aim of the present study was to determine the antioxidant activity of four standard substances (Trolox, ascorbic acid, gallic acid and uric acid) in two different versions of the TEAC (trolox equivalent antioxidant capacity) assay, in the FRAP (ferric reducing antioxidant power) assay and in the PCL (photochemiluminescence) assay as well as the total phenolics content using the method of Folin-Ciocalteu. Analyses were done on three different days within several months. All substances were measured in four to seven different concentrations.

There was no effect of dilution for Trolox in the first version of TEAC assay and in the PCL assay, for ascorbic acid in the second version of TEAC, PCL, FRAP and Folin-Ciocalteu assay, for gallic acid in the first version of TEAC assay and for uric acid in the FRAP assay. In the PCL assay gallic acid showed an increasing antioxidant activity as its concentration increased (50-125 μ M). The remaining measurements showed conflicting results. Gallic acid possessed the strongest potential in all tests while the ranking order of the other substances varied. These findings confirm previous observations showing several single compounds reacting differently in diverse assays for measuring antioxidant capacity which is due to the different reaction mechanisms. Within-run coefficients of variation for all assays and all compounds tested were in an acceptable range, on average 6 and 7% in the two versions of the TEAC assay, 7% in the FRAP assay, 10% in the PCL assay (except gallic acid) and 6% in the Folin-Ciocalteu assay. In contrast, repeated measurements in longer time intervals resulted in considerably higher coefficients of variation which shows the problem of reproducibility. It is assumed that single compounds and complex food extracts behave in different ways. Thus, despite the four standard substances, a tea extract, a strawberry nectar and a tomato extract were analysed in several concentrations by using the five different tests.

Changes of the vitamin C concentration in the serum and of the tissue Total Antioxidant Status (TAS) in the precancerous states of the stomach cancer

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The aim of the study was to evaluate the changes of the vitamin C concentration in the serum against the background of the Total Antioxidant Status (TAS) of the gastric mucosa in considering the occurrence of the precancerous states of the stomach cancer. Material and methods. The analysis included 91 patients (61 men and 30 women) aged 31 to 65 years who were examined because of pains in the upper part of the alimentary canal. In 31 of them chronic gastritis was diagnosed, in 23 – gastric peptic ulcer, and in the remaining 37 – duodenal ulcer. In the course of the diagnosis the following points were included: a) gastroscopic examination with histopathological evaluation of the segments taken from the stomach stump mucosa; b) the evaluation of the TAS of the gastric mucosa (test of the firm Randox, TAS reference values: 1,3 – 1,77 mmol/l); c) estimation of the vitamin C concentration in the serum. The vitamin C concentration in the serum was evaluated chromatographically. The supply of the vitamin in all persons analysed was standardized.

Results and conclusions. 1. The range of the reference values of the vitamin C concentration in the serum amounting to 0.91±0.18 mg/dl was established on the basis of the results obtained from the examination of reference group. 2. The increase of the pathology of the gastric mucosa, starting from the normal mucosa, through the inflammatory changes, to the atrophic changes is associated with the fall of the ascorbates concentration in the serum as well as with the decrease of the average TAS value of the gastric mucosa (vit. C: 0.84±0.16, 0.63±0.19, 0.58±0.25, 0.51±0.18 mg/dl; TAS: 1.39±0.27, 0.87±0.21, 6.1±0.17, 1.01±0.25 mmol/ml respectively). 3. However, in the cells affected by the dysplastic changes, there comes about a construction of the supplies of the antioxidants despite a hitherto low vitamin C concentration in the serum.

Importance of vitamin E supplementation during suckling and postweaning for α -tocopherol status and immune responses of piglets

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Birth and weaning are two critical stages where it is difficult to maintain a proper vitamin E status for optimal growth and health status in pigs. Vitamin E deficiency has been found to predispose pigs to different diseases, including *Escherichia coli* infection. On the other hand, dietary vitamin E increased cellular and humoral immunity in pigs. The purpose of this study was to test two nutritional strategies to increase α -tocopherol (α -toc) and immune status of piglets. Experiment 1 involved 12 crossbreed sows that were fed increasing levels of all-rac- α -tocopheryl acetate (**All-Ace**) (70, 150, and 250 IU/kg) during lactation. After weaning (28 d), piglets were fed the same amount of All-Ace (70 IU/kg feed). Experiment 2 involved 7 litters of piglets obtained from sows fed 250 IU All-Ace/kg feed, and the piglets were fed three dietary levels of vitamin E during the postweaning period (85, 150, and 300 IU All-Ace/kg feed) from 28 to 56 d. Experiment 1 showed that increasing maternal dietary vitamin E supplementation increased the concentration of α -toc in plasma ($P=0.02$) and milk ($P=0.007$) of sows. When lipid-standardized, plasma α -toc was increased in suckling piglets of sows fed 250 IU of All-Ace compared with other sow-groups ($P=0.005$). At 28 d of age, α -toc concentration in tissues was increased with supplementation of high All-Ace levels to the sows; however, after weaning, a decrease in α -toc concentration in most tissues (except liver) was observed. In experiment 2, supplementation of 300 IU All-Ace/kg feed to the piglets increased α -toc concentration in serum at 42, 49, and 56 d of age compared to 85 IU. At 56 d, α -toc concentration of liver ($P<0.001$), adipose tissue ($P<0.001$), heart ($P=0.002$) and muscles ($P<0.001$) was increased in piglets fed 150 or 300 IU All-Ace/kg feed when compared to 85 IU. When considering the difference between tissue α -toc at 28 and 56 d, addition of 150 IU Ace/kg feed seemed to be enough to maintain the vitamin E status postweaning in piglets weaned from sows fed 250 IU All-Ace/kg feed. The maternal vitamin E supplementation improved the *Escherichia coli* antibody responses in piglets postweaning ($P<0.001$), whereas minor influence was observed with regard to the influence of increasing vitamin E supplementation to postweaners on their immune responses.

Effects of enhanced vitamin B supplementation in fattening pigs

(Auswirkungen einer erhöhten Vitamin B-Versorgung in der Schweinemast)

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Two groups of fattening pigs fed with diets differing in vitamin B supplementation were investigated to clarify the effect of a dietary enhanced vitamin B content. Group 1 which is regarded as control group received a diet with a vitamin B supplementation like the diet used in the fattening performance testing, the diet of group B contained about eight times more vitamin B than the NRC standard (Table).

Table: Vitamin supplementation of mixed feed

Supplements (mg/kg mixed feed)	B1	B2	B6	B12	Niacin	Pantothenic Acid	Folic Acid	Biotin
1 (Control)	-	1.5	-	0.01	18	3	-	-
2 (Experiment)	3	15.5	2	0.072	94	56	2	0.05

B¹ - Thiamin, B² - Riboflavin, B⁶ - Pyridoxine

In Köllitsch/Saxony one experiment was carried out with 15 fattening pigs per group kept on fully slatted floor (experiment A). In Remderoda/Thuringia two experiments were carried out with 16 fattening castrates per group (experiment B and C). The experiments started when the animals were 10 weeks old. The body weight of each animal was taken at the beginning, at the end of the pre fattening period, and the end of the experiment.

The higher vitamin B content of the diet took an insignificant effect on the fattening performance and carcass parameters. On average of the three experiments 1.0% lower feed intake, 2.2% increased life weight gain, and 3.1% improved feed efficiency were observed. There was no effect on mortality. However, the percentage of skeletal muscle was diminished by 2.1%-points in experiments A whereas an increased percentage of skeletal muscle by 1.2% and 0.7%-points was observed in experiments B and C.

The higher vitamin B content of the diet in group 2 costs 1.09 €/t mixed feed compared to group 1. The fattening performance could be improved but not significantly by an 8 times higher vitamin B supplementation than the NRC standard in the three experiments as well as in investigations performed by Coehlo (2001) and Weiss and Quanz (2003). There was no tendency in changing the carcass parameters.

It can be pointed out that the higher vitamin B supplementation of the diets of fattening pigs tended to have a positive effect. The costs for the higher vitamin B content are low. However, the results of the three experiments do not allow to give a general recommendation for an enhanced vitamin B content of the diets of fattening pigs.

Investigations on the biotin requirement of broiler chicken

(Untersuchungen zum Biotinbedarf von Masthähnchen)

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Former investigations indicated a considerable reduction of feed intake and growth performance of broiler chicken, when biotin concentration of the feed was only 20% below recommendations. Therefore a further trial was conducted to confirm the small allowance between optimal and suboptimal biotin supply. A basic diet was mixed covering the nutrient requirements of broiler chicken with the exception of biotin, which amounted to only 70% of the recommended content of 150 µg available biotin per kg diet. The basic diet was supplemented with 20, 30, 50, 75 or 200 µg biotin per kg. The resulting six diets were fed for 5 weeks to a total of 360 male day-old chicken (ROSS), housed in groups of 10 in cages resulting in 6 replicates per diet. Feed intake, body weight gain, and feed-to-gain efficiency confirmed former observations, that a 20%-deficiency of biotin impairs performance of broiler chicken significantly by more than 6% (cf. table). Exceeding the actual biotin recommendations (GfE 2000), growth performance increased slightly, additionally, biotin concentration in the liver rose considerably.

Biotin in the diet	% of recommendation					
	70	80	90	100	120	200
Feed intake g/d	75.1	75.8	78.2	79.0	79.1	80.9
Final body weight g	1739 ^b	1788 ^{ab}	1874 ^{ab}	1872 ^{ab}	1905 ^a	1900 ^a
Feed efficiency (final week) g feed/g weight gain	1.86	1.71	1.58	1.63	1.66	1.67
Biotin in the liver µg/g	2.17 ^a	2.57 ^b	3.24 ^c	3.80 ^d	4.30 ^e	4.61 ^f

^{a,b} means without same superscripts differ significantly (SNK test)

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Analysis of pantothenic acid and folic acid: conclusions from method comparisons

(Analytik von Pantothensäure und Folsäure: Folgerungen aus Methodenvergleichen)

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Method comparisons were performed for the quantitation of pantothenic acid and folic acid.

For folic acid (FA) in enriched cereal products, a stable isotope dilution assay (SIDA) based on the use of [²H₄]-FA as internal standard was compared to a simple HPLC method with UV detection in a collaborative trial. Whereas SIDA resulted in data quite close to the labelled amounts of FA, the HPLC-UV method gave quite different and imprecise data. Further experiments revealed that extraction at pH 11 and the use of large capacity solid phase extraction (SPE) cartridges significantly improved the results.

For pantothenic acid (PA), different foods and feedstuffs were analyzed by the recently developed SIDA and by the standard method, a microbiological assay (MA). SIDA involved the use of [¹³C₃, ¹⁵N]-pantothenic acid as the internal standard and detection by liquid chromatography-tandem mass spectrometry. For samples derived from plants a good accordance between the MA and the SIDA of total PA was found, whereas for the products of animal origin higher contents were measured by MA than by SIDA. From the results of treatments by pantothenase and phosphatase on the one hand and papain and diastase on the other, it was concluded that MA is able to measure a significant amount of bound PA. Furthermore, the data imply that microbial enzymes were able to cleave PA conjugates more effectively than pantothenase and phosphatase treatment.

Although significant process in terms of precision and accuracy has been achieved by using stable isotope dilution technology in enriched foods, for evaluations of the bioavailability of naturally occurring vitamins, microbiological assays may still be of use. However, there are still open questions regarding the underlying mechanisms and substances responsible for the growth of microorganisms.

Transfer of antibiotics used in animal husbandry from slurry into food

(Transfer von Antibiotika aus der Tierhaltung über Gülle in Nahrungsmittel)

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Low antibiotic concentrations are known to cause bacterial resistance in human and animal microbes. Little is known about the environmental effects of these drugs and their transfer into plants after using animal excreta as fertilizer. To achieve more information pigs were medicated twice with sulfadiazine (SD), trimethoprim (TMP) and chlortetracycline (CTC) and excretions collected. The slurry was administered as fertilizer on wheat and lettuce cultivations. Antibiotic residues in slurry, soil, wheat and lettuce were analysed by LC-MS/MS methods.

During 8 months storage overall antibiotic concentration of SFD decreased from 502 mg/kg slurry (1st medication period) and 559 mg/kg slurry (2nd period) to 29% and 50% of the original amount, respectively; CTC decreased from ~110 mg/kg slurry to 38% and 86%. Overall administration of antibiotics to soil was 557 mg SFD and 176 mg CTC/m² after single and 922 mg SFD and 284 mg CTC/m² soil after double application of slurry. Antibiotic recovery rates from soil reached peaks after 2nd slurry administration and were reduced within 4 to 5 months to 32% and 17% of maximum SFD and CTC recovery rates after single and 30% and 9% of SFD and CTC recovery after double slurry administration. Initially wheat roots contained 0.3 to 0.5mg SFD and 0.9 to 1.1mg CTC/kg DM. This amount was reduced to 0.1mg CTC/kg DM at harvest. Green leaves and stems contained 0.6 to 1.1mg CTC/kg DM but only traces of SFD. At harvest virtually no antibiotics were detected in straw, but wheat grain of the double fertilized plot 1 contained 0.05mg CTC/kg DM. Hence, strong evidence is given for the transfer of antibiotic residues in slurry via soil into plants which may be used for human consumption. Studies on several hydroponically grown plants reveal also the uptake of antibiotics by roots from nutrient and transport in plants. **Acknowledgement:** The authors are grateful to the "Ministerium für Umwelt und Naturschutz, Landwirtschaft und Verbraucherschutz (MUNLV) des Landes NRW" for financial support.

Effect of L-carnitine supplementation of sows on body composition and lipid metabolism of piglets

(Einfluss einer L-Carnitin Supplementation bei Sauen auf Körperzusammensetzung und Lipidstoffwechsel der Ferkel)

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Recent studies showed that dietary L-carnitine supplementation of sows during pregnancy increases birth weights of piglets and their body weight gain during the suckling period. This improved postnatal growth of piglets could be attributed to a different body composition. Therefore the aim of this study was to investigate the effect of supplementation sows' diets with L-carnitine on the body composition of newborn and suckling piglets. For this a total of 27 crossbred gilts (German land race x Large white) were allotted to a treatment (n=14) and a control group (n=13) and inseminated with sperm from Pietrain boars. All sows were fed commercially available diets with low carnitine concentrations (gestation diet: 10mg/kg; lactation diet: 3mg/kg). Sows of the treatment group received per day additionally 125 mg of L-carnitine during pregnancy and 250 mg of L-carnitine during lactation. From each litter an average weighted piglet was killed on the 1st, the 10th and the 20th day of life to determine the carcass composition. Because L-carnitine is involved in the lipid metabolism, metabolic parameters, i.e. concentrations of triglycerides, cholesterol and free fatty acids in liver and plasma of the newborn and suckling piglets, were also determined. There were no differences between the carcass composition of piglets of control sows and those of sows treated with L-carnitine at any time of measurement, and there were less differences in concentrations of lipids in liver and plasma between both groups of piglets. However, piglets of supplemented sows had higher concentrations of total carnitine in blood and carcass (p<0.05). This study shows that piglets of sows treated with L-carnitine are provided with more L-carnitine and have therefore a better carnitine status at birth and during suckling than piglets of control sows. Their body composition, i.e. deposition of fat and protein, and their lipid metabolism in this early phase of growth are not influenced by supplementation of sows with L-carnitine. Probably the increased postnatal growth of piglets is not caused by a different body composition, but may be the result of an increased milk consumption.

Studies on the net absorption of L-carnitine in the ileum of broiler chicken

(Untersuchungen zur Nettoabsorption von L-Carnitin im Ileum beim Broiler)

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Es wurde untersucht, wie hoch die Nettoabsorptionsrate (NAR) für supplementiertes L-Carnitin in unterschiedlichen Abschnitten des Ileums beim Broiler ist. Je 60 männliche Broiler (Ross) wurden mit einer von sechs Dosierungen (0, 25, 50, 100, 200, 400 mg/kg) von L-Carnitin und TiO₂ (5 g/kg) als unverdaulichem Marker auf der Grundlage einer üblichen und bedarfsgerecht formulierten Futtermischung versorgt. Die Tiere wurden in Gruppen zu je 10 in Käfigen gehalten und ab dem 22. Lebenstag den Behandlungen zugeordnet (n=6 Käfige je Behandlung). Nach 7-tägiger Fütterung und Tötung wurde ein definierter Abschnitt des Dünndarms zwischen dem Meckelschen Divertikulum und dem Ileum entnommen. Dieser Darmabschnitt wurde gedreht, der Chymus je Drittel entnommen und für alle Tiere eines Käfigs gepoolt und gefriergetrocknet. Zudem wurden an den letzten drei Versuchstagen Exkrementproben gewonnen. Die Bestimmung der Gehalte an freiem L-Carnitin erfolgte mittels ESI-MS/MS. Die NAR wurde unter Berücksichtigung der Konzentrationen von TiO₂ berechnet. Es traten keine signifikanten Interaktionen bei der NAR des L-Carnitins zwischen der Dosierungshöhe und dem Ileumabschnitt auf. Im Durchschnitt aller Dosierungsstufen war die NAR mit 84 % im letzten Ileumdrittel signifikant höher als im ersten (79 %) und nicht signifikant verschieden vom mittleren (82 %). Hieraus wird geschlossen, dass die Nettoabsorption von L-Carnitin am Meckelschen Divertikulum noch nicht abgeschlossen ist. Im letzten Ileumabschnitt betrug die NAR bis zu einer Dosierung von 50 mg/kg mindestens 94 %. Mit weiter steigender Dosierung ging die NAR kontinuierlich bis auf 54 % zurück. Dies zeigt, dass im Dünndarm eine Anpassung der Carnitinabsorption an die Höhe der Zufuhr stattfindet. Ein Vergleich der Carnitinnengen, die am Ende des Ileums fließen bzw. mit den Exkrementen ausgeschieden werden, legt zudem die Schlussfolgerung nahe, dass die Anpassung der Absorption der wesentliche Regulationsweg des Broilers ist und die renale Ausscheidung nur eine geringe Bedeutung hat.

Influence of supplemented Phytase on the Cu- and Zn-Content of pig carcasses

(*Einfluss zugesetzter Phytase auf den Cu- und Zn-Gehalt von Schweineschlachtkörpern*)

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50 carcasses from all castrated males and 3 carcasses from females of a trial with 100 growing-fattening pigs, (50 castrated males : 50 females), were analysed to investigate:

1. Influence of the level of Cu- and Zn-concentration in feed.
2. Influence of chemical binding form.
3. Supplementation of phytase of microbial origin on Cu- and Zn-concentration in the whole body at about 115 kg LW.

There were three levels of feed concentration, only native feed content (7mg Cu/kg feed, 30 mg Zn/kg feed), and two levels of supplementation (2 or 9 mg Cu/kg feed and 35 or 95 mg Zn/kg feed).

The supplementations of Cu and Zn were given as sulphate (CuSO₄ and ZnSO₄) or as an amino acid – trace element – complex.

All supplementation levels and trace element sources were added either with or without phytase (700 units/kg feed).

The intake of Cu and Zn varied, mainly depending on the high of supply, from 997 mg Cu and 4296 mg Zn up to 3816 mg Cu respectively 29840 mg Zn per animal. The mean intake was 2176 ± 703 mg Cu respectively 15609 ± 7482 mg Zn per slaughtered animal. The analyses of variance did not show any influence of phytase supply on balance of the trace elements which are investigated (see Table).

Table: Intake, balance, excretion and carcass content of Cu and Zn depending on phytase supply (mg)

Phytase Supply	n	Intake		Balance		Excretion		Cont. per kg carcass	
		Cu	Zn	Cu	Zn	Cu	Zn	Cu	Zn
Yes	27	2135	15318	89	1873	2046	13445	0,86	18,20
No	26	2218	15911	87	1776	2131	14135	0,83	16,90

Influence of different non starch polysaccharide degrading feed enzymes on the intestinal microbiota in piglets

(*Einfluss verschiedener Nicht-Stärke-Polysaccharide spaltender Futterenzyme auf die intestinale Mikrobiota in Ferkeln*)

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The aim of this study was to compare two non starch polysaccharide (NSP) degrading enzyme preparations regarding their influence on several bacterial parameters in the intestine of fattening piglets. Weaned piglets (mean weight 7.6 kg +/- 1.3 kg) were fed a basal diet with 72% wheat and 10% wheat bran as the only source of NSP for 4 weeks either non supplemented (control) or supplemented with 100 mg/kg of a multi-enzyme preparation (3000 IE xylanase/g; 2800 IE glucanase/g; 140 IE arabinogalactanase/g, 190 IE galactomannanase/g, 160 IE cellulase/g) or with 200 mg/kg of a single enzyme preparation (3600 IE xylanase/g). Live weight of the animals was not significantly different, but feed conversion ratio tended (p = 0.1) to be improved in the enzyme supplemented groups. Intestinal viscosity (jejunum, colon) was reduced by both enzyme preparations. An inverse relationship was found for viscosity and bacterial metabolites in the colon. As highest amounts of volatile fatty acids and lactate were found in the monoenzyme group that displayed the lowest viscosity, followed by the multienzyme- and the control group, which displayed highest viscosity. The multienzyme preparation yielded the highest amounts of n-butyrate and n-valerate, while the monoenzyme preparation led to similar or higher amounts of the iso-forms of butyrate and valerate as the control group. Bacterial NSP degrading enzyme activities increased due to enzyme inclusion, most notably for lichenan (mixed linked glucan) in animals fed the monoenzyme preparation supplemented diet. Further results will be presented.

The results of this study point towards modifications of the intestinal microbiota due to enzyme supplementation. Fermentation of available substrates seemed most pronounced in the monoenzyme preparation, however composition of metabolites indicates a different microbiota depending on the nature of the supplemented enzyme.

Effect of calcium level, microbial phytase and citric acid on laying performance, bone mineralization and phosphorus disappearance in the digestive tract of laying hens fed corn soybean meal diets

(Einfluss von Kalziumgehalt, mikrobieller Phytase und Zitronensäure auf Legeleistung, Knochenmineralisierung und Phosphorverschwinden im Verdauungstrakt von Legehennen nach Verabreichung von Mais-Soja-Diäten)

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A 38 week trial was conducted to investigate the effect of calcium level, microbial phytase and citric acid on the performance parameters and bone mineralization in laying hens fed corn soybean meal diet. A total of 72 LOHMAN BROWN-CLASSIC laying hens, 23 wk of age were randomly assigned into a 2x2x2 factorial arrangement of treatments with two levels of calcium (2.6 and 3.8%), two levels of phytase (0 and 300 FTU/kg) and two levels of citric acid (0 and 2.5%). The phosphorus level was 0.33% tP (0.12% Non-phytate phosphorus) in all dietary treatments. After the performance trial was terminated, the digesta from the crop and the proximal small intestine were obtained from each hen and the total phosphorus disappearance was calculated using TiO₂ as an indigestible marker. Body weight, egg production, egg mass and feed conversion ratio were significantly reduced by feeding 2.6% calcium, adding microbial phytase did not prevent the adverse effect of low calcium diets (2.6%). Tibia ash and calcium in tibia ash were increased by adding microbial phytase. Feed conversion ratio, calcium and phosphorus in tibia ash were improved by adding citric acid, whereas body weight and feed intake were decreased. Phosphorus disappearance of the crop contents was reduced when hens were fed high calcium diets (3.8%) compared to hens which were fed low calcium diets (2.6%). Our study indicated that the major site of phytase activity (RONOZYME-P5000®) in the digestive tract of laying hens is in the crop. No significant effect of dietary treatments on phosphorus disappearance in the proximal small intestine was observed.

Calcium and phosphorus intake, apparent absorption, balance and normative requirement of adults – Are supplementations necessary?

(Verzehr, scheinbare Absorption, Bilanz und Bedarf von Calcium und Phosphor Erwachsener - Sind Supplementationen erforderlich?)

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With the duplicate portion technique and 10 test populations – each consisting of 7 women and 7 men at the age of 20 to 69 years – we estimated the calcium and phosphorus intake on 7 successive days. The calcium and phosphorus balance was measured in 3 test teams. On average, men took in 30% more calcium and phosphorus than women. Their increased consumption of both macro elements corresponds to their 24% higher dry matter intake. Habitation varies the calcium and phosphorus intake of both genders significantly by approximately 67% and 36%, respectively.

Each test team consumed significantly more phosphorus than calcium. On average, the calcium to phosphorus proportion was 1 : 1.58 varying between 1 : 2.10 and 1.31. The average daily calcium and phosphorus consumption amounted to 510 and 670 mg Ca/day, respectively, in women and 810 and 1060 mg P/day, respectively, in men. The frequency of the calcium and phosphorus intake in both genders follows a Gauss distribution. Lowest consumption on the average of a week was < 200 mg/day by women and < 400 mg/day by men for calcium, and < 400 and < 500 µg/day, respectively, for phosphorus. Highest consumptions amounted to 900 and 1000 mg Ca/day, and 1300 and 1800 mg P/day by women and men, respectively.

Calcium is mainly excreted through faeces (80%) and phosphorus through urine (20%). The apparent absorption rate of calcium in women was ± 0, that of men 16%. The apparent absorption rate of phosphorus amounted to 55% in women and 67% in men, respectively. The calcium balance of women was -24% and that of men -9%, the phosphorus balance of women amounted to -16% and that of men to -5%. With increasing age (> 40 years), the calcium balance of women becomes more negative than that of men. The apparent absorption rate of men is significantly higher than that of women. Osteoporosis is much more common in women than in men.

The normative calcium requirement of women is 500 mg/day, of breast-feeding women 750 mg/day, and of men 600 mg/day. Supplementation of young women with a calcium intake of 695 mg/day increased the faecal calcium excretion by the supplemented calcium amount. The calcium balance of young women with an intake of 500 mg Ca/day or 9 mg Ca/kg body weight is equalized.

Approximately 20% of women and 20% of men consume < 500 and 600 mg Ca/day on the average of a week, respectively. They need a calcium supplementation. The recommendation for a phosphorus consumption of 700 mg/day is satisfied in both genders of humans.

Impact of benzoic acid on early weaned piglets

(*Einfluss von Benzoesäure auf früh abgesetzte Ferkel*)

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The aim of the trial was to study the effectiveness of benzoic acid at the dietary inclusion level of 5 g per kg feed on piglet performance and on composition of the intestinal microbiota. In addition the tolerance of piglets to the feeding of benzoic acid at the 1% level was examined in a wheat based diet. A total of fifty four castrated male and female piglets were used from the 24th to the 66th day of live. The piglets were allocated to flat decks of 2 piglets per pen and were allotted equally according to the litters and gender. Each treatment consisted of 18 piglets.

Both inclusion levels of benzoic acids resulted in a significant higher hippuric acid concentration in urine as well as reduced urea concentrations in blood ($p < 0.05$) compared to controls. Urine pH, additional blood parameters, body weight, feed intake and feed conversion were not effected significantly.

However, up to 50% of colon and ileal digesta samples of piglets in the control group were found to harbor potentially enteropathogenic *Escherichia coli* O8, O138, and O141 serogroups, as determined by agglutination test of *E. coli* enriched cultures and purified isolates. In contrast, an up to 100% reduction in occurrence of the O138 and O141 serogroups was found in piglets belonging to the groups receiving benzoic acid. Using denaturing gradient gel electrophoresis (DGGE) of 16S rDNA-amplicons the dominant eubacterial colon and ileal microbiota and the *Lactobacillus* spp. community, respectively, were compared between all treatment groups. Treatment dependent variations in community structure based on intra- and intergroup similarity and microbial diversity as revealed by DGGE banding patterns as well as the overall richness of the microbiota in digesta samples will be discussed.

Addition of inulin to diets low or high in NSP does not stimulate bifidobacteria and lactobacilli in the intestine of growing pigs.

(*Inulin in einer NSP-armen und NSP-reichen Diät fördert nicht das Populationswachstum von Bifidobakterien und Laktobazillen im Darmtrakt von wachsenden Schweinen*)

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The microbiota of the human and porcine gastrointestinal tract is a complex ecosystem consisting of more than 400 bacterial species. Interest is now focused on modulation of bacterial composition to stimulate health-beneficial bacteria strains by feeding prebiotics. Inulin largely escapes digestion in the upper part of the human intestine, reaching the large intestine unfermented, and thus stimulates bifidobacteria and lactobacilli in the colon. These bacteria strains are thought to be beneficial for the host. The future ban of antibiotics as growth promoters in farm animals fuels the search for alternatives. To explore possible changes in the bacterial composition growing pigs ($n=8$ /group) were fed two diets with different NSP levels (CD, 16%; SD 1%) with or without 3% inulin over a period of 21 and 42 days, respectively. Fluorescence-*In-Situ*-Hybridization (FISH) using 16S rRNA-based oligonucleotide probes and *Denaturing Gradient Gel Electrophoresis* (DGGE) by using group-specific PCR primers were performed to detect luminal and gut wall-associated bacteria, respectively, in the small and large intestine. We found that nearly 30 - 50% of inulin was fermented in the small intestine. Inulin did not influence luminal bifidobacteria and lactobacilli counts in jejunum and colon ($P>0.06$). A masking effect of NSP level on the bifidogenic impact of inulin could be excluded. Within the gut wall-associated lactobacilli strains *Lactobacillus amylovorus* seemed to be a dominant species. In conclusion, a largely individual intestinal microbiota hampered the verification of diet-related effects.

Effects of a probiotic *Lactobacillus acidophilus* strain on food tolerance in dogs with non-specific dietary hypersensitivity

(Effekte eines probiotischen *Lactobacillus acidophilus* Stammes auf die Futtermitteltoleranz von Hunden mit unspezifischer Futtermittelsensitivität)

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The objective of this study was to investigate the effects of a probiotic *L. acidophilus* strain DSM 13241 on food tolerance in dogs with non-specific dietary sensitivity.

6 adult German Shorthair Pointers (4 males, 2 females, body weight 30.8±2.0 kg) with a history of non-specific dietary sensitivity were fed a control dry diet, the same diet with 6 x 10⁶ colony forming units (CFU)/g *L. acidophilus* DSM 13241 for 12 weeks each and the control diet for additional 4 weeks. Frequency of defecations, scoring of fecal quality, fecal dry matter and fecal unbound water were determined. Fecal samples were cultured for *Clostridium perfringens*, *Escherichia coli*, *Lactobacillus spp.* and *Bifidobacterium spp.* *In situ* hybridization was performed in fecal samples for *Clostridium histolyticum*, *E. coli*, lactobacilli and bifidobacteria. The digestibility of test and control diets was also assessed. Statistics were performed by ANOVA and Student's t-Test, p<0.05.

Feeding *L. acidophilus* DSM 13241 led to a slightly lower frequency of defecations, slightly improved fecal consistency and increased fecal dry matter (p<0.05). It resulted in higher fecal numbers of *Lactobacillus spp.* and *Bifidobacterium spp.* and in a slightly decreased number of *C. perfringens* and *E. coli* as measured by selective bacterial culture. Fluorescence *in situ* hybridization showed less variations in the bacterial concentrations. Digestibility of crude protein and crude fat was higher during the probiotic intake.

This study showed that *L. acidophilus* DSM 13241 can stabilize the digestive processes in dogs with non-specific dietary sensitivity. The observed improvement in fecal consistency would have considerable practical importance, since *L. acidophilus* probiotics are generally regarded as safe (GRAS status) and as such are acceptable for long-term application.

Influence of different pre-, pro- and synbiotics on the digestibility processes in pigs

(Zum Einfluss verschiedener Prae-, Pro- und Synbiotika auf die Verdauungsvorgänge beim Schwein.)

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Pre-, pro- and synbiotics are increasingly focussed in pig nutrition. Therefore the aim of the present studies was on one hand to provide basic data about the precaecal and faecal digestibility of pre- and synbiotics and on the other hand to investigate the influence of pre-, pro- and synbiotics on nutrient digestibility and microbial parameters (bacterial counts, pH, lactic acid (LA), volatile fatty acids (VFA), NH₃) in the digestive tract.

A series of metabolic trials was carried out with 22 growing male castrated pigs. 11 pigs were fitted with an end-to-end ileorectal anastomosis with preserved ileo-caeco-colic valve; the remaining pigs were used as intact partners. A basic diet consisting mainly of corn, wheat, barley and soybean meal (per kg 13.4 MJ ME, 150 g CP) was offered in amounts of 1500 to 1900 g/pig/d. The pre-, pro- and synbiotic feed additives were supplemented as follows (per kg): lactulose (LAC I: 1.5%; LAC II: 2%), inulin (INU I: long-chain; INU II: middle-chain; 2%, resp.), *Enterococcus faecium* (E.f. I: DSM 10663, 8x10⁹ CFU; E.f. II: DSM 7134, 5x10⁸ CFU), Mannanligosaccharides (MOS; 0.3%) and the synbiotic combination INU II + E.f. I and MOS + E.f. II. The diets containing the test substances were offered for periods of 12 – 40 days.

The precaecal digestibility of the prebiotics was 79.4% (LAC I), 76.9% (LAC II), 97.7% (INU I), 57.3 (INU II), and 55.5 (INU II + E.f. I). All prebiotics were completely digested faecally. Nutrient digestibility and bacterial counts, pH, LA, VFA, and NH₃ were widely unaffected by the different supplemented substances.

Summarizing, the selected prebiotics are digested and absorbed by pigs to a considerable degree (lactulose, middle chain inulin) or even completely (long chain inulin) in the ileum, regardless of the addition of a probiotic. Because of the high amounts of prebiotic components in usual pig feed additional beneficial effects of a supplementation are questionable. In the presented experiments the proof of positive effects of pre-, pro- and synbiotics in pig nutrition was only partly successful.

Modifying gut flora in pigs by phytochemicals

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Regulation 1831/2003 (EC) lays down provisions phasing out the authorisations of antimicrobial growth promoters in animal feed (AMGP's) in the EU as from January 2006. The antimicrobial properties of certain bioactive chemicals (phytochemicals) have been described extensively. However, there is little quantitative data on their antimicrobial activity against the normal gut flora of the pig. In this research, the *in vitro* antimicrobial activity of 7 pure phytochemicals (monoterpenes; carvacrol, thymol, eucalyptol and terpinen-4-ol; phenylpropenes; trans-anethol, eugenol and cinnamaldehyde) against the main components of the pig gut flora has been evaluated. The duration of the *in vitro* incubation was 3, 4 and 24h for simulation of gastric, small intestinal and caecal digestion respectively, allowing an exponential growth of the bacteria. A control (0 ppm in medium) and three concentrations were tested per botanical.

Carvacrol, cinnamaldehyde, thymol and eugenol give opportunities to modulate the flora of the gastrointestinal tract (GIT) of pigs. Their antimicrobial effect varies according to the site and bacterial group considered. Eugenol had no effect in gastric conditions, but was very effective in small intestinal conditions. In caecal conditions, it reduced only *E. coli*. Cinnamaldehyde showed a low antimicrobial activity in gastric conditions (pH 3). The use of cinnamaldehyde and eugenol can result in a higher lactobacilli/*E. coli* ratio in the GIT. Thymol and carvacrol lowered significantly total anaerobic bacteria, lactobacilli, streptococci and *E. coli* in all parts of the GIT. The phytochemicals in the incubation medium have been quantified by a GC-method. Data on microbial metabolization of the phytochemicals during incubation are given.

[V1] Essential fatty acids and tocopherols of evening primrose (*Oenothera biennis* L.) oil

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The possibility of production and use of γ -linoleic acid (GLA) is the object of extensive investigations lately. Namely, new evidence on the nutritive value of this oil has been stated on the basis of investigations of multidisciplinary teams of scientists and experts (nutritionists, biochemists, biophysicists, physiologists etc.). Besides vitamins, minerals and other functional components, certain fatty acids have an important role in everyday diet also. GLA is essential for persons with disturbed metabolism, where the transformation of α -linoleic to γ -linoleic acid is impossible. This depression of bio-conversion is especially expressed under stress. In such cases, the direct intake of GLA is recommended. The paper points to the nutritive significance of evening primrose oil. The parameters which characterize the seed and quality of oil, including the fatty acid composition were investigated (determined), the content and composition of tocopherols and stability of oil. The fatty acid composition was determined by GC method and the dominant fatty acids are: palmitic, stearic, oleic, linoleic and γ -linoleic with 6,83% in pressed and 2,85% in the extracted oil. The tocopherols were determined by HPLC method, and their content was 282 mg/kg. The oxidative stability of this oil is relatively poor. The induction period of pressed oil, determined by Rancimat test, at 100°C, was 8.5 hours.

[V2]

Bioactive compounds in tomatoes and tomato products – investigations of different commercially available foodstuffs*(Bioaktive Substanzen in Tomaten und Tomatenprodukten – Untersuchungen verschiedener kommerziell erhältlicher Lebensmittel)*Fröhlich Kati, Burkhardt Susanne, Böhm V

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Tomatoes and tomato products are very popular in Germany and contribute for 22% to the total vegetable consumption. In 2003, consumption of tomatoes and tomato products per head and year was 18.6 kg (raw tomatoes 8 kg). In addition to carotenoids, tomatoes deliver considerable amounts of vitamin C and tocopherols. Recent epidemiological studies indicated that these phytochemicals may play a number of key health-promoting roles in the human body. Animal model studies, together with epidemiologic observations, suggested that tomato consumption may have organ-specific chemopreventive effects on some types of cancer and cardiovascular diseases. The non pro-vitamin A active carotene lycopene is one of the major phytochemicals in tomatoes contributing to the anti-carcinogenic function. Tomatoes are consumed mainly as processed tomatoes. Homogenization as well as heat treatment of tomatoes disrupt cell membranes, with heat treatment being suggested to disrupt also the protein-carotenoid-complex. Therefore, processing increased the bioavailability of carotenoids in humans. In contrast, processing of tomatoes also led to significantly decreased vitamin C concentrations in the products. Aim of the research was to determine contents of carotenoids and vitamin C in samples of tomatoes, tomato juice, tomato ketchup, tomato sauces, sieved tomatoes (tetrapack) and pizza tomatoes (tin), reflecting the product diversity on the German market.

Total lycopene concentrations ranged between 4.1 mg/100 g and 24.8 mg/100 g and contents of β -carotene were 0.1-1.8 mg/100 g. Vitamin C was analysed in amounts of 0.32-42.3 mg/100 g. The results indicated that raw tomatoes have lower total lycopene concentrations compared to tomato products due to dehydration during the processing. Regarding β -carotene, raw tomatoes showed the highest concentrations, indicating that β -carotene is less resistant against homogenization and heat treatment than lycopene. Compared to untreated tomatoes, processed products contained lower contents of vitamin C due to its lability.

[V3]

Vitamin C and antioxidative potential of oranges and other citrus fruits*(Vitamin C und antioxidatives Potential von Orangen und anderen Zitrusfrüchten)*Gutte Josefine, Burkhardt Susanne, Böhm V

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Dietary antioxidants, found in fruits and vegetables, might prevent oxidative damage in humans which is one cause for cancer. Aim of this investigation was to determine the contents of vitamin C, total phenolics and antioxidant capacity of different citrus fruits as well as the respective contribution of vitamin C to the sum parameters. In addition, the effect of frozen storage at -18°C on the content of vitamin C was ascertained. Edible parts of 12 samples of citrus fruits from different food markets, with nine oranges, one lemon, one lime and one grapefruit, were homogenized and analysed after a frozen storage period of for 14 months, vitamin C also prior to that. Vitamin C was determined photometrically using 2,4-dinitrophenylhydrazine. The content of total phenolics was also assessed photometrically by using the Folin-Ciocalteu method. Antioxidant capacity was investigated by using the ferric reducing antioxidant power (FRAP) assay and the Trolox-equivalent antioxidant capacity (TEAC) assay. Oranges had the highest vitamin C content (46-72 mg/100 g) compared to the other investigated citrus fruits, lime the lowest (37 mg/100 g). After 14 months of frozen storage only two orange samples showed a significant decrease in vitamin C-content. Total phenolics content expressed as gallic acid equivalents (GAE) was similar in all tested fruits (61–81 mg GAE/100 g). Especially in grapefruit there was a high contribution of phenolic compounds other than vitamin C to the total phenolics content (70-77%). The highest antioxidant capacity was determined for lemon (FRAP: 0.71 mmol/100 g, TEAC: 0.35 mmol/100 g), the lowest one for lime (FRAP: 0.46 mmol/100 g, TEAC: 0.21 mmol/100 g). In oranges at least 78% of the antioxidant capacity is caused by vitamin C. There is a significant correlation between the values of the two antioxidant capacity assays ($r=0.922$, $p<0.01$) whereas only low relationships between vitamin C, total phenolics content and antioxidant capacity were found. Thus, citrus fruits enrich human nutrition, especially with regard to the prevention of diseases resulting from oxidative damage.

[V4]

Urinary excretion of antioxidants following consumption of *Hibiscus sabdariffa* L. extract*(Exkretion von Antioxidantien im Urin nach Zufuhr von Hibiscus sabdariffa L. Extrakt)*Netzel¹ M, Straß^{1,2} Gabriele, Bitsch² Irmgard, Kler³ A, Kries³ E, Kammerer⁴ DR, Schieber⁴ A, Caille⁴ R, Frank⁵ T, Bitsch¹ R¹Institute of Nutrition, Friedrich-Schiller-University, Dornburger Strasse 29, D-07743 Jena, Germany; ²Institute of Nutrition, Justus-Liebig-University, Wilhelm Strasse 20, D-35392 Giessen, Germany; ³Plantextrakt GmbH & Co KG, Dutendorfer Strasse 5-7, D-91487 Vestenbergsgreuth; Germany; ⁴Institute of Food Technology, Section Plant Foodstuff Technology, Hohenheim University, August-von-Hartmann-Strasse 3, D-70599 Stuttgart, Germany; ⁵Solvay Pharmaceuticals GmbH, Hans-Boeckler-Allee 20, D-30173 Hannover, Germany

Hibiscus sabdariffa L. is a traditional Chinese rose tea and is used effectively in native medicines against hypertension, inflammation, and liver disorders. The flowers of *Hibiscus* species contain several antioxidant compounds like phenolic acids, flavonoids, and especially anthocyanins. The aim of the present pilot study was to assess the renal excretion of *Hibiscus sabdariffa* L. extract (HSE) antioxidants in one healthy volunteer. After a single oral dose of 200 ml of HSE (217 mg anthocyanins and 263 mg total phenolics, equivalent to an antioxidant capacity of 3.20 mmol [FRAP assay], 2.03 mmol [PCL assay], and 3.30 mmol [TEAC assay]), the parameters obtained in urine were compared to those obtained after the ingestion of water (control). Urine was collected in intervals up to 24 h post-intake. After HSE ingestion, delphinidin-3-sambubioside, cyanidin-3-sambubioside, and delphinidin-3-monoglucuronide were excreted in the volunteer's urine (0.031 % of the administered dose) and unambiguously identified by HPLC-ESI-MS/MS. Compared to the ingestion of water, HSE consumption resulted in an increased urinary excretion of total phenolics (+12%), other antioxidant compounds (+13% [FRAP assay], +22% [PCL assay], +36% [TEAC assay]), and hippuric acid (potential colon metabolite; +104%) within 24 h. In conclusion, the results of this pilot study may indicate an *in vivo* antioxidant effect of *Hibiscus* antioxidants in healthy humans.

[V5]

Oxidative stability of virgin olive and high oleic sunflower oilVuksa Vjera¹, Dimic Etelka¹, Tesanovic D², Romanic R¹¹Faculty of Technology, 21000 Novi Sad, Bul. Cara Lazara 1, Serbia and Montenegro²Faculty of Natural Sciences, 21000 Novi Sad, Trg Dositeja Obradovica 3, Serbia and Montenegro

Since some lipids affect the human organism in a specific way, the interest of experts and of consumers for their use is increasing, in order to maintain or improve the human health. According to this, the demands of contemporary consumers are directed at natural »cold pressed oils which contain more in original specific nutritive components like vitamins, sterols, carotenoids, phospholipids etc.

Olive oil, as the typical representative of cold pressed oils is in use for the longest period. However, the interest for the natural edible nonrefined oils and introduction of new raw materials - oilseeds is increasing. Special attention is paid to high-oleic sunflower oil, since the fatty acid profile is very similar to olive oil.

All edible vegetable oils are more or less prone to oxidative changes, and as the result, harmful free radicals are formed. For that reason, the nutritive value and oxidative stability of the mentioned two kinds of oils were compared. The investigations showed that cold pressed high-oleic sunflower oil (oleic acid > 80%), regarding the primary oxidative products and FFA, is of better quality compared to extra quality virgin olive oil. The vitamin E content was higher in sunflower oil (about 800 mg/kg) compared to olive oil (80 – 280 mg/kg). However, the oxidative stability of olive oil is higher: the induction period at 120°C is about 7 hours, and for sunflower oil only 4 hours.

[V6]

Antioxidative status and meat quality of pigs influenced by dietary supplementation of vitamin E and by post mortem injection of calcium ascorbate

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Vitamin E and C are primary antioxidants in biological systems and break the chain of lipid peroxidation. The objective of this study was to evaluate further the effects of dietary vitamin E supplementation and calcium ascorbate injected in the muscle postmortem on level of alpha-tocopherol, ascorbic acid and calcium in muscle, antioxidant capacity and parameters of meat quality.

Treatment consisted of supplementation of vitamin E (500 mg alpha-tocopherylacetate/kg feed) for the minimum 30 days to finishing pigs before slaughtering. Pieces of meat (longissimus dorsi) were injected with 10% by weight of 1.5% calcium ascorbate solution. Dietary supplementation of fat soluble antioxidant vitamin E to grow-finishing pigs significantly ($P < 0.05$) increases the concentration of alpha-tocopherol in meat (longissimus dorsi). Meat quality parameters (drip loss, pH) were positively influenced. Lipid oxidation measured as TBARS (MDA) and antiperoxidative capacity (Fe^{2+} /ascorbate induced) of meat were significantly ($P < 0.05$) influenced by supplementation of vitamin E to grow-finishing pigs. Water soluble antioxidant calcium ascorbate injected in the meat increases the concentration of calcium ($P < 0.05$) and ascorbic acid ($P < 0.05$), stabilized color ("a" value) in chill-stored meat and improved ($P < 0.05$) antiperoxidative capacity (Fe^{2+} /ascorbate induced). Further research is needed to optimize the dosage.

[V7]

Utilisation of flavonoids in black currant seeds

(*Verwertung von Flavonoiden in schwarzen Johannisbeersamen*)

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To arrive of conclusions for the use of flavonoids and respectively flavonoid extracts (e.g. bakery products) the analysis of interventional studies (in vitro/ in vivo) is an essential informative basis.

In the context of the research project a new method using the capillary electrophoresis technique was developed to determine as many black currant seed flavonoids as possible in one run.

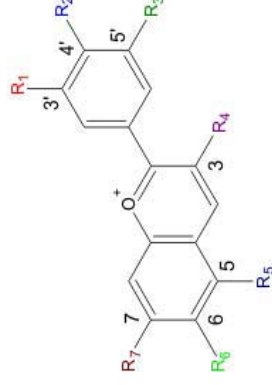


Figure: basic structure anthocyan: flavylium cation

Anthocyanidin	R ₁	R ₂	R ₃	R ₄	R ₅	R ₆	R ₇
Delphinidin	OH	OH	OH	OH	OH	H	OH
Cyanidin	OH	OH	H	OH	OH	H	OH



[V8]

Cell protective effectsHealth promoting effects of wheat bran arabinoxylans – *in vitro* investigations with human colon cells

(Zellschützende Wirkungen von Arabinoxylanen der Weizenkleie – *in vitro* Untersuchungen mit humanen Kolonzellen)

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Arabinoxylans are the most important dietary fibre components of wheat bran. It was the aim of this study to characterise chemoprotective effects of aqueous solutions of wheat bran fractions, namely water extractable (WeAx) and alkali extractable arabinoxylans (AeAx), and their products formed during the gut-flora-mediated fermentation in human colon cells. For this WeAx and AeAx were isolated from wheat bran, dissolved in cell culture medium or fermented with human faeces to produce fermentation supernatants (FS), which were analysed for content of short chain fatty acids (SCFA) using HPLC. Human colon HT29 cells were treated with the samples and we investigated possible antiproliferative activities by quantifying residual DNA with a fluorescent dye. Viability was determined by trypan blue exclusion and DNA damage using the comet assay, to assess possible antioxidant and antigenotoxic effects. The glutathione S-transferase (GST) activity was determined spectrophotometrically. Key results were that the fermentation of the arabinoxylans resulted in a significant increase of SCFA. Growth of the HT29 cells was efficiently inhibited by all FS including the control. A short term pre-incubation with WeAx decreased the level of H₂O₂ (75 µM) induced DNA damage significantly by 67 %. Fermented WeAx or AeAx were less protective. In contrast, all fermented samples reduced the genotoxicity of the lipid peroxidation product HNE (200 µM). We measured a significant induction in GST activity, which may point to an improved detoxification of HNE. Our results indicated that wheat bran ingredients before and after fermentation may act antiproliferativ and antigenotoxic, thus demonstrating chemoprotective properties in human colon cells. This research project was supported by the FEI (Forschungskreis der Ernährungsindustrie e.V., Bonn), the AIF and the Ministry of Economics and Labour. AIF-FV 13065 BG.

[V9]

Carob fibre and Gallic Acid – Impact on proliferation parameters in human colon cell lines

(Carob und Gallensäure – Einfluss auf Parameter der Proliferation in humanen Kolonzelllinien)

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Introduction: Carob fibre is a food ingredient from the Mediterranean carob pod (*Ceratonia siliqua* L.). During a mild procedure soluble carob constituents are mainly removed by water extraction and the insoluble dietary fibre is retained in the residue, in addition to tannins and other polyphenols. Carob fibre was investigated for its ability to modulate a variety of different cellular parameters of proliferation using the human colon carcinoma cell line HT29 and the human colon adenoma cell line LT97. **Methods:** For this, an aqueous extract of carob fibre was prepared (100 g carob fibre per 1 litre cell culture medium) and cells were incubated with 0.05 g to 20 g carob fibre per 1 litre cell culture medium. After 12, 24, 48 and 72 h treatment, growth and survival of the cells were measured by determining metabolic activity (reduction of resazurin) and cell number (incorporation of fluorochrome DAPI into DNA of remaining cells). Results were calculated in percentage of the medium control and the metabolic activity is based on the cell number. **Results:** The aqueous extract effectively decreased number of HT29 cells (from 100% in the control to 8 ± 7 % after 72 h with 20 g/l carob fibre). The effects were associated with dose and time. The reduction of metabolic activity was particularly apparent after 12 and 24 hours treatment (up to 32% after 12 h with 2 g/l carob fibre), but was only temporary. In contrast, cell number of LT97 cultures was less impaired by the treatment with equal amounts of carob extract (70 ± 6 % of the medium control after 72 h with 20 g/l carob fibre) and metabolic activity correlated highly with cell number. **Conclusion:** Carob fibre modulates the growth of both colon carcinoma and colon adenoma cells. The impact of the complex aqueous extract seems to depend on growth kinetics of the cell line. Highly proliferating carcinoma cells were more susceptible to the growth inhibitory properties of this plant extract, than cells with a lower cell turn over.

[V10]

The effect of apple polyphenol extracts on proliferation of colon adenoma (LT97) and carcinoma (HT29) cells

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Introduction: Epidemiological findings suggest that plant foods decrease colorectal tumour risks. This could be due to a number of different phytoprotectants which act chemopreventive by inhibiting the growth of tumour cells. Although apples are rich in such phytochemicals (polyphenols), no attempt has been made to identify the relevant bioactive component(s). **Aim:** We evaluated antiproliferative properties of apple polyphenols in colon adenoma (LT97) and carcinoma (HT29) cells to assess potential chemoprotective activities. **Material and Methods:** We have used different apple polyphenol extracts "AE" (samples 1, 2 and 3) from different cultivars (2002-2004), which were characterized for their polyphenols and flavonoids by HPLC. LT97 and HT29 cells were first grown in 96-well plates for 72 and 48 h and then incubated with either medium alone (control) or apple extracts. The concentrations of AE were ranged from 8.5 – 85 µg (dry matter: equivalent to 5 – 50 µM of phloridzin). Cell growth was determined indirectly by measuring the total cell-DNA staining with the flouochrome 4', 6-Diamidino-2-phenylindole (DAPI) after 24, 48 and 72 h. **Results and Conclusions:** All three AE contained high and comparable total amounts of polyphenols (478-533 mg/g). The comparison of the three AE samples revealed that sample 3 contains the highest amounts of quercetin derivatives and phloridzin (5.3 fold more than samples 1 and 2), probably due to the different qualities of the apple cultivars. Associated with this was the finding that sample 3 inhibited the growth of colon cells more than the other two samples. The LT97 adenoma and preneoplastic cells were more sensitive than the rapidly proliferating tumor-derived cells HT29. This suggests that AE contain polyphenols which suppress growth of adenoma and tumor cells, and thus may act chemoprotective, especially in the earlier stages of tumor development.

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[V11]

The changes of the vitamin C concentration in the serum of patients with skin cancer

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The aim of the study was to estimate the changes of the vitamin C concentration in the serum of patients with non-melanomatous skin cancers (NMSC).
Material and methods. The studies included 113 patients (52 men and 61 women) aged 32 to 84 years, treated because of NMSC. In all of them the diagnosis of the neoplasm was confirmed histopathologically. In 76 patients a basal cell cancer was diagnosed, in the remaining 37 – a spinocellular cancer. In this group of patients, as well as in the two others: a reference group (RG) – blood donors (36 persons), and a control group (CG) – 70 patients with non-neoplastic skin diseases (alopecia areata, lichen planus or with common warts) were conducted the determinations of the vitamin C concentration in the blood serum with the chromatographic method. The statistical analysis of the results obtained was conducted using the SAS programme, t-Student's test and Wicoxon test. All the decision were taken on the critical level p<0.05.

Results and conclusions. The vitamin C reference value (RG) in the serum was 0.91±0.18 mg/dl. The vitamin C concentration in the serum of patients with NMSC varied from 0.38 to 1.42 mg/dl, in the CG – from 0.52 to 1.24 mg/dl, whereas in the RG from 0.80 to 1.22 mg/dl.

Following the studies carried out, it was confirmed that in patients with NMSC a statistically significant decrease occurs of the vitamin C concentration in the serum in comparison with the RG and the CG. Yet these deficits mainly concern the cancer of the skin of the face and of the neck, as well as the cancer of the skin of the limbs.

[V12]

Excretion of polyphenols and metabolites in human urine following consumption of apple juice*(Exkretion von Polyphenolen und Metaboliten im Urin nach Zufuhr von Apfelsaft)*Netzel¹ M, Straß^{1,2} Gabriele, Roßberg¹ Anke, Bitsch² Irmgard, Thielen³ Christine, Dietrich³ H, Kammerer⁴ DR, Schieber⁴ A, Carle⁴ R, Frank⁵ T, Bitsch¹ R¹Institute of Nutrition, Friedrich-Schiller-University, Dornburger Strasse 29, D-07743 Jena, Germany²Institute of Nutrition, Justus-Liebig-University, Wilhelm Strasse 20, D-35392 Giessen, Germany³Institute of Enology and Beverage Research, Research Institute of Geisenheim, Rudesheimer Strasse 28, D-65366 Geisenheim, Germany⁴Institute of Food Technology, Section Plant Foodstuff Technology, Hohenheim University, August-von-Hartmann-Strasse 3, D-70599 Stuttgart, Germany⁵Salvay Pharmaceuticals GmbH, Hans-Boeckler-Allee 20, D-30173 Hannover, Germany

In the German diet important sources of polyphenols are fruit juices. Apple juice is still the undisputed leader among the fruit juices with a per capita consumption of approximately 13 L / year (VdF 2003). The aim of the present study was to assess the urinary excretion of apple/ apple juice polyphenols as well as of their potential metabolites in eight healthy humans. After a single oral dose of 700 mL of apple juice, produced from the cultivar *Weisser-Trierer-Weinapfel (WTW)*; containing 1107 mg total phenolics, 330 mg chlorogenic acid, 69 mg phloridzin, 9 mg epicatechin, 7 mg phloretin xyloglucoside and 4 mg coumaroylquinic acid), the apple polyphenols were recovered predominantly as metabolites (glucuronides) in the volunteers' urine. Compared to the ingestion of water (control treatment), *WTW* juice consumption resulted in a significantly ($p < 0.05$) increased urinary excretion of total phenolics (+ 36%) and hippuric acid (+ 210%); potential colon metabolite) within 24 h. In conclusion, these metabolites (glucuronides and hippuric acid), and not the native apple polyphenols, may be responsible for at least some of the health effects attributed to apple/ apple juice consumption.

[V13]

Effect of vitamin E and polyphenols on ochratoxin A-induced cytotoxicity in liver (HepG2) cellsHundhausen C^a, Bösch-Saadatmandi Christine^a, Augustin K^a, Blank R^b, Wolfram S^b, Rimbach G^a^aInstitute of Human Nutrition and Food Science, Christian-Albrechts-University of Kiel, Hermann-Rodewald-Str.6, 24098 Kiel, Germany^bInstitute of Animal Nutrition and Physiology, Christian-Albrechts-University of Kiel, 24098 Kiel, Hermann-Rodewald-Str.9, 24098 Kiel, Germany

It has been shown that oxidative damage contributes to the wide range of toxic effects of the mycotoxin ochratoxin A (OTA). Therefore, we examined the effects of α -tocopherol (α -TOC) and different polyphenols – catechin (CAT), daidzein (DAI), epicatechin (EC), epigallocatechin gallate (EGCG), genistein (GEN), and quercetin (QUE) – on OTA-induced cytotoxicity in HepG2 liver cells. Incubation of HepG2 cells with increasing concentrations of OTA resulted in a dose- and time-dependent cytotoxicity as measured by the neutral red assay. Half lethal concentrations (LC50) of OTA were 35 and 10 μ M after 48 and 72 h incubation, respectively. Incubation of HepG2 cells with α -TOC as well as with different polyphenols (exhibiting different antioxidant potency as determined by the FRAP, TEAC and DPPH assays) did not counteract OTA-induced cytotoxicity. These findings indicate that OTA may exert its toxic effects by affecting other hepatic mechanisms than those directly modulated by α -TOC and polyphenols.

[V14]

Risk assessment of vitamins and minerals in foods*(Risikobewertung von Vitaminen und Mineralstoffen in Lebensmitteln)*Grossklaus R, Domke A, Niemann B, Przyrembel H, Richter K, Schmidt E, Weißenborn A, Ziegenhagen R

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The BfR is involved in the risk assessment of vitamins and minerals, including trace elements, in foods. A two-volume report is now available in the series "BfR Wissenschaft" (1, 2). It is intended as a basis for discussion and decision-making for risk managers in Germany and the European Union when setting maximum levels for nutrients in food supplements and fortified foods. The aim of the poster is to provide an insight into the principles of risk assessment of vitamins and minerals including trace elements. Risk assessment of nutrients differs somewhat from that of chemical residues or contaminants in foods. When setting maximum levels for nutrients in foods, both the safe daily intakes of a vitamin and mineral (so-called tolerable upper intake levels) and the uptake of these nutrients from common foods by the population must be taken into account, as well as the recommended daily intakes for a nutrient. Tolerable upper intake levels have been derived or defined by various scientific bodies. The BfR endeavours to identify possible methods of setting maximum levels of vitamins and minerals in a single food or food supplement. Such maximum levels should allow supplementation levels sufficient for correcting nutrient deficits in population groups without exceeding the tolerable daily intake levels (to a major degree). Depending on the desired level of protection, various options for setting maximum levels are outlined in this two-volume report and the respective advantages and disadvantages are discussed. The poster will give some examples of the quantitative risk assessment of vitamins and minerals for the derivation of maximum levels in individual foods.

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[V15]

Vitamins and Calcium, as Additives in milk /cow/*(Vitamine und Kalzium als Zusatzstoffe in Milch (Kuh))*Meilikia E, Kvanchakhadze R, Sekhniashvili N, Tsereteli D, Sekhniashvili Z

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National Center of Nutrition in Georgia in 1998-2004 has implemented the state prophylactic program investigation of the caused diseases as a result of insufficiency of the microelements (J, Fe, F etc.), mineral substances and vitamins deficiency. The result of the investigation had showed that the vitamins deficiency among population (is more than 50% and calcium –more than 25%) is widely spread. The relative risk of the Avitaminose development is 2.5; it means that it's higher among urban population than among rural. In Georgia such diseases as Atherosclerosis, Osteoporosis, Rachitis and others are caused by the vitamins and calcium deficiency.

The Children, suckling mothers and pregnant women are especially considered to the sensitive group relative to the calcium and vitamins deficiency.

A sufficient quantity of calcium and vitamins forms therefore the base prophylaxis against Avitaminosis, Rachitis, Atherosclerosis, Osteoporosis and other illnesses.

The fortified cow's milk began 3 years ago, the price of the abovementioned milks is accessible to the population.

Milk is a necessary product for the health, especially for children, pregnant and suckling mothers. The most important advantage at the milk is that it contains vitally important nutrients such as: proteins, fat, coal hydrates, mineral substances which are easily absorbed in human organism. In the process of pasteurization the following vitamins: A, B₁; B₂; B₃; B₆; B₁₂; B_c; E; PP; H and C are added to the cow's milk. The richness of the vitaminised milk is 2.5%, the density is 1027. The acid degree is 0^oT –20.

100gr milk contains the following vitamins: A-0.25ME; D-0.25ME; E-1.9 mg; B₁-0.35 mg; B₂-0.35 mg; B₃(niacin)-1.75mg; B₆-0.4 mg; B₁₂-0.75; B_c(folacin)-100 mikg, PP-4.5 mg; H-50.0; C-15.0.

CaHPO₄ is also added to the fresh and pasteurized cow's milk. In 100gr cow's milk fortified by calcium the calcium substitute is 0.5%; the calcium's density in 300mg amounts to 1028 and the acid degree is 0^oT-19.

The preliminary data of the investigations carried out among population those who were given calcium and vitamins fortified cow's milk showed positive effects.

[V16]

Stability of RRR- α -Tocopheryl-Acetate in premixes and feed processingPeisker, M., Dersjant-Li Yueming

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A comparative study was carried out to assess the stability of natural (*RRR- α -Tocopherol*) and synthetic (*all-rac- α -Tocopherol*) vitamin E preparations in acetate form. Two premixes (standard; aggressive with elevated levels of Cu, choline, salt) were prepared with the two vitamin preparations. The standard premix was added to a piglet feed in mash form, which was pelleted (75°C) and extruded (90, 100, 110°C). During six months, storage stability at ambient (20°C) and elevated (40°C) temperature was tested for the following treatments: 1-standard premix, 2-aggressive premix, 3-mash feed, 4-pelleted feed, 5-7 extruded feed (90, 100, 110°C).

Manufacturing stability: The vitamin E recovery after pelleting was 100%. The extrusion process, however, reduced vitamin E recovery by about 40%. Extrusion temperature did not impact on the extent of *α -Tocopherol* losses in the process. No differences in processing stability have been observed between natural and synthetic vitamin E preparations.

Storage stability: Vitamin E in acetate form was stable in the tested period of 6 months. Neither premix type (standard, aggressive) nor storage temperature (20°C, 40°C) had an effect on storage stability. No differences were observed on storage stability between natural and synthetic vitamin E preparations during the 6 months period. The averaged data for mash, pellets and extruded feeds seem to indicate that mash and pellets are more stable than extruded feeds after 6 months of storage.

In conclusion, the processing and storage stability in premixes and feeds supplemented with natural vitamin E (*RRR- α -Tocopherol*) is the same as for synthetic vitamin E (*all-rac- α -Tocopherol*) when the products are in acetate form.

[V17]

Microwave-vacuum dried strawberries and their contribution to a healthy diet*(Mikrowellen-Vakuum-getrocknete Erdbeeren und ihr Beitrag zu einer gesunden Ernährung)*Bellmann Susann¹, Kühnert S², Rohm H², Scholze G³, Böhm V¹¹ Institute of Nutrition, Friedrich Schiller University Jena, 07743 Jena, Germany,² Institute of Food Technology and Bioprocess Engineering, Dresden University of Technology, Dresden, Germany,³ Zittauer Fruchtveredlungs GmbH, Zittau, Germany

Aim of the presented study was to investigate the intestinal absorption of vitamin C and polyphenolics as well as the antioxidant capacity of plasma and urine samples after consumption of microwave-vacuum dried strawberries compared to raw strawberries. Prior to this intervention study the drying technique was optimised in order to reduce the loss of vitamins and antioxidant compounds during processing.

Eight healthy adults consumed 600 g frozen strawberries and 50 g dried fruits using a cross-over design. Blood samples were taken prior to the intervention and 30/60/90/120/180/240/300 and 360 min after ingestion of strawberries. Urine samples were collected during 24 hours after eating the strawberries.

Vitamin C was determined photometrically after oxidation to dehydroascorbic acid and reaction with 2,4-dinitrophenylhydrazine. The polyphenolics were assessed photometrically by using the Folin-Ciocalteu method. Antioxidant capacity was determined by using three different assay systems (Ferric Reducing Antioxidant Power (FRAP), Photoluminescence (PCL), Trolox Equivalent Antioxidant Capacity (TEAC)).

The parameters analysed increased in plasma as well as in urine after consumption of both types of strawberries. Maximum values were reached delayed after consumption of dried strawberries compared to raw strawberries. In addition, the elimination of some parameters was slower after ingestion of dried strawberries. Thus, microwave-vacuum dried strawberries are a healthy, convenient snack which delivers a number of valuable plant ingredients to the consumer.

[V18]

Vitamin E and A concentrations trace element levels, and some antioxidant parameters in the blood of bladder cancer patientsWasowicz W¹, Gromadzinska J¹, Jablonowski Z², Grzelinska Z¹, Pytlińska E¹¹Department of Toxicology and Carcinogenesis, Institute of Occupational Medicine, Lodz,²Department of Urology, Medical University, Lodz, Poland

It is generally believed that processes involving generation and promotion of reactive oxygen species and/or their metabolites play an important role in the initiation and development of numerous pathologic conditions including cancer. It is currently believed that antioxidants may directly reduce concentration of oxygen and its active forms (superoxide radical anion, hydrogen peroxide), bind metal ions, scavenge free radicals capable of initiating chain reactions with organic compounds. Enzymatic antioxidants include glutathione peroxidases (GPx), superoxide dismutase (SOD), glutathione S-transferase, and ceruloplasmin (Cp). Non-enzymatic antioxidants comprise tocopherols (vitamin E), β -carotene (provitamin A), ascorbic acid (vitamin C), lycopene, glutathione, uric acid, and bilirubin. Microelements (selenium, zinc and copper) associated with an active site or occurring in antioxidative enzymes as structural elements also play an important role.

The aim of the study was to determine vitamin E, A and β -carotene concentrations, selenium, zinc and copper levels and some antioxidant parameters (GPx, SOD, Cp activities, total antioxidant capacity (TAC)) in the blood of bladder cancer patients. The study covered 65 bladder cancer patients (men only) and 157 appropriate controls. The results of the study indicated statistically lower vitamin A and E concentrations in cancer patients ($p < 0.0001$) as compared to controls. Copper level was significantly elevated ($p < 0.0001$) and was found to be 1.24 ± 0.28 mg/l in cancer patients vs. 1.00 ± 0.18 mg/l in controls. We did not find any differences between selenium and zinc concentrations in cancer subjects as compared to controls. Glutathione peroxidase activity in red blood cells of bladder cancer patients was found to be 22.0 ± 5.3 u/g Hb. This value was significantly higher than in healthy controls (17.3 ± 3.9 u/g Hb; $p < 0.0001$). Activities of SOD in erythrocytes ($p < 0.001$) and Cp in plasma (NS) as well as TAC concentration ($p < 0.0001$) were significantly higher in cancer patients. Statistically significant linear correlations between selected parameters determined were found. Further and more thorough research are needed to comprehend the relationship among the incidence of bladder cancer and the involvement of antioxidants in this process.

[V19]

Dietary ascorbic acid supplementation: the good and the badBerzina Nadezhda, Vasilyeva Svetlana, Basova Natalya, Apsite Mirdza

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Ascorbic acid is marketed as a dietary supplement because of its antioxidant properties. However, we report here that ascorbic acid administered to diet in different doses can exhibit a prooxidant, as well as an antioxidant effect in chicks. The aim of the present study was to investigate how L-ascorbic acid dietary loading can influence on antioxidant and immune status of experimental chickens. Five groups of one to 30-day-old male Lohmann Brown chickens were fed the diets differed only by L-ascorbic acid supplements (0, 100, 500, 1000 and 2000 mg/kg). At the end of the experiment the birds were weighted and sacrificed. To assess the metabolic and immune changes induced by L-ascorbic acid the next indices were studied: in blood – hematological parameters; in blood serum – creatinine and uric acid content, lysozyme and circulating immune complexes level; in liver – MDA, GSHpx activity, glutathione level, Cd concentration; in immunocompetent organs – T and B immunocyte number. Permeability of small intestine epithelium was evaluated by glycine accumulation *in vitro*. The high loading of ascorbic acid (1000 and 2000 mg/kg) caused a significant increase of MDA and a fall of GSHpx activity and glutathione level in chicken liver. These indices change shows the oxidative stress development in tissue. In case the supplementation of low doses of dietary ascorbic acid was used, a reduction of creatinine and uric acid level in blood serum has been observed. However, these protein catabolism parameters were enhanced under high level of ascorbic acid intake. The increasing of ascorbic acid content in diet promoted the accumulation of heavy metal Cd in chicken liver. It can be connected with a rising of intestinal epithelium permeability. In the chicken groups that received ascorbic acid addition till 500 mg/kg the increase of phagocytic activity of blood neutrophils and number of T and B immunocyte in according immunocompetent organs was observed. It indicated the immunostimulating effect of these ascorbic acid supplements. Further dietary ascorbic acid increase has changed the direction of immune responses in chickens. The indices of natural immunity in blood serum show the immunosuppressive effect of ascorbic acid doses over 1000 mg/kg.

[V20]

Vitamins A, E and antioxidants enzymes in rat liver and heart under different light and geroprotectors influence

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The longevity is significantly dependent on reactive oxygen substances the level of which is regulated by the antioxidant system. Light conditions are influence the lipid peroxidation intensity, which is especially actual for the northern-European population. The aim of this investigation was to study different light and geroprotector (melatonin and epitalone) influence on various antioxidant system components - vitamins E and A level, as well as key antioxidant enzymes activity – superoxide dismutase (SOD) and catalase, in rats liver and heart.

Four groups of rats kept from one to six-month old were kept at 12-hour day and night cycles (Group 1), natural illumination (Group 2), constant illumination (Group 3) and in complete darkness (Group 4). The 4-months old animals from first, second and third groups were divided into three subgroups: first - received melatonin with drinking water in a doze 10 mg/L, second – 0.1 µg of epitalone with hypodermic injection, third - received placebo.

The vitamins A and E concentration and enzymes activity in the investigated organs greater depends on light conditions than on substances. The vitamins level in liver was more sensitive to experimental procedure compared with the enzymes activity. The light conditions did not influence the concentration of vitamin E in heart. The maximum SOD activity in liver was observed in animals kept in complete darkness. The maximum number of significant changes in vitamins concentration and antioxidant enzymes activity under the effect of melatonin and epitalone was found at constant illumination.

Thus, the present research has demonstrated high stability of non-enzyme (vitamins E and A) and enzyme (SOD and catalase) antioxidant system components, to changing light conditions and introducing geroprotectors: melatonin and epitalone.

[V21]

Concentration of 25-hydroxy vitamin D in plasma of dairy cows differently exposed to solar radiation

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Eighteen lactating dairy cows were subjected to different exposure to solar radiation. The treatments tested were (i) grazing on pasture vs. grass feeding indoors, (ii) exposure to daylight at different daytimes (one hour during sunrise vs. one hour at noon, while remaining in the barn for the rest of the day) and lowland grazing (at 400 m a.s.l., Period I) vs. high alpine grazing (at 2000 m a.s.l. with presumed higher UV radiation, Period II). No direct effects of any of these treatments on the 25-OH-vitamin D concentration in the blood plasma of the cows were found, although a mineral mix free of vitamin D had been used. By contrast, a general increase of the plasma levels with the onset of the warm summer weather was found in all groups. It is concluded that the concentration of the hydroxylated forms of vitamin D does not respond to the kind of treatments investigated. However, this does not allow to assume the same for the non-hydroxylated form of vitamin D, because the lack of effects on 25-OH vitamin D also could have been caused by a limited hydroxylation capacity of the liver.

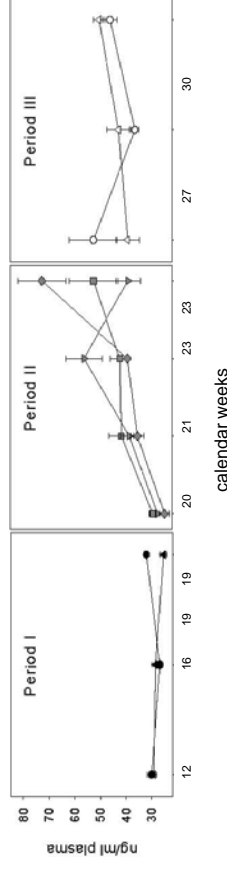


Figure 1. Concentration of 25-OH-vitamin D in blood plasma of dairy cows in subsequent experimental periods. Periods I (400 m) and III (2000 m): ●, indoor grass-fed group; ▲, indoor pasture group. Period II (400 m): ▼, grass-fed group, let out in the morning; ◆, grass-fed group, let out at noon; ◆, silage-concentrate group, let out at noon.

[V22]

Studies on the biotin flow at the duodenum of dairy cows fed differently composed ration*(Untersuchungen zum Biotin-Fluss am Duodenum von Milchkühen bei Fütterung unterschiedlich zusammengesetzter Rationen)*Lebzien P¹, Schröder B², Abel HU², Flachowsky G¹¹ Institut für Tierernährung, FAL Braunschweig, Bundesallee 50, 38116 Braunschweig² Institut für Tierphysiologie und Tierernährung, Universität Göttingen, Kellnerweg 6, 37077 Göttingen

It is well established that rumen microbes are able to synthesize B-vitamins, but knowledge of the effects of feed composition on net biotin output is scarce.

Research reported herein was conducted to determine effects of diets differing in concentrate-to-roughage ratio and type of grain on the duodenal biotin flow. Six cows, each fitted with cannulae in the rumen and in the proximal duodenum were used in four trials. Rations consisted of (I) 100% grass hay, (II) 81% corn silage and 19 % protein-mixture, (III + IV) 43% grass silage and 57% concentrate mixture containing 87 % wheat grain or corn respectively, 11% soybean meal and 2% oil. The cows were pre-fed the rations for 21 days. Thereafter duodenal digesta was sampled every two hours in two periods of 5 days each. Cr₂O₃ was used as a flow marker and microbial portion of total N at the duodenum was estimated by NIRS.

Daily intake of organic matter and biotin, the amount of fermented organic matter and flow of microbial-N and biotin at the duodenum are shown in the following table:

Ration	(I) (n=2)	(II) (n=4)	(III) (n=3)	(IV) (n=3)
Intake/day organ.matter (kg)	8.33	10.27	16.12	16.29
biotin (mg)	2.55	2.35	4.49	4.20
Fermented organic matter (kg/day)	5.94	7.83	13.13	11.59
Duodenal flow/day microbial-N (g)	120	175	395	356
biotin (mg)	1.83	4.37	5.89	6.16

Flow of biotin at the duodenum was not significantly related to biotin intake ($r^2=0.40$) but to amount of fermented organic matter ($r^2=0.72$) and amount of microbial protein ($r^2=0.71$).

[V23]

Effects of chronic ethanol treatment on levels of vitamin B1, B2, and B6 in rat brain*(Auswirkungen einer chronischen Ethanolapplikation auf die Vitamin B1, B2 und B6 Spiegel im Rattengehirn)*Netzel M¹, Borsch C², Bitsch Irmgard²¹Institute of Nutrition, Friedrich-Schiller-University, Domburger Strasse 29, D-07743 Jena, Germany²Institute of Nutrition, Justus-Liebig-University, Wilhelm Strasse 20, D-35392 Giessen, Germany

Vitamin B1 (Thiamine) and vitamin B6 (Pyridoxine) act in form of their coenzymes (thiamine diphosphate [TDP] and pyridoxal-5-phosphate [PLP], respectively) in the carbohydrate and amino acid metabolism. Vitamin B2 (Riboflavin) catalyses numerous oxidation-reduction reactions. The major function of riboflavin is to serve as the precursor of the coenzymes FMN (flavin mononucleotide) and FAD (flavin adenine dinucleotide) and of covalently bound flavins. Because FAD is part of the respiratory chain, vitamin B2 is central to energy production. The aim of the present study was to investigate the long-term ethanol related effects on vitamin B1, B2, and B6 status in brain tissue, to get new facts for a potential therapeutically treatment. Male Wistar rats (n = 14) were separated into a control group (n = 7) and an alcohol group (n = 7) at random. The animals received a semi synthetic diet (1.9 mg Thiamine Hydrochloride, 18.2 mg Pyridoxine, and 7.5 mg Riboflavin per kg diet), which was sufficient in all essential nutrients. Alcohol (20% ethanol-solution) was given instead of tap water. The ethanol calories were substituted by sucrose and the groups were fed isocalorically for 12 months. Vitamin B1, B2, and B6 (as a sum of all detectable vitamers) were analysed in brain tissue by HPLC. Chronic ethanol intake caused significantly (p < 0.05) reduced levels of vitamin B6 (- 19 %), PLP (- 26%), FAD and FMN (- 18%) as well as TDP (- 21%). Vitamin B1 and B2 were slightly reduced (p > 0.05) of about 4 and 12%, respectively. The results suggest that ethanol markedly impairs the conversion of Thiamine, Riboflavin, and Pyridoxine to their physiological active coenzymes, which may contribute to the depression of brain metabolism observed in alcoholism.

[V24]

Efficacy of benfotiamine vs. thiamine hydrochloride on thiamine status in ethanol treated rats*(Wirksamkeit von Benfotiamin und Thiamin-Hydrochlorid auf den Thiaminstatus der alkoholbehandelten Ratte)*Nezel M¹, Raubach C², Jaworski A², Bitsch Irmgard²¹Institute of Nutrition, Friedrich-Schiller-University, Dornburger Strasse 29, D-07743 Jena, Germany²Institute of Nutrition, Justus-Liebig-University, Wilhelm Strasse 20, D-35392 Giessen, Germany

The purpose of the present investigation was to study the efficacy of benfotiamine (lipid-soluble thiamine derivative), compared to thiamine hydrochloride (water-soluble salt) on thiamine status in ethanol treated rats. The study was performed with 21 male Wistar rats, which were separated into 3 treatment groups (7 animals each) at random. Group C (control): thiamine hydrochloride (11.9 mg/kg diet) and tap water; group TE (thiamine/ethanol): thiamine hydrochloride (11.9 mg/kg diet) and ethanol (20%-solution) per oral ad libitum; group BE (benfotiamine/ethanol): benfotiamine (16.45 mg/kg diet; equimolar to thiamine hydrochloride in the treatment groups TE and C) and ethanol (20%-solution) per oral ad libitum. The ethanol calories were substituted by sucrose and the groups were fed isocalorically for 3 months. Thiamine, thiamine monophosphate (TMP), and thiamine diphosphate (TDP; biological active coenzyme) were determined in blood, brain, heart, liver and kidney by HPLC. The ethanol treatment caused significantly ($p < 0.05$) reduced levels of TDP in blood (- 22%), liver (- 24%) and kidney (- 17%), and of total thiamine in blood (- 13%) and liver (-19%), respectively. An improved TDP and total thiamine status could be observed in the benfotiamine fed animals (compared with the ethanol free control as well as with the thiamine hydrochloride/ethanol group). Treatment with benfotiamine could be an effective strategy in the prevention of alcohol induced thiamine/ TDP deficiency.

[V25]

Vegetable diets without and with vitamin B12 supplements in laying hens*(Pflanzliches Legehennenfutter ohne und mit Vitamin B12-Supplementation)*Richter G.¹, Arnhold W.², Ochrimenko Wl¹, Köhler H¹¹ Thuringian State Institute of Agriculture, Auf der Höhe 6, D-07751 Jena OT Remderoda² BASU-Mineral Feed Inc., Bergstr. 2, D-99518 Bad Sulza

Two investigations were carried out to investigate the influence of 20 mcg vitamin B12 supplementation /kg vegetable diet (wheat, barley, corn, soybean meal, sunflower meal) in laying hens. In a 40 weeks lasting experiment A 5 x 2 LSL-laying hens per group were kept in cages during the 24th – 64th week of age. In experiment B (22 weeks) 8 x 2 LB-hens per group were also kept in cages during the 38th – 60th week of age. The number of eggs was registered daily, and the eggs were weighed 3 times per week. The parameters of egg quality (outside and inside) were measured permanently on 80 eggs per group in experiment A, and on 78 eggs per group in experiment B. The vitamin B12 concentration was analysed in lyophilised eatable eggs, in the liver, and in excrements and caecum content dried at 60 degree centigrade. The vitamin B12 analysis was carried out by means of the microbiological method with *Lactobacillus leichmannii* (ATCC 7830).

On average of both experiments no differences of the zootechnical parameters were observed between the groups without and with vitamin B12 supplementation. The mean produced egg mass in both experiments was 1.8% higher in the groups without vitamin B12 supplementation compared to hens with vitamin B12 supplementation. Furthermore, the hens without B12 supplementation gained 30 g/animal more weight ($p > 0.05$).

In experiment a one hen developed symptoms like nervous disturbances, paralysis, and cachexy. The quality of feathers and the dry matter content of eggs, liver, and caecum content did not differ between the groups without and with vitamin B12 supplementation.

Among the registered outside egg quality parameters like deformation, shell breakage strength, shell density, shell colour as well as the inside egg quality parameters like albumin height, Haugh-units, and blood spots the shell breakage strength, the shell density, and the Haugh-units were significantly increased in the groups without supplementation.

The vitamin B12 supplementation of the diet increased the vitamin B12 concentration in the egg contents by 19%, in the liver by 66%, and in the excrements by 49%. Furthermore, the vitamin B12 concentration of 20.000 mcg / kg dry matter in the caecum content was estimated as very high (Table).

Table: Vitamin B12 concentration in various substances (mcg/kg dry matter, 3 experiments A und B)

Substances	n	+ 20 µg B ₁₂ /kg		without B ₁₂ suppl.		t-Test
		3	S ±	3	S ±	
Feed (88 % DM)	3	30,3	1,5	5,3	4,9	p < 0,001
Eatable egg	6	185	24	155	11	p < 0,01
Liver	4	2.278	194	1.375	519	p < 0,01
Caecum content	3	19.723	5.013	21.647	8.187	>
Excrements	5	1.839	619	1.236	665	>

[V26]

Blood vitamin concentrations in dogs before and after intake of diets with specified vitamin contents*(Vitaminskonzentrationen im Blut bei Hunden mit unterschiedlicher Vitaminaufnahme)*Tran JL¹, Horvath C², Krammer S³, Höller U³, Zentek J^{1,4}¹Department of Veterinary Public Health and Food Science, Institute of Nutrition²Clinical Department for Small Animals and Horses, Clinic of Internal Medicine for Small Animal and Horses, University of Veterinary Medicine, Vienna³DSM Nutritional Products Ltd, Animal Nutrition and Health and Analytical Research Center, Basel⁴Institute of Animal Nutrition, Free University of Berlin, Berlin

The objective was to investigate blood vitamin concentrations in healthy dogs and to determine the effects of defined dietary vitamin intakes on blood vitamin levels and hair and skin quality. A total of 64 healthy dogs aged from 1 to 8 years without any history of skin or coat problems were used. All animals were fed commercial complete diets before the start of the study. According to weight and gender the animals were assigned to four groups with different vitamin concentrations, adjusted to meet or exceed the dietary requirements. Diet 1 had the lowest analyzed vitamin levels; diets 2, 3, and 4 had 2-3 fold increased concentrations of water soluble vitamins when compared to diet 1 (Table).

	Diet 1	Diet 2	Diet 3	Diet 4
Vitamin A, IU	4320	5450	3720	3050
Vitamin D, IU	700	800	600	650
Vitamin E, mg	86	87	94	95
Ascorbic acid, mg	70	57	59	66
Vitamin B1, mg	6	16	17	17
Vitamin B2, mg	5	17	19	19
Vitamin B6, mg	6	20	15	16
Vitamin B12, mg	0.09	0.1	0.1	0.1
Pantothenate, mg	15	34	31	33
Niacin, mg	25	70	78	80
Folic acid, mg	0.6	2.7	3.6	4.7
Biotin, mg	0.3	0.6	1.3	2.6

The blood vitamin levels of the dogs were investigated at day 0 and after the feeding of diets with specified vitamin levels at day 122. Skin and coat quality was evaluated by a defined dermatological scoring system. Plasma biotin concentrations increased in group 4 (P<0.05) with the highest biotin intake (2.6 mg/kg diet). Pyridoxal-5-phosphate (P=0.001) blood levels and plasma α -tocopherol levels (P<0.05) decreased in group 1, having the lowest intakes. Cobalamin levels increased and retinol and 25-hydroxycholecalciferol levels decreased significantly in all groups compared to baseline values determined at day 0. Serum pantothenate increased significantly in groups 2 to 4, which received amounts twice as high as group 1. Dermatological investigations did not detect significant changes of skin and coat conditions. In conclusion, this study gives a survey of blood vitamin concentrations in healthy dogs and provides a data base for the evaluation of the vitamin status in health and disease. Comparing the blood vitamin concentrations before and after the intake of the defined diets allows the conclusion, that the dietary intakes of some vitamins were lower under field conditions, however, the intake of most other vitamins seems to have exceeded the levels in the experimental diets.

[V27]

Effect of baking and freezer storage time on added folic acid stability in wheat and rye breads

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The objective of this study was to examine the effect of baking process and freezer storage time on added folic acid to wheat and rye breads. Breads were produced using the formulation containing enriched wheat and rye flours with 0.4 mg folic acid/100g product and baker's yeast. Breads were also produced with untreated flours containing no folic acid. The amount of doughs for bread making were 250g, baking temp. 230 °C, and time of 30 i 40 min. for wheat and rye breads, respectively. Breads were frozen at -20 °C and held for up to 16 weeks. Fresh breads and breads frozen for 5, 10 and 16 weeks were measured for folic acid content.

Folic acid was extracted with Hepes/Ches buffer (pH=7.85) followed by destruction of matrix by amylase and protease. The extracts were purified with strong anion-exchange (SAX) cartridges. Folic acid was determined by HPLC using a Phenomenex Luna C 18 column (5 μ m, 250 x 4.6 mm). The chromatographic condition for gradient elution were as follows: flow rate, 1 ml/min; volume injected, 1 ml; column temperature, 23 °C; UV detection, 290 nm. Gradient elution was performed with acetonitrile-33 mM phosphate buffer, pH 2.3. The run time was 30 min and the time between injections 42 min. The gradient was started at 5% (v/v) acetonitrile, maintained isocratically for the first 8 min, thereafter the acetonitrile concentration was raised linearly to 17,5% (v/v) within 25 min. Peak identification was based on the retention time, by comparison of the ratios of UV peak with that of the standard compounds.

For both rye and wheat breads there was a significant decrease of folic acid (~15%) from flour to bread stage. No folic acid was lost during storage of the fortified wheat and rye breads in a freezer for 5 weeks but after 10 and 16 weeks of storage folic acid concentration decreased significantly (p<0.05).

[V28]

Determination of B vitamins in infant milk by high-performance liquid chromatographyMichalak J, Kuncewicz A, Gujska E

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A liquid chromatographic method for determining of the B group vitamins in powdered infant milk was conducted. The procedure was developed to control of five water-soluble vitamins, B₁ (thiamine), B₂ (riboflavin), B₆ (pyridoxamine phosphate, pyridoxal-HCl, pyridoxal-2HC) in infant foods. Traditional methods for vitamin determination usually depend on the accuracy and sensitivity required, and the interferences encountered by the sample matrix. Nowadays, there is a growing need for more rapid and specific methods for vitamin analysis. High-performance liquid chromatographic (HPLC) techniques allow rapid separation and qualification of B vitamins in food using reversed-phase columns and UV detection. The vitamins were extracted from powdered infant milk using the hydrochloric acid hydrolysis followed by enzymatic digestion with taka-diastase for vitamin B₁, B₂ and with acid phosphatase for B₆ group vitamins. The procedure includes studies about the composition of the mobile phase, flow-rate and temperature. Chromatographic analysis of all vitamins was carried out using a reversed-phase C₁₈ column with acetonitrile/0.05M potassium dihydrogen phosphate (10:90, v/v) containing 0.3 x 10³M sodium octane sulfonate as mobile phase. The mobile phase was then adjusted to pH 2 with orthophosphoric acid. The separation was performed at 30°C at a flow rate of 1 ml min⁻¹, the UV-detection at 260, 268 and 290 nm. The injection volume was 50 µl. All five compounds were separated in less than 35 min. Peaks identification were based on the retention time, by comparison of the ratio of UV spectra with that of standard commercial compounds. Validation was performed using the certified reference materials (milk powder CRM 421). Linearity, precision, recovery and sensitivity were satisfactory. The method was successfully applied for determination of the B group vitamins in powdered infant milk.

[A1]

Studies on the bioavailability of various L-carnitine compounds*(Untersuchungen zur Bioverfügbarkeit verschiedener L-Carnitin-Verbindungen)*Keller Uta, Feigner Juliane, Becker Karin, Kluge H, Eder K

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L-carnitine is essential for beta-oxidation of long chain fatty acids. Physical exercise or diseases such as ischemia, neuropathy, AIDS, or hemodialysis develop a need for additional L-carnitine. To meet the requirement under these conditions, dietary supplements can be helpful. Base of free L-carnitine is highly water soluble, the absolute bioavailability is relatively low. It is largely unknown whether chemical modification of free base of L-carnitine into its salts or esters influences its absorption kinetics. Therefore, the aim of the present study was to compare absorption rate and kinetics of various L-carnitine esters (acetyl-L-carnitine and lauroyl-L-carnitine) and organic salts (L-carnitine L-tartrate, L-carnitine fumarate, L-carnitine magnesium citrate) in piglets as model animals. Six groups of five or six piglets each were administered orally a single dose of 40 mg L-carnitine equivalents/kg body weight of each of those L-carnitine compounds. A seventh group served as a control. Free and total plasma carnitine concentrations were determined 1, 2, 3.5, 7, 24 and 32 h after administration of the single dose. Area-under-the-curve (AUC) values were calculated to assess the bioavailability of the L-carnitine compounds. AUC values, calculated for the time interval between 0 and 32 h, for both free and total carnitine were similar for base of free L-carnitine and the three L-carnitine salts (L-carnitine L-tartrate, L-carnitine fumarate, L-carnitine magnesium citrate) while those of the two esters (acetyl-L-carnitine, lauroyl-L-carnitine) were lower. Administration of L-carnitine L-tartrate yielded a higher plasma free carnitine AUC value for the time interval between 0 and 3.5 h than administration of the other compounds. The data of this study suggest that L-carnitine salts have a similar bioavailability as base of free L-carnitine while L-carnitine esters have a lower one. The study also suggests that L-carnitine L-tartrate is faster absorbed than the other L-carnitine compounds.

[A2]

Effects of L-carnitine supplementation on the somatotrophic axis and masses of placenta in pregnant sows

(*Effekte einer L-Carnitinsupplementation auf die somatotrophe Achse und die Massen der Plazenten bei trächtigen Sauen*)

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Recent studies have shown that L-carnitine supplementation in pregnant sows improves intrauterine growth of piglets. Placental function is essential for growth of the fetuses, and it is strongly influenced by the somatotrophic axis, particularly by maternal plasma concentrations of IGF-1. The present study aimed to investigate whether L-carnitine supplementation in pregnant sows influences plasma concentrations of hormones of the somatotrophic axis and masses of placenta. In the first experiment, 40 gilts were divided into two groups. In the second experiment, 20 sows in the third parity were divided into a control group and a treatment group. All the sows received nutritionally adequate diets. Sows of the treatment group received a supplement of 125 mg L-carnitine per day. In the first experiment, sows supplemented with L-carnitine had higher plasma concentrations of IGF-1 (+43%, $P < 0.05$), IGF-2 (+147%, $P < 0.05$) and lower plasma concentration of IGFBP-3 (-14%, $P < 0.05$) at day 80 of pregnancy than control sows. The plasma concentrations of somatotropin, somatostatin and insulin did not differ between the two groups of sows. The number of piglets born did not differ between both groups of sows but piglets of sows supplemented with L-carnitine were slightly heavier than those of control sows (+9%, $P < 0.25$). In the second experiment, sows supplemented with L-carnitine had also slightly heavier piglets than control sows (+10%, $P < 0.25$). Sows supplemented with L-carnitine had also higher placental masses (+19%, $P < 0.15$) and higher contents of total protein (+40%, $P < 0.01$) and DNA (+29%, $P < 0.10$) in placenta than control sows.

The study shows that dietary L-carnitine supplementation during pregnancy increases the plasma concentrations of IGF-1, -2 and placental masses. This suggests that the improved intrauterine growth of the piglets in sows treated with L-carnitine may be the result of an improved transplacental nutrient supply.

[A3]

Impact of fish n-3 polyunsaturated fatty acids on growth of the HT29 human colon tumour cell line

(*Einfluss von fischeigenen n-3 mehrfach ungesättigten Fettsäuren auf das Wachstum der humanen HT29 Kolonkrebiszelllinie.*)

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Objective: Colorectal cancer is one of the most common cancers in western countries. Epidemiological studies suggest a high fish intake to be associated with a decreased risk for colorectal cancer, but research is still needed to improve understanding of cancer preventive mechanisms of such a diet. Therefore the major fatty acid constituents of fish oil, namely the n-3 polyunsaturated fatty acids (PUFAs) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which are thought to be the bioactive compounds in fish, were used for investigating growth modulatory effects on HT29 colon tumour cells. **Methods:** The effect of ethanol dissolved fish PUFAs EPA (C20:5, n-3) and DHA (C22:6, n-3) in comparison to the common plant PUFA linoleic acid (LA, C18:2, n-6) [10-1000 µM] on growth of the HT29 human colon tumour cell line was determined. For this we used the 4',6-Diamidino-2-phenylindole (DAPI) assay for DNA quantification as well as the Cell Titer Blue (CTB) assay for measuring mitochondrial metabolic activity after 24, 48 and 72h treatment. **Results:** Both fish n-3 PUFAs EPA and DHA reduced the DNA amount and mitochondrial metabolic activity of HT29 colon tumour cells in a dose- and time-dependent relationship. In comparison to EPA DHA showed a stronger inverse association on DNA amount (DAPI EC₅₀ 24h 555 vs. 178 µM; 48h 431 vs. 135 µM; 72h 390 vs. 117 µM) as well as on the mitochondrial metabolic activity (CTB EC₅₀ 24h 926 vs. 291 µM; 48h 764 vs. 185 µM; 72h 651 vs. 155 µM). A treatment with LA had neither an impact on the DNA amount, as a mirror of the cell number, nor on the mitochondrial metabolic activity. High concentrations of all PUFAs (≥ 500 µM) showed microscopically detectable cytotoxic effects. **Conclusions:** Our data show, that both fish n-3 PUFAs EPA and DHA are potent inhibitors of colon tumour cell growth by affecting cell numbers (calculated from the DNA amount) and metabolic activity of HT29 colon tumour cells in contrast to the n-6 PUFA LA.

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[A4]

Effect of conjugated linoleic acids on growth and nutrient digestibility in dogs*(Einfluss von konjugierten Linolsäuren auf Wachstum und Nährstoffverdaulichkeit beim Hund)*

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Conjugated linoleic acid (CLA) is known to have an effect on body composition in various mammals like mice, rats, pigs and humans. In general lean muscle mass is increasing and fat, especially depot fat is depressed while body mass keeps unchanged. In pigs an improved feed conversion rate was reported when CLA was added to the diets.

Two experiments were conducted to investigate the effect of CLA on growth performance and nutrient digestibility in dogs. In both experiments, there were two treatments (control: 5% sunflower oil, 0% CLA; treatment: 5% CLA, 0% sunflower oil). The CLA (Luta-CLA® 60) used in this experiment contained a mixture of isomers consisting of 30% c9, t11 and 30% t10, c12. All diets were formulated to meet AAFCO standards. In experiment 1, twenty adult mixed breed dogs (10 males and 10 females) were allotted to treatments by sex and weight. Dogs were housed individually in kennels that measured 2m x 3m. Feed and water were provided *ad libitum*. Body weight, body condition scores and feed intake were measured weekly. Over the entire experiment, there was a positive trend ($P > 0.05$) for the dogs fed CLA to have lower weight gain (1.05 kg for CLA and 2.13 kg for control) and have less fat deposition than dogs on the control diet (0.05 mm for CLA and 0.10 mm for control). In experiment 2, 6 Beagle dogs/treatment were used to compare nutrient digestibilities of two experimental diets (0% CLA and 5% CLA). Dogs were housed individually in metabolism crates. Total fecal collection was done over a 5-day period. There were no differences ($P > 0.05$) in digestibility coefficients between the two treatments. Results from experiment 1 indicate that CLA can be fed to control the rate of weight gain and the amount of fat deposition in adult dogs. Results from experiment 2 demonstrate that CLA does not have a negative impact on nutrient digestibility.

[A5]

The use of sodium butyrate (adimix c) in layer nutritionPietsch M³, Schwarzer C², Arnouts S¹¹INVE TECHNOLOGIES nv, Hoogveld 93, B-9200 DENDERMONDE, BELGIUM²NUTRI-AD INTERNATIONAL NV, Kloostersstraat 1 Bus 7 B-2460 KASTERLEE, BELGIUM³BFI, Telfort Court, CH1 6LT CHESTER, England

The use of organic acids in animal nutrition is, according to EU legislation, merely the subject of use as an antimicrobial in feed preservation. Unlike other acidifiers butyric acid not only has antimicrobial activities, but has also biological functions at cellular level. From research in human nutrition, it is well known that butyrate is used as primary source of energy by epithelial cells. Therefore it can be hypothesized that due the effect of butyric acid on absorption of nutrients, it might improve technical performance and might also have an effect on the absorption of Ca and P in layers feeds. This is especially the case with older layers with decreasing lay performance, when cage fatigue and bone demineralization can become problematic.

A trial consisting of 5 treatments 7 repetitions (5 times 4 ISABROWN layers) was set up in which different concentrations of ADIMIX C (Sodium Butyrate) were tested (0, 50, 100, 250 and 500 ppm). ADIMIX C is added to a high quality wheat/corn based feed that is fed during 2 times 28 days. Animals were 54 weeks of age at the start of trial and were housed at a density of 450 cm² on a 18L:6D light scheme

Besides technical performances (lay performance, average egg weight, daily egg mass output and feed efficiency), eggs of a 24 h lay period were be collected biweekly to evaluate the following parameters:

- Egg bending and egg shell cracks to evaluate the effect of butyrate on the egg shell strength through improves Calcium absorption
- Egg dirtiness (feces and blood spots) to evaluate the effect on intestinal metabolism and absorption (or malabsorption)

Results of the first four weeks of the trial indicate a positive result of the application of ADIMIX C on lay performance improving it from 84.3 % (control) to 88.0 % at 500 ppm. As average egg weight was not affected (well noticed in layers at early stage of lay), the daily egg mass output increased from 56.0 g/a/d (control) to 57.8 g/a/d at 500 ppm ADIMIX C.

It can be concluded that the supplementation of ADIMIX C in diets for older layers has a positive effect on technical performance.

[A6]

Effect of dietary supplementation with algal omega-3 lipids on fatty acid composition of egg yolksSobajic S, Djuricic I, Djordjevic B, Konic-Ristic A

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Changes in the composition of the feed of laying hens can influence composition and nutritional value of eggs. This possibility has been used to produce so-called “functional eggs” with lower cholesterol and saturated fatty acid content, increased iodine or omega-3 fatty acids content. Especially omega-3 enriched eggs are interesting because they can be valuable sources of eicosapentaenoic and docosahexaenoic acids (EPA and DHA). Different omega-3 sources are used for dietary supplementation of hens, as flaxseed and fish meal. The aim of this experiment was to investigate the effect of hen’s diet supplementation with DHA-rich oil from microalga *Schizochytrium* on fatty acid composition of egg lipids.

Study was conducted with 60 hens divided into control group and omega-3 supplemented group. Control group was fed basal diet with added sunflower oil. In the basal feed of omega-3 supplemented group 0.7% and 1% of DHA-rich oil was added. The eggs were analyzed for fatty acid composition on 0, 7 and 30 days feeding period. The fatty acid composition of extracted lipids was analyzed using gas chromatography.

DHA-rich oil from microalga *Schizochytrium* has high content of DHA and omega-6 docosapentaenoic acid (DPA). Table 1 shows a comparison of polyunsaturated fatty acid content of control and group supplemented with 1% DHA-oil.

Table 1: Fatty acid content of egg yolks (mg/100 g whole egg, without shell)

Fatty acids	Control	omega-3 supplemented group
Polyunsaturated fatty acids (PUFAs)	1130	1220
Omega-3 fatty acids	30	140
Docosahexaenoic acid (DHA)	22	130

DHA content in egg lipids increased 6 times after period of 30 days, while the content of omega-6 fatty acids, linoleic and DPA, and was not changed. Algal DPA was not accumulated in the egg yolk while algal DHA was successfully incorporated into egg lipids.

[A7]

Adipose tissue lipids of pigs fed sunflower oil supplemented dietTsvetkova Veneta, Angelov L.

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The effects of dietary unsaturated fatty acids on the lipogenesis and the accumulation of lipids in the pig adipose tissue have been investigated. The influence of frozen storage for three and six months has also been studied. The adipose tissue from the pigs fed standard feedstuffs (control group) and with supplement of 2 % sunflower oil to the mixture (experimental group) has been analyzed for total content of triacylglycerols (Tg), free fatty acids (FFA), cholesterol and activity of lipogenetic enzymes. The fatty acid composition of Tg and FFA in fresh tissue and in tissue stored at -25°C has been determined. The results show a significant increase ($P \leq 0.01$) of the FFA levels in the control as well as in the experimental groups stored for 3 and 6 months. The cholesterol lowering effect of the unsaturated oil is not proved. The activity of NADP-malate dehydrogenase in the experimental group is decreased ($P \leq 0.01$), which may be influenced by the added oil. The adipose tissue Tg of the experimental animals show higher concentration of 18:2 ($P \leq 0.01$) and lower of 16:0 ($P \leq 0.01$) vs. the control group in fresh tissue. In the two groups the level of the 18:1 is increased and those of the 18:2 and 18:3 are decreased at storage mainly for six months. The fatty acid composition of FFA in the experimental animals show higher values for the unsaturated 16:1 and 18:2 and lower ones for the saturated 16:0 ($P \leq 0.05$). When stored the higher levels of the FFA may be connected with a moderate increase of the molar concentrations of 18:1 and 18:2, simultaneously with a certain decrease of 16:0 and 18:0 in both groups. The increase in the degree of unsaturation doesn't lead to the formation of a high oxidative potential and to deterioration of the nutritive and technological qualities of the pig adipose tissue in frozen storage for six months.

[A8]

Influence of long-term intervention with supplemented dairy products on serum lipids*(Einfluss einer Langzeitintervention mit ergänzten Milchprodukten auf die Serumlipide)*Dawczynski C¹, Hein G², Schubert R¹, Deufel T², Jahreis G¹¹Department of Nutritional Physiology, Institute of Nutrition, Friedrich Schiller University of Jena, Dornburger Str. 24, D-07743 Jena, Germany²Department of Internal Medicine III, Friedrich Schiller University of Jena, Erlanger Allee 101, D-07747, Germany

Regular consumption of n-3 long chain polyunsaturated fatty acids (LC-PUFA) of marine origin can improve serum lipids and reduce cardiovascular risk. Dairy products are poor in LC-PUFA.

Dairy products like yogurt, cheese and butter were enriched with special oils which are rich in n-3 fatty acids, especially eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). This study aimed to analyse the influence of LC-PUFA supplemented products on serum lipids in humans.

Design: 45 subjects were randomised into two groups in a double-blind, placebo-controlled crossover study. Both groups received placebo products or supplemented dairy products consecutively for three months with a two-month washout phase between both periods. The daily dose of n-3 fatty acids amounted to 2.3 g, consisting of 1.35 g EPA+DHA and 0.95 g α -linolenic acid (LA). Clinical examinations were elevated at the beginning and at the end of each period.

Results: 39 test persons aged 57.9 ± 10.8 years completed the study. Total cholesterol, LDL cholesterol, LDL/HDL ratio and triacylglycerides were not affected by the intervention with dairy products. Interestingly, HDL cholesterol increased significantly ($p < 0.05$) by the consumption on dairy products rich in n-3 fatty acids but increased in the placebo period as well ($p < 0.01$). Total cholesterol/HDL cholesterol ratio tended to decrease due to the intake of n-3 fatty acids ($p < 0.1$) and decreased significantly in the placebo period ($p < 0.01$). Supplementation with dairy products rich in n-3 fatty acids tended to be associated with lower concentrations of lipoprotein(a) ($p < 0.1$).

Conclusions: The results showed that the supplementation of dairy products rich in LC-PUFA had only marginal influence on serum lipids and affected only selected risk factors of cardiovascular diseases.

[A9]

Assessment of genetically modified prebiotic potato tubers concerning their nutritive value and the fate of DNA*(Ernährungsphysiologische Bewertung von gentechnisch veränderten präbiotischen Kartoffeln)*Böhme H¹, Hommel B², Broll H², Flachowsky G¹¹ Institut für Tierernährung, FAL Braunschweig, Bundesallee 50, 38116 Braunschweig² Institut für integrierten Pflanzenschutz, BBA, Stahnsdorfer Damm 81, 14532 Kleinmachnow³ Bundesinstitut für Risikobewertung, Thielallee 88-92, 14195 Berlin

Silage from transgenic inulin synthesising potatoes was compared to that of the parental line (i.e. cv. Désirée). Inulin synthesis was transferred via constitutive expression of two 1-fructosyltransferase genes of globe artichoke, with the result that tubers represent the whole artichoke inulin spectrum at a concentration of 5%.

The content of crude nutrients, minerals and amino acids did not show significant differences due to the genetic modification. However, glycoalkaloids were analysed to be 25% increased (904 vs. 728 mg/kg DM) in the freeze-dried material. Significant differences in digestibility (%) were not determined but a tendency towards decreased fibre and increased NFE digestibility.

	Isogenic		Transgenic	
	Crude nutrients (% of DM)	Digestibility (%)	Crude nutrients (% of DM)	Digestibility (%)
CP	10.7	76.9	10.6	73.0
EE	0.6	66.3	0.6	49.9
CF	2.5	81.0	2.8	72.6
NFE	80.7	90.7	80.2	94.2

The iso- and transgenic silage were fed at a level of 40% DM in the diet over a period of 42 days to 4 pigs each. After slaughter, samples from organs and digesta were taken and analysed for identifying the fate of the foreign DNA, applying 4 different real time PCR systems. No plant-specific DNA was detected in any organ, chloroplast specific DNA was found in the digesta, sampled in the different segments from stomach to rectum.

[A10]

Effect of dietary transgenic maize on absorption and retention of mineral elements

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Effect of conventional and transgenic maize (Roundup Ready - RR), with an introduced gene of glyphosate resistance (two lots from Monsanto, USA) on minerals was studied in 30 rats (75g) divided into three feeding groups. Ca, Mg, K and Fe intake, apparent absorption and retention did not differ between groups. Higher P retention and absorption was stated only in one group RR2 ($p \leq 0.05$). The Zn apparent absorption was lower ($p \leq 0.05$) in RR2 group than in other groups. However Zn retention in RR2 and control groups were similar, and in both lower than in RR1 group. Although similar Cu intake in all groups, the significantly lower apparent Cu absorption in RR1 group than in the others was observed, however the Cu retention did not differ between groups. The results of this study, and the previous ones, as well as the fact that plant mineral composition can change dramatically independently on variety and species, allow us to conclude that genetically modified (RR) maize revealed equivalence in mineral composition with isogenic maize. The differences in mineral absorption and retention in rats offered transgenic maize of different origin (year and place of growing) were in many cases bigger than compared to animals fed isogenic maize.

[A11]

The use of Immunmilch preparations as colostrum substitute in the eradication and control of paratuberculosis

(Einsatz von Immunmilchpräparaten als Biestmilchersatz in der Paratuberkulosesanierung)

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Feeding experiments with an Immunmilch preparation produced and developed by Phytobiotics, Eitville, were carried out on 30 calves within the scope of a graduate thesis on a Thuringia dairy farm that holds 200 cows. The Immunmilch formula used in this study is an IBR-free, freeze dried colostrum that does not contain traceable amounts of the paratuberculosis pathogen.

In these experiments, two different dosage variants of the Immunmilch formula were given to the female calves of the study within four hours postpartum, depending on their birth weight (variant A 1.5 g immunoglobulins per kg live weight, variant B 3 g immunoglobulins per kg live weight). The male calves were fed colostrum collected and banked from the dairy farm's own paratuberculosis-free cows. 24 hours after birth, the calves underwent blood tests which were analysed to measure the gammaglobulin and protein content in the blood. The calves were weighed after birth and at the age of 14 days. From this data the daily weight gain was calculated. Additionally, the calves' state of health was recorded.

It was found that the two different dosages of immunoglobulin were only slightly reflected in the blood count of the group of female calves. Although the blood of the female calves showed significantly lower concentrations of Gammaglobulin and Protein as did the blood of the male calves that were fed with colostrum, they showed good results in the postpartum development (average daily weight gain of the female calves 9.8 kg, of the male calves 12.6 kg, the average number of days the female calves suffered from diarrhea was 1 day, that of the male calves 0.44 days).

Economically, the Immunmilch formula, which costs between 100 and 120 €/kg, can be rated as a very efficient colostrum substitute in the prevention of paratuberculosis in livestock. However, within the scope of general calf rearing it will hardly manage to hold its own on the market.

[A12]

The effect of benzoic acid on growth performance, digestibility of nutrients, nitrogen balance and gastro-intestinal microflora in piglets

(Einfluss von Benzoesäure auf Wachstum, Nährstoffverdaulichkeit, Stickstoffbilanz und die gastrointestinale Mikroflora beim Ferkel)

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To investigate the effects of benzoic acid on growth performance, digestibility of nutrients, nitrogen balance and gastro-intestinal microflora of piglets, a performance experiment and a separate balance study were conducted. In the performance experiment, four different dietary treatments were involved: 1 – basal diet (negative control); 2 – basal diet supplemented with benzoic acid at 5 g/kg; 3 – basal diet supplemented with benzoic acid at 10 g/kg; 4 – basal diet supplemented with potassium diformate at 12 g/kg. Each dietary treatment was assigned to 9 replicate groups, each consisting of 2 piglets. Supplementing the diet with benzoic acid resulted in a dose-dependent increase of feed intake and body weight gain and in an improved feed conversion ratio. In piglets fed the diet supplemented with benzoic acid at 10 g/kg, feed intake, body weight gain and feed conversion ratio were 9%, 15% and 6% better than in control piglets. Growth performance of the piglets fed the diet with benzoic acid at 10 g/kg was similar to that of piglets fed the diet supplemented potassium diformate. In the balance experiment, 3 groups of 6 piglets each were fed either a control diet or diets supplemented with benzoic acid at 5 or 10 g/kg, respectively. Benzoic acid did not affect significantly the digestibility of the nutrients but increased the nitrogen retention. Piglets fed the diets supplemented with benzoic acid at 5 or 10 g/kg retained 5 and 6%, respectively, more nitrogen than control piglets. Benzoic acid also reduced the number of bacteria in stomach, duodenum and ileum in a dose-dependent increase. Moreover, benzoic acid strongly reduced the concentration of acetic acid in the duodenum. In conclusion, this study shows that benzoic acid exerts strong antimicrobial effects in the gastro-intestinal tract of piglets and therefore increases growth performance and nitrogen retention.

[A13]

Studies on the effect of Creatine on growth performance and on carcass quality of broiler chickens

(Untersuchungen zum Einfluss von Kreatin auf das Wachstum und die Ganzkörperzusammensetzung von Broilern)

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Creatine is a substance occurring naturally in the animal body. The major proportion of the total Creatine pool is found in skeletal muscle. The present four studies were focused on the observation of the growth performance of male growing broiler chickens in dependence on Creatine feeding. Influences on carcass quality were investigated at 35 days (Experiments 1, 2, 3) or 84 days (Experiment 4) of age. 350/210/336 Lohmann Meat (Experiments 1/2/3) and 180 ISA 257 (Experiment 4) day-old broiler chickens were distributed to dietary treatments with 8-10 pens per group. The basal diet was formulated to contain 0, 0.5 g, 1 g, 2 g, 5 g or 10 g Creatine per kg. Feed and water were provided for ad libitum consumption. Body weight was recorded for each broiler individually whereas feed intake was weighed back weekly on a pen-basis. In Experiment 1, Creatine showed significant growth promoting effects (Tab. 1). In Experiment 2 and 3 Creatine supplementation didn't change final live weight at day 35 of age. In Experiment 4 final body weight was depressed after 1 g Creatine supplementation at day 84 of age. No effects on the carcass quality of broilers could be shown in the four Experiments.

Table 1: The influence of Creatine on final body weight, g/broiler (Mean, P_≤0.05)

Experiment	Control	Creatine 0.5 g	Creatine 1 g	Creatine 2 g	Creatine 5 g	Creatine 10 g
1	2092 b	-	2237 a	2215 a	2229 a	2187 a
2	2360	2351	2308	-	-	-
3	2264	2280	-	-	-	-
4	4321 a	4205 ab	4144 b	-	-	-

[A14]

Studies on the effect of Creatine on performance of laying hens*(Untersuchungen zum Einfluss von Kreatin auf die Leistungsmerkmale von Legehennen)*Halle Ingrid

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Creatine is a substance occurring naturally and is most abundant in fish and meat. Creatine is obtained through the diet and synthesized in the liver, kidney and pancreas. Vegetarians have lower serum Creatine concentrations than nonvegetarians. End of this fact and the situation that the feeding of animal meat meal to hens is forbidden, the question arose whether a supplementation of Creatine to diet improves the performance of breeding hens.

A total of 144 laying hybrids were allocated to 4 groups with 36 hens per group. Hens were kept singly in a cage battery. The basal diet was formulated to contain 0, 0.5 g, 1 g or 2 g Creatine per kg. The experiment commenced when the hens were 22 weeks old and lasted 13 laying months. Eggs laid were recorded daily and feed consumption monthly. Eggs were weighed four times in two weeks each month. Hens were artificial inseminated; eggs were collected and stored in the incubator at a hen age of 30, 40 and 65 weeks.

The laying intensity and egg mass production of hens fed the diets, supplemented with Creatine, were not improved. Neither hatchability nor chickens weight at hatching were significantly affected by Creatine supplementation.

Table 1: Laying and breeding performance of hens (P>0.05)

	Control	Creatine 0.5 g	Creatine 1 g	Creatine 2 g
Laying intensity, %	93	93	94	92
Egg mass, g/H/d	56	57	57	56
Hatchability, %	91	90	87	88
Chicken weight, g	39	39	39	39

[A15]

Investigations on the use of trace elements for authentication of the origin of poultry and beefFranke BM^{1,2,3}, Haldimann M², Bütikofer U³, Gremaud G², Hadorn R³, Kreuzer M¹¹Institute of Animal Science, Animal Nutrition, ETH Zurich, ETH-Zentrum, CH-8092 Zurich, Switzerland²Swiss Office of Public Health, CH-3003 Berne-Liebefeld, Switzerland³Agroscope Liebefeld-Posteaus, CH-3003 Berne-Liebefeld, Switzerland

Consumer demands for authentication of the origin of meat are rapidly increasing. This issue may be addressed by appropriate analytical tools. Trace elements might help to discriminate between different origins. Rare and non-essential elements are more promising in that respect since they are typically not supplemented to diets and thus could be region-specific. To test this hypothesis, two different commodities were selected: poultry where most production factors (feed, genotype), except tap water, are widely uncoupled from regional origin and a dried beef product where the production of the raw beef much more depends on geographic origin. Poultry breasts (n=25) were collected from Thailand, France, Germany, Hungary, Brazil and Switzerland. The dried beef meat samples (n=18) originated from Switzerland (two regions and, in one region, raw beef from Switzerland or Brazil), Austria, Australia Canada and USA. The authenticity of the poultry samples was confirmed by official customs documents. Dried beef was either collected at the point of production or imported directly from the producers. Poultry samples were frozen and beef samples were stored at +5°C before being homogenized, subjected to micro-wave assisted pressure digestion with nitric acid and analysed for a total of 75 elements/isotopes using a sector field ICP-MS (Element 2, Finnigan MAT, Bremen, D). Compartment analysis was performed to determine the extent to which the variation among samples was explained by a combination of elements. Accordingly, in poultry 60% of the variation was explained by B, Ca, Co, Ti, while Ca and Ti alone explained 42%. In beef, 86% was explained by Ca, Cu, Li, Pd, Rb, Sc, Sr, Ti, U and V, while Li, Rb and Ti alone explained 80%. Rare earth elements could not be detected in any of the samples. The present results demonstrate a large potential for discrimination, maybe in combination with other methods for meat authentication.

[A16]

Fruits and vegetables as a source of the mineral elements in the food ration planned for nutrition of young men doing military service in the Polish Army

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The aim of the work was to analyse the quantity of fruits and vegetables planned in daily food ration for young men doing military service in the winter season in the Air Cavalry units of the Polish Army. Participation of fruits and vegetables in delivery of selected mineral elements was analysed as well. Total of 50 daily menus, used for young men's nutrition planning were the base for the examination. Using the calculation software FOOD 2 delivery of sodium, potassium, calcium, phosphorus, iron, zinc, copper and manganese with fruits and vegetables was calculated. It was found that 458,5 g vegetables (403-540 g) and 373 g fruits (323-417 g), fresh or processed, were planned for consumption during the winter. Planned vegetables were very different e.g. onion, sauerkraut or bean. Mandarins and bananas besides apples were the fruits included in nutrition planning.

Content of selected mineral elements in planned daily food ration (in mg)

	Sodium	Potassium	Calcium	Phosphorus	Magnesium	Iron	Zinc	Copper	Manganese
Vegetables	566.2	1238.4	175.6	204.5	89.9	3.76	2.32	0.46	0.21
Fruits	8.5	783.9	69.4	68.5	58.5	1.36	0.48	0.35	0.53
Norm (at the safe level)	625	3500	1100	800	350	11	14	2.0-2.5	5

Vegetables met the requirements for sodium in the biggest amount (90.6%) but for the manganese in the lowest one (4.0%). Fruits met the requirements for potassium in the biggest amount (22.4%) and for zinc in the lowest one (3.4%). Planned in the daily food rations quantities of fruits and vegetables exceeded the recommended by WHO amounts (at least 500 g daily).

[A17]

Bioavailability of magnesium ions in aspect of supplementation

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One of the most important parameter which describe the efficiency of drug is their bioavailability. In our studies we determined "in vitro" and "in vivo" the bioavailability of magnesium ions from proposed solid dosage forms. The proposed solid dosage forms contained organic and inorganic magnesium salts. We observed that bioavailability of magnesium ions was good. From previously examinations we calculated that the mean daily intake of magnesium ions was 206 mg. We concluded that in many cases hypermagnesemia states is often.

[A18]

Iodized tea influence in childhood

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Georgia is one among all those countries who somehow face the problem of iodine deficiency. The soil and water in the region are low in iodine due to the geography. Despite efforts in the sub-region iodine deficiency is still a major health problem. In Georgia IDD have always been an endemic disease and this problem is still actual especially in high mountain regions. In 1997-2003 several epidemiological studies by Georgian and foreign specialists showed that in different regions of Georgia the prevalence of goiter was fluctuating 32% and 55% in adults and between 29-78% in children. All above-mentioned made important to find alternative sources of iodization of food products in Georgia.

The most effective method of the fight against iodine deficiency is iodine prophylaxis, based on providing population of iodine-deficit regions with iodide product (salt, bread, milk, butter, tea). It is established that day and night consuming of iodine products containing 100mcg of iodine, decreases the disease of thyroid gland by 50-60% during a year. As Georgia traditionally is so called "tea country" exactly this product was chosen for this purpose. KJ was added in tea by original technology proper amount with innovative way, using special mixer-dosimeter. Georgian National Intellectual Property Center, under number U829 and U828 patent this method.

1. Iodisation doesn't affect tea organoleptic characters.
2. Iodine content in tea is stable; re-iodisation is of no need.
3. This technology gives exact dosing possibility 25 mg iodine per each gram of tea, which allows controlling consumed amount of iodine per person.

Iodized tea could be used as for prevention of IDD as well as alternative way for individual iodine preventive process for those persons, who have medical restrictions and can not use proper amount of table salt. We have examined 228 children of the age 8-12 in children house before becoming iodized tea and after 6 month. Children were in the same social condition we have researched thyroid gland by ultra sonography. And iodine deficiency we determine by iodine excretion in urine. 228 children were decided in two groups: Group I = 158 children with goiter and group II = 70 children without goiter. Before iodized tea using in I group 33% had goiter of I degree and 6% (9child) II degree. Median of iodine in I group was 7.1 mcg% (it's correspond to light deficiency) in the II group – 9.3 mcg% (it correspond to normal index low border). The tea was given to the children of the both groups during 6 month twice a day (100mg iodine) on breakfast and supper. After 6 month the condition in both groups was improved. Frequency of goiter speeding in the group decreased at 16%. In the II group 2 children had no improvement. In 7 children the goiter of II degree increased in goiter of I degree. About iodine excretion in urine the median increased in the I group to 10.9 mcg% ($p<0.05$) and in the II group 12.9 mcg% ($p<0.05$). It means the iodine balance was normal.

As it has been shown in practice the use of iodized tea have definitely positive results on children. It can be used in adult sportsman, hard physical workers and pregnant women and nursing mothers.

[A19]

Influence of iodine and selenium supplementation to the diet of ewes (Blackhead dairy breed) on the milk composition and trace element content of milk

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The study was carried out to establish the effect of Se and I supplementation during the winter feeding of Blackhead sheep breed on the milk performance, trace element contents (Se, I, Zn) and daily secretion of essential nutrients in the raw milk during the first 56 days of lactation (suckling period). Twenty ewes of Blackhead dairy breed, divided into two groups were fed according to ARC norms with different amounts of selenium and iodine during the experiment. The natural base diet included 0.08 mg Se/kg DM and 0.10 mg I /kg DM. The supplemented group received additionally *per os* 2.1 mg Se (as NaHSeO₃) and 2.8 mg I (as KI) every two weeks. The milk samples were taken on the 14th, 28th, 42nd and 56th day of lactation.

The low levels of Se and I in the ration of ewes led to decrease of daily milk production during the first 56 days of lactation by 17% (674 ml/d vs. 813 ml/d). The supplemented animals produced raw milk with lower percentage of fat (6.19±0.46) in comparison with the control group (5.11 ± 0.78). Addition of Se and I led to increase of daily protein secretion by 24.5%. The natural I-Se-deficiency influenced not only the milk performance but trace element contents and the daily Se-, I- and Zn-secretion, as well. The supplemented ewes secreted on the average 2.5-fold more selenium (32.2 µg/day vs. 12.8 µg/day), 4.5-fold more iodine 4.5 (26.6 µg/day vs. 5.8 µg/day) and 1.22-fold more zinc (7.2 mg/day vs. 5.9 mg/day).

The results obtained in the experiment with Blackhead sheep breed during the winter feeding demonstrated clearly the necessity of correction in the selenium and iodine content the ration during the suckling period of lambs.

[A20]

Effect of natural magnesium additives on the rumen fermentation in vitro

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The effect of two types of caustic calcinated magnesite (Caustic magnesite, CM and Agromag, AG) upon the end products of in vitro fermentation (total gas, methane, total and individual fatty acids, VFA) and protozoan population in the rumen fluid collected from sheep. Meadow hay (MH), wheat straw (WS), amorphous cellulose (AC) and barley grain (BG) were used as substrates and incubated with the buffered rumen fluid using an in vitro gas measuring technique during 72 h of incubation. The rumen protozoa, *Entodinium* spp., *Trichostomatids* and large *Entodiniomorphids* and the total protozoan concentration were counted after 24 h of incubation. The methane production was significantly decreased with CM or AG, respectively: by 58% or 62% (MH), by 65% (WS), by 52% (AC), by 58% (BG). The total VFA concentration was significantly lower compared to control for CM plus MH, WS, AC, BG and AG plus WS. The total VFA concentration was significantly higher compared to control for AG plus AC. The effect of the both additives on ciliate population was not uniform and depended on the substrates used and protozoan type. Ciliate population was significantly increased in *Entodinium* spp. (AG plus BG) and *Diploplastron affinae* (CM or AG plus BG) compared to control. Tested additives significantly decreased population of *Entodinium* spp. (AG plus MH or AC), *Dasytricha ruminantium* (AG plus AC), *Ophryoscolex c.tricornatus*, *Eremoplastron dilobum* and *Polyplastron multivesiculatum* (CM or AG plus BG).

[A21]

Rare earth elements as alternative growth promoters for pigs

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Rare earth elements (REE) include the 15 lanthanides La, Ce, Pr, Nd, Pm, Sm, Eu, Gd, Tb, Dy, Ho, Er, Tm, Yb, Lu, and the elements Sc and Y. Despite their name, the REE are in fact not especially rare. Each is more common in the earth's crust than silver, gold or platinum. In China, REE were used in agriculture as fertilizer in plant production and as growth promoter in animal nutrition already for some decades. Many Chinese studies showed partly beneficial growth promoting effects whereas the most spectacular is seen in poultry and pig production. Several feeding trials with REE were already carried out under western conditions. So far mineral salt chlorides were used in most previous studies. In the present experiment we use REE-citrate as dietary supplement. In addition to the investigations on energy and nitrogen metabolism in pigs (see abstract Prause et al., 10th Symposium Jena 2005), the goal of the present study was to test the influence of REE-citrate as potential growth promoter for weaning piglets under Swiss feeding restrictions (focus on safe feed has priority rather than maximum growth rate). The feeding experiments (see Table 1) were carried out under test station conditions with a total of 147

Table 1: Experimental design

weaned Large White piglets (30 days of age) during 5 weeks with 10 replications per treatment. The feed was pelleted and given in dry form in automatic feeders (ad libitum).	Treatment		
	A	B	C
Citrate	100	-	-

Health status of the piglets and feces conditions was not affected by REE-citrate. Growth performance, calculated over the whole period, was not significantly influenced by dietary treatment, although feed conversion ratio was better in treatment B and C compared to A (1 - 4 %). Feed intake and daily weight gain were reduced by REE by 4 - 8 %. Energy and nitrogen digestibility, by using the indicator method, was highest in B and lowest in C. The results of the present study indicate that growth promoting effects of REE under test station conditions were limited and not consistent with numerous other studies, but feed conversion ratio was positively influenced.

[A22]

The impact of rare earth elements on energy-, carbon-, and nitrogen-balance of growing piglets

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The aim of this study was to determine, if Rare Earth Elements (REE) have an impact on digestibility of nutrients and/or an impact on energy, carbon, and nitrogen metabolism of growing piglets. Furthermore the effect of Rare Earth as growth promoter was investigated. For this purpose a field trial with 147 pigs and the same food was performed also (see abstract S. Gebert et al., 10th symposium Jena, 2005). Material and methods: 40 barrows with an initial mean body mass of 8 kg were assigned to three feeding variants. They were fed a conventional farrow feed added with either 100 mg citrate as control diet or a REE - citrate, at doses of 150 mg or 300 mg. Up to a weight of 25 kg the pigs were kept pairwise, later they were housed individually until they were slaughtered at 55 - 65 kg. In each of these two periods the pigs were put for 96 hours in respiration chambers. By analyzing respiration data, feces, urine and feed, it was possible to generate a nutrient and energy balance during these times. Feed intake and body weight data were collected at regular intervals during the fattening. Blood samples were taken periodically and after slaughtering tissues and bone samples were taken. Results: Daily body weight gain was not affected by supplementing REE. Feeding the low dose of REE significantly reduced feed conversion rate almost 8%, because daily feed intake was 9% lower than control. Pigs supplemented high dose of REE reduced feed intake 5% and had 2% better conversion rate than control, but not significant. During both periods of respiration digestibility of energy and nutrients was more efficient in the low dose treatment. In second respiration phase carbon, energy and specially nitrogen retention, proportional to food intake was also increased in this feeding group. But these were not significant. Further studies should be conducted to evaluate REE as a growth promoter and to detect the mode of action.

[A23]

Effects of the multi-element supplementation to the diet of ewes on the reproduction parameters and the growth of female lambs reared in endemic regions of Middle Bulgaria

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The extensive system of husbandry of small ruminants is associated with certain nutrient imbalances due to the irregular mineral supply through natural pastures. The effect of a complex mineral mixture on reproductive parameters and growth in sheep has been tested in a two-stage performed experiment. Twenty five ewes of the South Corriedale breed at the IV lactation were divided into the supplemented (n=13) and the deficient group (n=12). The supplemented ewes received 60 mg Zn, 0.187 mg Se and 0.187 mg J per kg DM in inorganic form. After weaning the female lambs, correspondingly divided into the supplemented (n=7) and the deficient group (n=7), were fed a restricted diet composed of grass hay, alfalfa and concentrates in proportion 30:30:40 over a period of 180 days. The supplemented diet was added with 25 mg Zn, 0.25 mg Se and 0.25 mg J per kg DM.

The supplementation of ewes during pregnancy and suckling period was found to reduce the mortality rate. The neonatal mortality was established to decrease from 38 % in the deficient group to 19% in the supplemented one. The postnatal mortality of the deficient group was accounted to 8%, whereas in the supplemented group it had not been registered. The complex addition of Zn, Se and J was found to change the sex ratio to the more number of male born lambs. The lambs of supplemented ewes had 0.4 kg higher birth weight (P>0.05) and differences were not assessed at weaning.

During the consequent experimental period of 180 days it was established that supplemented lambs had higher average daily gain by 23%. The very low trace element status of the deficient lambs marked trough the tissues concentration was accounted for the growth depression. At the end of experiment, under conditions of restricted diet, the supplemented lambs had higher live weight gain (4.19 kg) reached through more efficient feed utilization (intake/gain) by 29%.

Key words: ewes, lambs, zinc-selenium-iodine supplementation, reproduction, growth

[A24]

Organic acids in animal feeds – improving the decision making process*(Organische Säuren in Futtermitteln - den Entscheidungsprozess verbessern)*Keller S¹, Parker DS², Buttin B², Schasteen C³¹Röthel GmbH, Gudensberg, Germany²Novus Europe, Brussels,³Novus International Inc, St Louis, MO, USA

The EU-wide ban on the use of in-feed sub-therapeutic levels of antibiotics has focussed interest on the role of organic acids in animal feeds. Apart from optimising feed hygiene there is also the potential for encouraging a favourable microflora in the digestive tract. There are a number of factors to be considered when developing a strategy for acid use:

- 1) Level of inclusion that will ensure a consistent response – in some cases the actual concentration and acid composition in commercial blends is not stated providing no information upon which to make decisions on the optimum inclusion rate. 2) Single acid or blend – blends of methionine hydroxy analogue (Alimet[®]) and formic acid for example, have been shown at acid pH *in vitro* to be significantly more effective against *Salmonella enteritidis* than the individual acids under the same conditions. 3) Use of free acid or salt – whereas for handling the use of acid salts has advantages data from *in vitro* experiments show that acid salts may be less effective at modifying the microbial population than inclusion of the free acid in feed. 4) Encapsulated or protected acids – this approach will only be cost effective if the protective matrix is truly acid resistant and also if the blend of acids released is active at the pH of the target intestinal site. For example, incubation of *S. enteritidis* at pH 7.0 (Initial Optical Density (OD) at 600nm of 1.06) with either sorbic or fumaric acids at 0.5% resulted in final ODs of 0.74 for formic acid and 0.22 for sorbic acid, indicating that sorbic acid was more effective than formic acid at this pH.

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[A25]

The antimicrobial and acidification effects of ACTIVATE WD, an organic acid blend containing ALIMET[®] with other organic acids, in poultry drinking water*(Antimikrobielle und ansäuernde Effekte von ACTIVATE WD, einer Mischung organischer Säuren bestehend aus ALIMET[®] mit anderen organischen Säuren, im Tränkwasser für Geflügel)*Schasteen C¹, Parker DS², Keller S³, Wu J¹, Davis M¹, Fiene S¹¹Novus International, Inc., St Charles, MO 63304²Novus Europe, Brussels³Röthel GmbH, Gudensberg, Germany

The antimicrobial property of DL-2-hydroxy-4-(methylthio) butanoic acid (HMTBA or Alimet[®]) has been previously documented (Hao and Schasteen, 1999; Enthoven 2002) in culture broth. We have also evaluated the effect of formic, fumaric, propionic, butyric, lactic, HMTBA and HMTBA-containing organic acid blends (Activate[®]) on *Salmonella* growth in feed using a simulation of conditions (moisture and pH) encountered in the stomach and proximal to the small intestine. The addition of HMTBA to individual or a blend of organic acids resulted in a more than additive increase in antibacterial effect against *salmonella* in this assay. We have also done field testing of our HMTBA-containing organic acid blend for the drinking water, Activate WD. Three trials (1 22K broiler house treated and 1 untreated per trial) at different locations were conducted with Activate WD (0.04-0.08%, pH ~3) included in drinking water for the last 10-14 days prior to processing. Cloacal swabs of birds were positive for *salmonella* prior to starting Activate WD while swabs taken after treatment and prior to transporting the birds to the processing facility were found to be *salmonella* negative. This does not guarantee the chickens have no *salmonella* present in their intestine but indicates that they were not shedding. Live weight increases were seen in all 3 trials with an average increase of 140.7 grams/bird compared to non-treated houses. Activate WD treated birds water intake was not different than untreated controls. The presence of HMTBA-containing organic acid blends in the feed and or water provide antimicrobial and acidification benefits.

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[A26]

Interest for utilizing a single multi-enzyme preparation on different types of diets from post-weaning piglets to slaughter pigs

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The interest to utilize a single multi-enzyme preparation (Rovabio™ Excel, Adisseo, France) in order to increase the nutritive value of swine diets was examined by means of different digestibility and performance trials. Different dietary regimes, namely wheat-, wheat/barley- or corn-based diets, as well as different physiological stages, from post-weaning piglets up to slaughter animals, were studied. For each variable two studies were conducted by changing the dietary compositions. In piglets, faecal digestibilities of Crude Energy and Crude Protein were determined using either total faecal collection or grab samples of faeces by means of determination of an indigestible marker. Digestibilities in growing pigs were determined on an ileal level using ileal-rectal anastomosed animals. Performance trials were performed with animals kept either individually or grouped in floor pens, according to classical performance trials. Data were analysed using the ANOVA or ANCOVA procedure of Statview.

The conducted trials proved that by the utilization of a single multi-enzyme preparation it is possible to increase energy digestibility (+ 2% in piglets fed corn- or barley-based diets; 4.4% and 2.8% for fattening pigs when fed diets based on barley or wheat, respectively). Moreover, animal performance was improved (6-7% in average daily gain for piglets; 3.6% and 4.6% for fatteners) independently of the cereal raw material used to formulate the experimental diets and the physiological stage of the animals. With a single multi-enzyme preparation it is therefore possible to increase the digestibility of different types of diets and therefore the animal performance. This effect can be observed for piglets as well as for growing-finishing pigs. For the nutritionist it is therefore possible to change feed raw materials or to formulate feeds for animals in different physiological stages without changing the enzyme preparation.

[A27]

Effects of the phytogetic feed additive FRESTA F Conc. in weaned piglets

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Ninety six crossbred piglets (Landrace x Yorkshire x Duroc), averaging 5.5 kg in body weight, were allocated to four treatments and fed on a control diet (NC = negative control, no antibiotics, no phytobiotics) or the control diet supplemented with either a combination (CTC + Sulfathiazole + Penicillin) of antimicrobial growth promoters (PC = positive control, 0.1% antibiotics) or a phytogetic feed additive (NCF = negative control + 0.03% FRESTA F Conc.) or antibiotics plus FRESTA F Conc. (PCF = positive control + 300ppm FRESTA F Conc.) over a period of 49 days. The addition of antibiotics and FRESTA F Conc. numerically increased daily weight gain (3% and 7%, respectively) and reduced feed conversion ratio (3% and 4%, respectively) compared with NC. There was a synergistic and significant effect of antibiotics and phytobiotics on growth performance. The improved growth performance of piglets can -at least- partly be explained by an increased nitrogen digestibility in the PC (+2%), the NCF (103%), and the PCF (105%) diet compared with the NC diet. The immune response in piglets was also improved by the feed additives. Piglets fed the PC and NCF diets had numerical higher counts for red (RBC) and white blood cells (WBC) than piglets fed the NC diet after 49 days in experiment. WBC and RBC were even higher in the PCF treatment and statistically different from the negative control. There was also an increase in the concentration of immunoglobulin G (IgG) due to the addition of the feed additives. PC and NCF significantly increased IgG levels by about 25% and PCF by about 58%, respectively. Total protein and albumin in blood were not different between treatment groups. There were no differences in faecal acetic, butyric, and propionic acid production, but ammonia nitrogen and hydrogen sulphide were reduced by the feed additives up to 29% (NH3-N) and 24% (H2S), respectively. It is concluded that the phytogetic feed additive FRESTA F Conc. can be used to replace antibiotic growth promoters in piglets without negative effects on performance.

[A28]

Production of prebiotic meat products and sausages*(Herstellung prebiotischer Fleisch- und Wurstwaren)*

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Auch der aktuelle Ernährungsbericht 2004 der Deutschen Gesellschaft für Ernährung (DGE) e. V. zeigt, dass für bestimmte Lebensmittelinhaltsstoffe die empfohlene tägliche Aufnahmemenge, welche die Funktionalität eines gesunden menschlichen Körpers gewährleisten soll, unterschritten wird. Über eine ausgewogene Ernährung mit viel Obst und Gemüse sowie Ballaststoffen ist ein „normaler“ gesunder Verbraucher durchaus in der Lage, sich ausreichend mit den benötigten Lebensmittelinhaltsstoffen zu versorgen. Durch eine Anreicherung von Lebensmitteln mit bestimmten Inhaltsstoffen steht jedoch die Möglichkeit zur Verfügung, gerade solchen Verbrauchern eine ausgeglichene Ernährung zu ermöglichen, die derer im Besonderen bedürfen. Der Schwerpunkt der vorliegenden Forschungsarbeit liegt in der Anreicherung von Fleisch- und Wurstwaren (Brüh-, Koch- und Rohwurst) mit essentiellen Inhaltsstoffen wie z. B. Ballast- und Mineralstoffen. Hierbei kommen Weizen- und Haferfasern, ein Selen-Zink-Mineralstoffkomplex und unter anderem auch Spirulina-Algen und Fruchtkernpresskuchen zum Einsatz. Untersucht werden soll, welche Auswirkungen eine derart veränderte inhaltsstoffliche Zusammensetzung auf die technologische Verarbeitbarkeit der Wurstbräte und auf die daraus hergestellte Produktqualität sowie die sensorische Eignung, die im speziellen Aufschluss über die Verzehrs- und Verzehrfähigkeit der hergestellten Erzeugnisse gibt, ausübt. Als Ziel steht die Optimierung der inhaltsstofflichen Zusammensetzung und der jeweiligen technologischen Verfahrensführung, um einen hohen ernährungsphysiologischen Status der Fleisch- und Wurstwaren gewährleisten zu können und prebiotische Effekte zu ermöglichen.

[A29]

Influence of probiotics on immunological parameters of healthy volunteers and patients with atopic dermatitis*(Der Einfluss von Probiotika auf immunologische Parameter gesunder Personen und Patienten mit atopischer Dermatitis)*Klein A¹, Vogelsang H², Jahreis G¹¹Institute of Nutrition, Friedrich Schiller University Jena;²Institute of Clinical Chemistry and Laboratory Diagnostics, Friedrich Schiller University of Jena

The supporting role of probiotics in the prevention and therapy of atopic diseases is widely described. The effect is based on the interaction of the colonic microflora with the gut associated lymphoid tissue. Recent studies have shown that in patients with atopic dermatitis exists an aberrant gut microbial composition. Phagocytic receptor activity of granulocytes, as a marker for unspecific cellular immune response, is down regulated in these patients whereas a stimulation of this parameter is observed in healthy subjects. In the present study we investigated the effects of *Lactobacillus paracasei* LPC37 on the immune system of 15 healthy subjects and 15 patients with moderate atopic dermatitis. The study was designed as a placebo-controlled and cross-over trial. Volunteers were randomized into two groups. They consumed either 200 ml/d probiotic or placebo yoghurt drink over 8 weeks. After a 2 week wash-out period intervention was changed between the groups. Blood and stool samples were collected at the beginning and the end of each period

Immunological parameters (selected clusters of differentiation) were measured via flow cytometry (FACScan, CELLQUEST software). Phagocytic activity of granulocytes was determined using the commercial test kit Phagotest®. The percentage of granulocytes that ingested FITC-labelled opsonized *Escherichia coli* was detected. Final results will be shown and discussed at the presentation.

[A30]

On the influence of β -glucans from *Saccharomyces cerevisiae* on sow and litter performance during lactation

(Zum Einfluss von β -Glukanen aus *Saccharomyces cerevisiae* auf die Leistung von Sauen und deren Ferkeln)

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In search of alternatives for antimicrobial growth promoters various feed additives are used in farm animal feeding to improve animal health and performance. Among these are 1,3/4,6- β -D-glucans extracted from yeast cell walls. They are supposed to stimulate the immune system thus enhancing animal performance as it was shown in previous studies with various types of animals.

To prove this, a trial was conducted feeding yeast cell wall extracts to 15 sows from a commercial herd between d112 of gestation and weaning (d21). The animals were divided into 3 groups of 5 sows each in the experiment and housed into individual farrowing crates. Diets consisted of a standard barley-soybean meal (13.0 MJ ME/kg, 17.5 % XP) and were supplemented with 0 g (control), 4 g (group 1) or 2.2 g (group 2) β -glucan per sow and day, respectively. From d12 of gestation until farrowing sows were fed restrictively with 1.5 kg of the lactation diet which was then increased daily by 700 g until *ad libitum* intake from d6 of lactation on. Creep feed was offered to suckling piglets starting on d10 after birth. Sows and piglets had free access to tap water.

The number of piglets born alive varied insignificantly between 10.2 (control group) and 12.2 (group 2). However, these differences were not due to β -glucan-supplementation so that cross-fostering occurred within 24 h *post partum*, irrespective of treatment. Nonetheless, there was a positive tendency towards higher litter weight at weaning for group 1 amounting to 11.9 kg compared to the control group ($p > 0.05$). Moreover, litter weight gain was 8.5 kg higher than in the control group ($p > 0.05$).

The results show that under practical farming conditions improved animal performance may be possible by use of β -glucans derived from yeast cell walls. However, further research is necessary to understand the immunological mechanisms forming the basis of this amelioration.

[A31]

Influence of Flavomycin® and BioPlus 2B on the rearing result of calves

(Einfluss von Flavomycin und BioPlus auf das Aufzuchtresultat bei Kälbern)

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The effects of a probiotic additive (BioPlus 2B, 400 mg/kg milk replacer; group 2) in contrast to Flavomycin treated calves (10 mg/kg milk replacer, group 1) on feed intake, live weight gain, animal health and different parameters of blood were tested in a feeding trial involving 40 calves. At the beginning of the trial the calves were 21-28 days old. In addition to the milk replacer (maximum 110 g/l water and a quantity of 7 l) they were fed with growing feed for calves *ad libitum* as well as dried chopped grass and straw (maximum 0.7 kg/animal per day).

The table shows that the application of BioPlus 2B in place of flavomycin resulted in none effects on the intake of dry matter, crude protein and metabolizable energy.

Table: Influence of the BioPlus and Flavomycin on the rearing result of calves ($p > 0.05$)

Group Preparation Dose	Dimension mg/kg milk replacer	1 Flavomycin® 10	2 BioPlus 2B 400
Dry matter	kg/animal/day	2.07 ± 0.22	1.92 ± 0.29
Crude protein	g/animal/day	431 ± 0.50	399 ± 0.60
Metabolizable energy	MJ ME/animal/day	26.6 ± 2.6	24.7 ± 3.3
Average daily gain			
1. - 28. day		606 ± 170	548 ± 215
1. - 56. day	g/animal/day	776 ± 155	713 ± 202
1. - 84. day		929 ± 130	866 ± 154
Diarrhea, 1. - 28. day	day/animal	2.64	2.90
Blood			
Leucocytes	x 10 ³ mm ³	11.0	9.9
Aspartataminotransferase	nkat/l serum	816	852

The group treated with BioPlus 2B shows a lower live weight gain of about 7% in contrast to the Flavomycin treated group over the whole experimental period (1. - 84. day of trial), in the first 28 days even a 10% lower live weight gain ($p > 0.05$). All measured blood parameters showed normally values and were not affected by the preparation.

[A32]

Effects of BioPlus 2B supplementation on piglets*(Auswirkungen einer BioPlus 2B-Supplementation bei Ferkeln)*Richter G.¹, Arnhold W.², Hartung H.¹, Leiterer M.¹¹ Thuringian State Institute of Agriculture, Auf der Höhe 6, D-07751 Jena OT Remderoda² BASU-Mineral Feed Inc., Bergstr. 2, D-99518 Bad Sulza

In three experiments the effect of the probioticum "BioPlus 2B" was investigated on 126 fattening castrates during the 28th – 70th day of life. The piglets were kept on fully slatted floor in flatdeck style. Bio Plus 2B is a mixture of Bac. licheniformis and Bac. subtilis in ratio 1 : 1. It was supplemented to the diet with 0.4 g / kg during the whole experiment. The diet of the control group did not contain performance enhancers. The piglets received a pelleted starter diet during the 5th and 6th week of life, and a pelleted growing diet during the 7th and 10th week of life. The diets were offered ad libitum. The experiments were designed that in the experiment A and B the piglets of each group were kept in 6 boxes, and in experiment C in 9 boxes with three animals. The results were statistically assessed with the standard deviation and the Tukey-test.

No tendency was observed that the supplementation of the diet with Bio Plus 2B had an effect on zootechnical parameters. Piglets with Bio Plus 2B supplement consumed 5.4% more feed in the 5th and 6th week of life compared to the control animals, but the feed intake did not differ between the groups in the 7th up to the 10th week. That's why the feed intake was only insignificantly increased by 0.7% in the Bio Plus 2B supplemented group during the whole 6-week-period of investigation.

The life weight gain followed the results of the feed intake on average of the three experiments. The piglets with Bio Plus 2B supplement gained 7.9% more weight in the starter period than the non supplemented animals. However, the differences remained insignificant. Due to the fact that in the following period the piglets with Bio Plus 2B supplementation had a lower life weight gain than the control piglets the same life weight gain was detected in both groups (438 g vs. 437 g). Bio Plus 2B improved the feed efficiency by 2.5% in the first two weeks of investigation. However, when the whole period was regarded no differences were observed between the groups without and with Bio Plus 2B supplementation (1.37 vs. 1.38).

It could be demonstrated that the Bio Plus 2B supplementation of the starter diet of piglets improved feed intake by 5.4%, life weight gain by 7.9 %, and feed efficiency by 2.5%. Unfortunately, this positive tendency did not continue in the following period of the 7th up to the 10th week of life. Furthermore, the positive results of experiment A could not be reproduced in experiment B and C. The piglets were in a good health status. No disturbances were observed. The feeding did not take effect on mortality. However, there was a tendency of a lower occurrence and intensity of diarrhoea in Bio Plus 2B supplemented piglets.

The results of these three experiments demonstrate that the supplementation with Bio Plus 2B of the diets of piglets did not improve the economical result.

[A33]

The influence of Cr⁺³ picolinate, nicotine and chloride on apoptosis in human mammary epithelial cells (HBL-100) and breast cancer cells (MCF-7).

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Two lines of human mammary epithelial cell were used; normal (HBL-100) as well as tumor cells (MCF-7). Cells were exposed to different Cr⁺³ compounds in amount of 10 µg Cr/l. Apoptosis was quantified by fluorescence microscopy with 7-aminoactinomycin D staining. Bax and poly(ADP-ribose)polymerase (PARP) expression were estimated by LSC methods. Chromium picolinate, most popular Cr nutritional supplement, used for 6 h leads to increased cell death. Cells from measured area showed typical features of apoptosis such: cell shrinkage, chromatin changes (condensation and margination), nucleus pyknosis and apoptotic body formation. The rise of apoptotic cells % in sub G1, increased amount of aggregated DNA, was observed in studied cell lines as a function of time exposure. Significant increase of proapoptotic Bax protein expression was noticed as well as PARP, known as enzyme participating in damaged DNA repair. Picolinic acid also was found to increase the DNA damage recognized as increased cell number in sub G1 phase and participation in apoptotic protein expression, however this influence was much lower than shown by chromium picolinate. Fact that only Cr picolinate but not Cr nicotine or CrCl₃ possesses such properties suggests importance of ligand in apoptosis induction by chromium (III). Picolinic acid also cause slight increase of cell % in phase sub G1. Obtained results indicate, that even small concentrations of Cr picolinate may stimulate apoptosis in human epithelial mammary cells.

[A34]

The *in vitro* effect of chromium Cr⁺³ and selenium (Se⁺⁴ and Se⁻²) ions on carnitine uptake and fatty acids β -oxidation efficiency in rat's lymphocytes.

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The *in vitro* effects chromium Cr⁺³, selenium Se⁺⁴ and selenium Se⁻² ions on carnitine uptake as well as of carnitine supplementation on the activity of β -oxidation in healthy rat lymphocytes were studied. Chromium ions added as chromium acetate (96.15 μ M = 5.0 ppm) stimulate carnitine uptake and exhibits additive effect on stimulation of this process by selenium Se⁺⁴, but not by selenium Se⁻² (6.33 μ M = 0.5 ppm each). It seems that the mode of action of inorganic Se⁺⁴ added in the form of sodium selenite and organic Se⁻² used in the form of seleno-methionine differs. There is not additional effect of chromium on carnitine uptake in the presence of Se⁻² ions. Fatty acids degradation in the absence of exogenous carnitine is stimulated only by Cr⁺³. Other combinations of ions were ineffective. After supplementation with 50 μ M carnitine activation of β -oxidation was observed in all experimental variants.

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