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Use of Benzophenanthridine Alkaloids as Animal Feed Additives and Remedy for Intestinal Inflammation

This invention concerns the use of one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs as animal feed additive for inhibiting or preventing the enzymatic decomposition of the essential aromatic amino acids in an animal feed, the animal feed as well as the use of benzophenanthridine alkaloids for the production of a remedy.

Aromatic amino acids, e.g. tryptophane and alanine are essential nutrients that cannot be produced by the body itself, except in the case of ruminants. They are an important component of a recipe for food and animal feed. In the animal feed they serve - like other amino acids as well - to ensure, in a certain proportion to the content of other essential amino acids, the necessary balance between the individual essential and non-essential amino acids. Their use incurs cost.

In the intestines of mammals, micro organisms are widespread, which are able to decompose amino acids with the help of their enzymes, e.g. the aromatic amino acids decarboxylase. As a result, the balance between the individual amino acids, that is initially used as a basis for the preparation of animal feeds, is no longer given. The meat quality of animals is reduced.

Indole and skatole, which cause undesired characteristics and effects, are created as intermediate products of the enzymatic decomposition of aromatic amino acids. Indole and skatole are toxic substances of the decomposition of tryptophane. Among other effects they are a burden on the liver, because the body has to detoxify these substances, in order not to become ill because of them. Indole and skatole lead to chronic, sub-clinical inflammations of the tissue of the mucosa of affected mammals and human beings. Indole and skatole, produced by micro organisms of the intestinal mucosa, facilitate the entry (invasion) of pathogenic parasites (intestinal helminths, clostridia, salmonella, coccidia) into the upper cells of the intestinal mucosa and reduce their defence readiness respectively.

In addition, residues of indole and skatole in the food may also unfavourably influence the consumer with regard to his health or in his well-being. An unfavourable relation between aromatic amino acids (in relation too low a quantity available) and the other essential amino acids leads to an imbalance, which causes a reduced utilisation of protein and a reduced content of lactalbumin. Food quality is also measured on the basis of protein content. Protein content (lactalbumin, lean meat) that is reduced due to the lack of aromatic amino acids leads to reduced quality and a reduction in prices.

In connection with Parkinson's disease in human beings, aromatic amino acid decarboxylase is seen as a possible cause that leads indirectly to the disease. A great number of micro organisms that live in the intestine and intestinal mucosa of animals produce enzymes that decompose protein and amino acids. The situation becomes critical, if essential nutrients are affected by this decomposition process and if furthermore toxic decomposition products are created that place a burden on the host or lead to residues in the foodstuffs. The cost of the feed optimisation is increased insofar as the loss of aromatic amino acids, which until now was unavoidable, was taken into consideration via a higher admixture to the feed. As consequence of the creation and the uninfluenced activity of the aromatic amino acid decarboxylase, the cost of feeding and of maintaining the health of economically useful animals and pets is increased.

The animal feed in accordance with the invention is first of all characterised by the fact that it contains an effective quantity of one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs in order to reduce the enzymatic decomposition of the essential aromatic amino acids.

The use of Sanguinarine and other benzophenanthridine alkaloids as performance enhancers for economically useful animals, the utilisation of these alkaloids for the treatment and prophylaxis respectively against salmonella infection, against coccidia and other bacterial germs and for the conservation of animal feed is known from WO-A-9 315 602 (Neufeld). In it the utilisation of these alkaloids for inhibiting or preventing the enzymatic decomposition of the AAAD (aromatic amino acids decarboxylase) is not mentioned.

Drsata et al., *J. Enzyme Inhibition*, 1995, Vol. 10, pg. 231-237, report on the effect of Sanguinarine as an inhibitor of AAAD, which plays an important role in the biosynthesis of dopamine, noradrenaline, adrenaline and serotonin. As dopamine is important in connection with Parkinson's disease and as noradrenaline is a potent vasoconstrictor and serotonin is counted among the inflammation mediator substances, the inhibition of AAAD is the subject of the study. The insight gained on the basis of it, does not suggest the use of benzophenanthridine alkaloids for the improvement of the protein result for economically useful animals or their use as remedy in the area of veterinary medicine.

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In accordance with the invention, one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs are used as animal feed additives in an effective quantity for inhibiting or preventing the enzymatic decomposition of the essential aromatic amino acids in an animal feed or animal feed additive. In accordance with another characteristic of the invention, the benzophenanthridine alkaloids can also be used in the form of plant material of the Papaveraceae or as extract thereof or in the form of the isolated alkaloids or alkaloid mixtures or synthetic analogs.

In accordance with another characteristic of the invention, one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs or plant components of the Papaveraceae containing benzophenanthridine alkaloid(s) for the manufacture of animal feeds or animal feed additives for producing such animal feeds are used for the improvement of the protein result in economically useful animals through the inhibiting or preventing of the enzymatic decomposition of aromatic amino acids in the digestive tract.

Another object of the invention is the use of benzophenanthridine alkaloids or their derivatives or synthetic analogs or of plant components of the Papaveraceae containing benzophenanthridine alkaloid(s) for the manufacture of a remedy for intestinal inflammation. In particular those that are caused by decomposition products of the amino acid metabolism, such as indole and skatole.

Chelerythrine and Sanguinarine as well as other benzophenanthridine alkaloids have been found to be potent inhibitors of the aromatic amino acid decarboxylase. It has been shown that Sanguinarine and Chelerythrine block the activities of the aromatic amino acid decarboxylase to a degree of 99 % and 92 % respectively. This blockade of the aromatic amino acid decarboxylase is irreversible and therefore of particular interest, because it will continue for as long as the enzyme molecule are within the effective range of the benzophenanthridine alkaloids. Sanguinarine and Chelerythrine, representatives of the benzophenanthridine alkaloids, terminally have an aromatic structure that is similar to that of the aromatic amino acids. Probably, the enzyme 'Aromatic Amino Acid Decarboxylase' confuses the terminal structure of the benzophenanthridine alkaloids with the ring structure of the aromatic amino acids and

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5 installs those in its active centre instead of the amino acid. As benzophenanthridine alkaloids do not have a carbon skeleton that can be split off, as is the case for the amino acids, there is no end product of the enzymatic reaction, for which reason it cannot be completed. The active centre of the aromatic amino acid decarboxylase is irreversibly blocked. The invention shows that it is possible to irreversibly block the activity and thus
10 the metabolic products of the aromatic amino acid decarboxylase via the admixture of a structure that is similar to the aromatic structures of the aromatic amino acids and to use this to advantage for animal feeds and for remedies.

The invention leads to a reduced decomposition and loss of essential aromatic amino
15 acids and thus to the improved supply of these nutrients to the animal. Tests have shown that as a result of this the content of lean meat and the content of lactalbumin can be clearly, in part significantly, improved. Further tests have shown that through the balance within the amino acid pattern that is improved in this way the protein balance for pigs is improved by up to 10 % in the final fattening stage.

20 The invention leads to improved animal health. Tests show that the use of the benzophenanthridine alkaloids results in a clearly reduced inflammation frequency in the intestines of animals.

25 The animal feed in accordance with the invention can contain all traditional feed substances that are common in animal husbandry. This includes starch-containing components such as grain, grain products and maize. The other proteins contained can be of natural origin or can be added as preparations, which applies in particular to the essential amino acids. Furthermore, there may be mineral additives such as trace
30 elements, phosphates, salts and calcium. Other additives such as enzymes, vitamins or substances acting as medicaments can also be added.

The amino acids contained in the animal feed can either or in part be natural in origin or artificial additives. Important aromatic amino acids are for example thryptophane and
35 phenylalanine.

In nature benzophenanthridine alkaloids occur for example in the rhizomes of the Canadian blood-wort *Sanguinaria Canadensis* or in *Chelidonium majus* or *Macleaya cordata* as well. These plants belong to the *Papaveraceae*. The rhizomes or the leaf
40 material or the entire plant material can be collected and dried. On the basis of the dried substance, the content of benzophenanthridine alkaloids usually is approx. 4 % of the weight. In order to compensate for fluctuations in harvests, it may be advantageous to adjust the content of the main alkaloid Sanguinarine to a value of, for

5 example, 1.5 % of the weight through the addition of filling materials. As approx. half of the natural alkaloid content comes from Sanguinarine, the overall alkaloid content thus would lie at approx. 3 % of the weight.

10 Other alkaloids of this group are Chelerythrine, Chelirubine, Sanguirubine, Chelilutine and Sanguilutine.

Within the framework of this invention it may also be advantageous to administer extracts of the plant material or of the isolated alkaloids and their derivatives or their synthetic analogs.

15 The quantity of alkaloid contained in the feed is limited downward only by effectiveness. Preferably, the total quantity of alkaloids per ton of animal feed will be greater than 1 mg. On the basis of the standardised, dried Papaveraceae root material, this corresponds to approx. 0.067 g per ton of animal feed.

20 In the animal feed fed and up to now examined in more detail, quantities between 5 g and 150 g of dried Papaveraceae per ton of animal feed, standardised to 1.5 % alkaloid Sanguinarine, corresponding to approx. 3 % total alkaloid, were administered.

25 This invention also comprises animal feed additives, such as for example pre-mixtures or mineral feeds or supplementary animal feeds for the preparation of animal feed, whereby the additive, in addition to the usual animal feed additives, contains benzophenanthridine alkaloids or their derivatives or analogs or plant components of Papaveraceae containing these alkaloids. Thus the invention comprises the animal feed itself, which is fed to
30 animals, as well as preliminary products, which are used by the user to produce the finished animal feed.

The invention furthermore concerns the use of benzophenanthridine alkaloids or their derivatives or analogs or plant components of Papaveraceae containing these alkaloids
35 for inhibiting or preventing the enzymatic decomposition of aromatic amino acids, in particular in the digestive tract of animals, in particular in combination with one of the animal feeds or animal feed additives described above.

If the animal feed contains a sufficient quantity of essential amino acids, then the
40 admixture of alkaloids can save the additional administration of expensive aromatic amino acids. If aromatic amino acids have to be admixed, e.g. thryptophane and phenylalanine, then the added quantity can be reduced due to the alkaloid, whereby cost is reduced. On the basis of the current market situation, savings of approx. 10 EURO

- 5 animal feed price can be assumed as guideline value, if in accordance with the invention alkaloid preparations with a value of 1 EURO are admixed to the animal feed.

- 10 In accordance with the invention the benzophenanthridine alkaloids can also be used to advantage as remedy for intestinal inflammations, in particular those that have been caused by decomposition products of the amino acid metabolism, such as indole and skatole. The remedy is characterised by the content of an effective quantity of benzophenanthridine alkaloids or their derivatives or synthetic analogs or plant components of Papaveraceae containing these alkaloids.

- 15 Preferably, the use as remedy is effected together with animal feeds or as additive to the drinking water. The effective quantities of the alkaloids lies within the same range as well.

Several execution examples are described in the following.

- 20 1. Example of a traditional animal feed recipe for chickens and pigs

(in weight percent)

Wheat	42 %
Maize	20 %
Sun Flower	3.6 %
Whole Meal Soy I	4 %
Whole Meal Soy II	16.6 %
Whole Meal Rape	4 %
Fodder Oil	6.5 %
Methionine	0.26 %
Lysine	0.48 %
Mono Calcium Phosphate	0.85 %
Calcium	0.5 %
Salt	0.025 %
Pre-mix	1.0 %
Enzymes	0.2 %
<u>Amino Acids:</u>	
Protein	19.2 %
Lysine	0.824 %
Methionine	0.276 %
Meth + Cystine	0.588 %
Threonine	0.8 %
Thryptophane	0.24 %

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5 2. Example in accordance with the invention

The animal feed for pigs corresponds to Example 1. The quantities of ad-mixed Papaveraceae are stated in grams of dried root material with 1.5 weight % alkaloid Sanguinarine per ton of animal feed. Animal breed: Great Yorkshire + Cross Breeding
10 Sows, 23-24 animals per group.

Groups of Tested Animals		a	b	c
Papaveraceae g/t	Start / Pre-Fattening 22 kg - 48 kg	0	0	15
	Final fattening 48 - 109 kg	0	30	30

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Result:

Classification and Slaughtered Body Value
(AA better than A better than B)

	0 control	Papaveraceae Doses g/t	
		Pre-Fattening / Final Fattening	
	(0/0)	(0/30)	(15/30)
Lean meat %	54.4	55.1	55.0 NS
Back bacon mm	17.8	17.7	17.6 NS
25 Muscle area	51.9	55.6	54.2 (p < 0.05)
Class AA + A %	77 %	96 %	92 %
Class B	23 %	4 %	8 %

Thus the share of Class AA+A is significantly increased in accordance with the invention.

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3. Example

Groups: 0 control 30 g Papaveraceae / t Animal Feed

35 144 piglets at the start, 72 per group, distributed to
8 repetitions per group
Restrictive feeding

Pre-Fattening / Final Fattening 31 - 115 kg:

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	0 control	Papaveraceae 30 g/t
Lean meat (FOM) %	54.9	55.2
Muscle area cm ²	51.7	52.0
Back bacon cm	20.6	20.1

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Economy : Basis 1.022 Euro / kg Slaughtered weight

Slaughtered body value Euro + 1.53

Papaveraceae Advantage Euro + 2.71

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Comment:

In a system with restrictive feeding, Papaveraceae increases the value of the slaughtered body by increasing the share of lean meat and muscle area and decreasing the back bacon value. Thus Papaveraceae increases the meat quality and the profitability by approx. 1.53 Euro per animal.

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4. Example:

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Poultry Feed :

5 groups of 28,000 birds each

Coccidiostaticum Salinomycin

25

Control

Papaveraceae

15 ppm Virginiamycin (Antibiotic)

50 g / t

Parasite spores in 1000 / g muck

(Coccidia-ocysts)

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The test showed a clearly more favourable evaluation of the intestinal inflammations and clearly reduced excretion of intestinal parasites.

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Patent Claims

1. Use of one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs as animal feed additives in an effective quantity for inhibiting or preventing the enzymatic decomposition of essential aromatic amino acids in animal feed or animal feed additives.
2. Use in accordance with Claim 1, characterised by the fact that the benzophenanthridine alkaloids are used in the form of plant material of the Papaveraceae or as extract thereof or in the form of the isolated alkaloids or alkaloid mixtures or synthetic analogs.
3. Use in accordance with Claim 1 or 2, characterised by the fact that the animal feed contains essential aromatic amino acids and benzophenanthridine alkaloids.
4. Use in accordance with Claim 1 to 3, characterised by the fact that the aromatic amino acids are additionally added to the animal feed.
5. Use in accordance with one of the preceding Claims, characterised by the fact that the quantity of alkaloids is greater than 1 mg per ton of animal feed.
6. Use of benzophenanthridine alkaloids or their derivatives or synthetic analogs or of plant components of the Papaveraceae containing benzophenanthridine alkaloid(s) for the manufacture of animal feeds or animal feed additives for producing such animal feeds for improving the protein balance in economically useful animals by inhibiting or preventing the enzymatic decomposition of aromatic amino acids in the digestive tract.
7. Use of benzophenanthridine alkaloids or their derivatives or synthetic analogs or of plant components of the Papaveraceae containing benzophenanthridine alkaloid(s) for the manufacture of a remedy against intestinal inflammations, in particular those that are caused by decomposition products of the amino acid metabolism, such as indole and skatole.
8. Animal feed, which contains traditional animal feed materials such as grain or grain products, maize, proteins and aromatic amino acids, vitamins, mineral additives such as salts, phosphates, calcium, enzymes and such like, or animal feed additives for the manufacture of such animal feeds, characterised by the fact that it contains one or several benzophenanthridine alkaloids or their derivatives or synthetic analogs in an effective quantity for the reduction of the enzymatic decomposition of the essential aromatic amino acids.

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