

## CONTAMINATION OF ANIMAL FEED WITH FUNGI AND BACTERIA IN SLOVENIA

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In the period from 1993 to 1997 the contamination of feedstuffs with *Salmonella*, *Clostridia*, other pathogenic bacteria, mesophilic aerobic bacteria, moulds and yeasts was examined. Among 2442 examined samples, 8.2% of them were unsuitable because of too high contamination with fungi, 2.1% samples (out of 1947 examined) contained too many yeasts spores and 1.8% samples (out of 1591 examined) were unsuitable because of too high contamination with mesophilic aerobic bacteria. *Salmonella* was found in 1.9% samples (out of 2005 examined feeds), 3.1% samples (among 351 examined) contained too high amount of *Clostridia*, and 5.5% samples (among 55 examined) contained other pathogenic bacteria.

Among 796 samples of complete feeding stuffs for poultry inspected on mesophilic aerobic bacteria 10 (1.3%) of them had too high content of bacteria, 42 (4.8%) samples (out of 882) of feedstuffs for poultry were unsuitable because of too high contamination with fungi and 8 (1.1%) samples (out of 706) were unsuitable because of too high contamination with yeasts. In respect to microbiological status pelleted feedstuffs for poultry are of much higher hygienic quality than unpelleted feedstuffs.

### INTRODUCTION

The quality of a feedstuff is not only dependent on the content of nutrients, the digestibility, and biological availability of the constituents but also on its hygienic condition. The term "hygienic status" of a feed comprises contamination with microorganisms as they occur in nature, their role in the decay and conversion of organic matter, and contamination with pathogenic microorganisms and their toxins(1-5).

The primary microflora in feedstuffs of vegetable origin is characterised by microorganisms living epiphytically and saprobially on the plants in the field (field microflora typical for the product). In contrast to this, the microflora of feedstuffs of animal origin mainly consists of contaminants originating from the sites of production and storage (secondary microflora, storage microflora). Feedstuffs can also be vectors of pathogenic microorganisms (bacteria like *E. coli*, *Salmonella*, *Clostridium perfringens*, viruses, fungi like *Stachybotrys atra*, *Aspergillus fumigatus*, parasites, etc.).

Considering the ecological and technological conditions by production of crop plants, as well as preservation and storage procedures, different types of feed contamination can be distinguished, e.g. primary microflora, typical for the product, secondary or storage microflora originated from the environment where a feedstuff was stored and a spoilage indicating microflora (spoilage causing microflora). Typical for unspoiled grains and their

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by-products as well as for compound feeds rich in grains are yellow pigmented colonies forming bacteria of the genera *Erwinia*, *Enterobacter* and *Pseudomonas* whereas grampositive species of the genera *Staphylococcus*, *Sarcina*, *Bacillus*, *Clostridium*, *Streptococcus* and *Enterococcus* are less common. Moulds of the genera *Aspergillus*,

*Penicillium*, *Mucor* and *Rhizopus* are found rarely compared to genera like *Alternaria*, *Verticillium*, *Helminthosporium*, *Cladosporium*, *Hormodendrum*, *Aureobasidium* and *Acremonium* as well as yeasts. Compound feeds consisting of unspoiled and technologically not much changed components reflect the microbiological spectra of their single components.

A spoilage indicating microflora is characterized by the predominance of bacteria and fungi of those genera which are less represented as long as the feed is unspoiled. These are representatives of the genera *Micrococcus*, *Staphylococcus*, *Bacillus* and *Streptomyces*. Spoilage by moulds during storage of a feed is normally initiated by osmotolerant yeasts or xerophilic moulds of the *Aspergillus*-*Glaucus* group. Only when the water activity is high enough the genera *Penicillium*, *Paecilomyces*, *Scopulariopsis*, *Trichoderma* and *Chaetomium* develop beside other *Aspergilli*. *Mucoraceae* have especially high moisture requirements. Favoured by humid weather or infested by pests, certain fungi (*Dematiaceae*, *Trichotheciae*, *Vericilliae*) can multiply rapidly and cause substantial changes in feeds in the field before harvest. *Fusarium* sp. hereby plays a special role. If the certain moisture content is exceeded it can multiply also after harvest. Many of these fungi are known as producers of mycotoxins.

In this work I will present results of bacteriological and mycological examination of feeds dealt in Slovenia in last five years and the legislation concerning this field in Slovenia

## MATERIAL AND METHODS

The examination of feed on pathogenic bacteria were done at the Institute for Microbiology and Parasitology at Veterinary faculty according to established methods. 2005 samples were inspected on *Salmonella*, 351 samples on *Clostridia*, and 55 samples on other pathogenic bacteria (*Streptococcus*, *Staphylococcus*, *E. coli*, *Listeria monocitogenes*). For the determination of mesophilic aerobic bacteria, moulds and yeasts content, the methods recommended by European Feed Microbiology Organisation (EFMO) were used. In the period from 1993 to 1997, 1591 samples of different feeds were examined on the content of mesophilic aerobic bacteria, 882 samples on the mould content and 706 samples on the yeasts content. The results (Table 1 and 2) were evaluated according to Slovene regulations (Pravilnik o zdravstveni ustreznosti krme, UL RS št. 20 z dne 12. 04. 1996), which are in accordance with EU regulations and recommendations.

*Table 1: The highest tolerable number of mesophilic aerobic microorganisms in feedstuffs in Slovenia*

Feedstuffs	Mesophilic aerobic bacteria million/g	Moulds 1000/g	Yeasts 1000/g
Feedstuffs of animal origin	2	10	50
Oil seed products and by-products	5	50	50
Cereal grains, cereal grain products and by-products	10	100	50
Complete feeding stuffs for young animals - meals	5	50	50
Complete feeding stuffs for adult animals - meals	5	100	50
Complete feeding stuffs - pellets	2	20	20



**Table 2:** The highest tolerable number of pathogenic bacteria in feedstuffs in Slovenia

Species of bacteria	Feedstuffs	The highest tolerable number
<i>Salmonella</i>	straight feeding stuffs, complete feeding stuffs and pet food	0 in 25g
<i>Enterobacteriaceae</i>	feedstuffs of animal origin and pet food in hermetically sealed containers	300 in 1g
<i>Clostridium</i> sp.	feedstuffs of animal origin, taken directly after heat treatment and pet food in hermetically sealed containers	0 in 1g
	other feedstuffs	100 in 1g

## RESULTS AND DISCUSSION

### The contamination of feedstuffs with bacteria

In last five years, at the Institute for Hygiene and Pathology of Animal Nutrition, 1591 samples of different feeds were investigated on the contamination with mesophilic aerobic bacteria, 2005 samples on *Salmonella*, 351 samples on *Clostridia*, and 55 samples on the other pathogenic bacteria (Table 3). 28 (1.8%) samples were not suitable because of too high contamination with mesophilic aerobic bacteria. 38 (1.9%) samples contained *Salmonella*, 11 (3.1%) samples contained *Clostridia*, and 3 samples (5.5%) contained other pathogenic bacteria. In most cases, the unsuitable feeds were of animal origin and unpelleted feedstuffs for young animals.

**Table 3:** The contamination of feedstuffs with bacteria in Slovenia in the period of 1993 to 1997

Feedstuffs	Mesophilic aerobic bacteria				<i>Salmonella</i>		<i>Clostridium</i> sp.		Other path. bact.	
	Number of examined samples	x 1000/g	Number of unsuitable samples	%	Number of examined samples	% of unsuitable samples	Number of examined samples	% of unsuitable samples	Number of examined samples	% of unsuitable samples
Feedstuffs of animal origin	157	1649	7	4.5	887	2.1	124	5.7	23	8.7
Oil seed products and by-products	67	764	2	3.0	53	1.9	10	0	7	0
Cereal grains, cereal grain products and by-products	319	3181	3	0.9	112	0.9	26	0	2	0
Complete feeding stuffs for young animals-meals	487	1529	9	1.9	434	1.6	77	2.6	14	4.2
Complete feeding stuffs for adult animals-meals	288	1247	5	1.7	391	1.8	63	1.6	5	0
Complete feeding stuffs - pellets	273	512	2	0.7	128	1.6	51	2.0	4	0
<b>TOGETHER</b>	<b>1591</b>	<b>1438</b>	<b>28</b>	<b>1.8</b>	<b>2005</b>	<b>1.9</b>	<b>351</b>	<b>3.1</b>	<b>55</b>	<b>5.5</b>

### The contamination of feedstuffs with moulds and yeasts

In Table 4 the results of the analysis of feedstuffs on moulds and yeasts are given. It is evident, that in the years 1993 to 1997, 2442 samples were inspected on moulds and 1947 samples on yeasts content. Most of them were cereals and cereal products. 201 (8.2%) samples were not hygienically suitable because of too high number of mould spores and 40 (2.1%) were not suitable because of too high number of yeast spores. In this respect, cereals, cereal products and unpelleted feedstuffs for young and adult animals were of the lowest hygienic quality. The reason was probably bad climatic conditions in the time of ripening and harvest of corn which was the main component of feedstuffs.

**Table 4:** The contamination of feedstuffs with moulds and yeasts in Slovenia in the period of 1993 to 1997

Feedstuffs	Moulds				Yeasts			
	Number of examined samples	$\bar{x}$ 1000/g	Number of unsuitable samples	%	Number of examined samples	$\bar{x}$ 1000/g	Number of unsuitable samples	%
Feedstuffs of animal origin	416	3.1	10	2.4	369	0.6	4	1.1
Oil seed products and by-products	97	7.6	4	4.1	51	0.4	2	3.9
Cereal grains, cereal grain products and by-products	624	19.8	87	13.9	446	5.7	9	2.0
Complete feeding stuffs for young animals - meals	498	11.5	49	9.8	367	9.3	11	3.0
Complete feeding stuffs for adult animals - meals	431	16.3	34	7.9	425	8.8	9	2.1
Complete feeding stuffs - pellets	376	4.6	17	4.5	289	0.9	5	1.7
<b>TOGETHER</b>	<b>2442</b>	<b>10.7</b>	<b>201</b>	<b>8.2</b>	<b>1947</b>	<b>4.3</b>	<b>40</b>	<b>2.1</b>

### The hygienic quality of feedstuffs for poultry

Among samples of feed for poultry (Table 5) 1,3% samples (out of 796 examined) contained too many mesophilic aerobic bacteria, 4,8% samples (out of 882 examined) contained too many mould spores and 1,1% samples (out of 706 examined) were too heavily contaminated with yeast.

In comparison to the early inspections, the hygienic quality of feeds and feedstuffs has been significantly improved. The reason is probably in using better straight feeding stuffs, specially corn, for feed mixtures production in our country and also in the recognition, that high quality feeds enables high production and appropriate animal health condition.

**Table 5:** The contamination of complete feeding stuffs for poultry with mesophilic aerobic bacteria, moulds and yeasts in Slovenia in the period 1993 - 1997

Feedstuffs	Mesophilic aerobic bacteria			Moulds			Yeasts		
	Number of examined samples	$\bar{x}$ 1000/g	% of unsuitable samples	Number of examined samples	$\bar{x}$ 1000/g	% of unsuitable samples	Number of examined samples	$\bar{x}$ 1000/g	% of unsuitable samples
Complete feeding stuffs for young animals - meals	259	1432	2.3	303	10.7	7.6	237	7.1	1.7
Complete feeding stuffs for young animals - pellets	221	617	0.5	264	3.9	3.4	251	0.8	0.4
Complete feeding stuffs for adult animals - meals	169	1376	1.2	198	15.3	3.5	143	8.6	1.4
Complete feeding stuffs for adult animals - pellets	147	719	0.7	117	6.2	2.7	75	1.0	1.3
<b>TOGETHER</b>	<b>796</b>	<b>1355</b>	<b>1.3</b>	<b>882</b>	<b>11.9</b>	<b>4.8</b>	<b>706</b>	<b>5.4</b>	<b>1.1</b>



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## КОНТАМИНАЦИЈА НА АНИМАЛНИТЕ ХРАНИВА СО ГАБИ И БАКТЕРИИ ВО СЛОВЕНИЈА

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Во периодот од 1993-1997 ја испитувавме контаминацијата на хранива за салмонела клостридии и други патогени бактерии, мезофилни аеробни бактерии, мувли и квасници. Испитани се 2442 мостри од кои 8,2% беа негодни за испитување поради висока контаминација со габи, 2,1% мостри (од 1947 испитани) имаа премногу спори на квасници и 1,8% мостри (од 1591 испитани) беа негодни за испитување поради високата контаминација со мезофилни аерофилни бактерии. Салмонела беше најдена во 1,9% мостри (од 2005 испитани), 3,1% мостри (од 351 испитани) имаа премногу клостридии и 5,5% мостри (од 55 испитани) содржеа други патогени бактерии.

Од 796 мостри на комплетни крмни смеси за живина испитани на мезофилни аеробни бактерии, 10 (1,3%) имаа многу бактерии, 42 (4,8%) мостри (од 882 испитани) беа негодни поради висока контаминација со габи и 8 (1,1%) мостри (од 706 испитани) беа негодни поради висока контаминација со квасници. Од аспект на микробиолошката исправност, пелетираната храна за живина е поквалитетна од непелетираната.