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INVESTIGATION OF THE TOXICOLOGICAL PROPERTIES OF A BIOMATERIAL DERIVED FROM COTTONSEED MEAL

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Annotation

This article discusses the production of a protein-rich bioproduct through microbiological treatment of cottonseed meal and the development of enriched compound feed formulations based on this bioproduct. The study examines the technological and physicochemical properties of the bioproduct, including its protein and gossypol content. According to the results, the protein content of the bioproduct reached 42%, while the gossypol content was 0.001%. In addition, the sanitary and bacteriological characteristics and heavy metal concentrations of the developed compound feed samples were analyzed. The findings demonstrate the potential for producing nutritionally enriched compound feeds using locally available raw materials.

Keywords: cottonseed meal, bioproduct, compound feed, toxicological safety.

How to Cite

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The Resolution No. PQ-120 of the President of the Republic of Uzbekistan, dated February 8, 2022, entitled “On Approval of the Program for the Development of the Livestock Sector and Its Sub-Sectors in the Republic of Uzbekistan for 2022–2026,” and Resolution No. 312 of the Cabinet of Ministers, dated April 27, 2018, entitled “On Measures to Organize Modern Industrial Production of Compound Feed in the Republic,” establish several priority objectives. These include the development of the livestock, poultry, and fishery sectors; the expansion of forage crop varieties; the formulation of scientifically based compound feed recipes; the development of technologies for producing locally sourced, protein-rich, and nutritionally balanced feed; the expansion of production capacities for feed required in intensive aquaculture; and the implementation of scientifically grounded feeding practices for livestock [1,2].

The advancement of scientific research in the livestock sector, the intensification of applied and innovative studies, the rational utilization of local raw materials, and the application of biotechnological methods to enhance the nutritional composition of compound feeds represent some of the most pressing challenges today [3].

In view of these considerations, significant efforts are being made in the Republic of Uzbekistan to ensure that the population has access to high-quality meat and dairy products. Research findings have demonstrated that deficiencies in certain vitamins and feed additives can lead to the development of various diseases in animals [4].

Globally, the permissible concentrations of toxic substances in compound feeds vary among countries. Each country establishes its own safety limits for heavy metals, mycotoxins, and pesticides in

compound feed. The following table presents the regulatory requirements for toxic substances in compound feeds in selected countries [5].

Table 1. Toxicological Requirements for Compound Feeds in Different Countries

№	**Name of Hazardous Substance**	South Korea	Japan	Canada	European Union	United States	Uzbekistan
1	Lead, mg/kg	10	3	5,0	5,0	5,0	10
2	Fluorine, mg/kg	250	50	-	-	-	-
3	Mercury, mg/kg	0,8	0,4	0,4	0,4	1,0	0,05
4	Cadmium, mg/kg	2	1	0,05	1,0	0,05	0,1
5	Arsenic, mg/kg	10	2	5,2	2,0	2,0	0,01
6	Chromium, mg/kg	100	-	-	-	-	-
7	Aflatoxin, mg/kg	10	20	20	20,0	20,0	20
8	Ochratoxin A, mg/kg	200	100	200	100,0	100,0	100,0
9	DDT, mg/kg	0,5	0,1	0,2	0,20	0,20	0,10
10	Carbofuran, mg/kg	0,2	0,1	-	-	-	-
11	Fipronil, mg/kg	0,02	0,01	-	0,01	0,01	-
12	Heptachlor, mg/kg	25	20	25	20,0	20,0	20
13	Lindane, µg/kg	100	50	100	50,0	50,0	50,0
14	Melanin, µg/kg	3,0	2,5	2,5	2,50	2,50	2,50
15	Aldrin, µg/kg	25,0	-	20	10	20,0	-

As shown in Table 1, the regulatory requirements for heavy metals, mycotoxins, and pesticide residues in compound feeds vary among countries [7].

The maximum permissible concentration of lead in compound feed is 10 mg/kg in South Korea, whereas the lowest limit is 3 mg/kg in Japan. The permissible cadmium concentration is 1 mg/kg in Japan, compared with 0.05 mg/kg in Canada and the United States. The maximum permissible arsenic concentration is 10 mg/kg in South Korea, 5.2 mg/kg in Canada, and 2 mg/kg in the remaining countries.

The presence of heavy metals, aflatoxins, and pesticide residues in products can adversely affect living organisms. As indicated in Table 1, all the countries considered have established regulatory limits for lead (Pb), mercury (Hg), cadmium (Cd), and arsenic (As), as well as for mycotoxins and certain pesticides. Macro- and microelements present in compound feeds play important roles in hormonal activity, the regulation of animal physiological functions, and other essential biological processes [6].

Strict requirements for mycotoxin levels are broadly comparable across the countries considered, with South Korea having the lowest permissible aflatoxin concentration. As shown in the table, the permissible levels of pesticide residues in compound feeds are also reported to be similar across the countries examined [8].

Information on the effects of toxic substances on animal organisms is presented in the following table.

Table 2. Adverse Effects of Hazardous Substances

№	Name of Hazardous Substance	Adverse Effects	Source
Group I. Effects of Heavy Metal Residues on Animal Organisms			
1.	Lead	Causes kidney damage, bone degeneration and fragility, impaired liver function, infertility, and reproductive system dysfunction.	[12; 14.]
2.	Mercury	Adversely affects brain and kidney function, causing necrosis and demethylation in proximal tubular cells.	[18.]
3.	Arsenic	Causes damage to the lungs and gastrointestinal tract.	[11.]
4.	Cadmium	Causes histopathological damage and impairs the locomotor system.	[9;23.]
Group II. Effects of Mycotoxins on Animal Organisms			
5.	Aflatoxin B	Causes growth depression and disrupts protein synthesis.	[5.]
6.	Ochratoxin A	Damages the small intestine, bone marrow, lymph nodes, and intestinal mucosa.	[12;13.]
Group III. Effects of Pesticides on Animal Organisms			
7.	DDT	Causes persistent stress in poultry.	[9;11.]
8.	Heptachlor	Causes renal failure and urinary disorders.	[12.]
9.	Melalin	Causes diarrhea, polyuria, and proteinuria.	[14;15.]

The toxic elements and residues listed in the preceding table can cause substantial economic losses in the livestock and poultry industries. This is because the simultaneous provision of contaminated compound feed to animals, particularly when administered according to established feeding rations, may lead to severe animal mortality. Furthermore, the accumulation of these toxic substances in meat, meat products, and eggs may pose a risk to human health, as the consumption of contaminated animal-derived products can result in human poisoning [9].

In this study, cottonseed meal was subjected to microbiological treatment to obtain a biomaterial, taking into account several key factors, including the reduction of gossypol content, improvement of dietary fiber digestibility and absorption, and enhancement of protein content. The research was aimed at developing new compound feed formulations based on the resulting biomaterial [10].

The following table presents the technological parameters of biomaterial production through the microbiological treatment of cottonseed meal, together with the physicochemical characteristics of the resulting biomaterial.

Table 3. Technological and Physicochemical Characteristics of Biomaterial Production from Cottonseed Meal

№	Parameter	Unit of Measurement	Value

Amount of Raw Material Used for Biomaterial Production			
1. 2. 3. 4.	Cottonseed meal Water Microorganism Carbohydrate (nutrient medium)	gr ml gr gr	1000 300-400 3-4 100-150
Technological and Physicochemical Parameters			
5. 6. 7. 8. 9. 10.	Temperature Maturation Period Moisture Content Protein Content Gossypol Content Proportion of Water-Soluble Proteins	^{0}C hour % % % %	30-35 20-24 60-65 42 0,001 62,4

The experimental results confirmed that, based on the recommended optimal formulation, the protein content of the 1,000 g biomaterial reached 42%, while the proportion of water-soluble protein was 62.4%. The results also demonstrated that the moisture content and maturation period met the established requirements.

Based on the proposed biomaterial formulation, the research led to the development of new formulations of enriched compound feeds using conventional raw materials and incorporating the obtained biomaterial. The composition of the enriched compound feeds produced using the biomaterial is presented in the following table.

Table 4. Compound Feed Samples Used in the Experiment

№	Product Name	Compound Feed Samples			
		1	2	3	4
1	Wheat bran	20	30	40	50
2	Cottonseed meal	50	35	25	20
3	Corn	15	15	15	15
4	Cottonseed hulls	10	10	10	10
5	Biomaterial	5	10	10	5
	Total	100	100	100	100

As shown in the table, wheat bran accounted for 20% of Sample 1, whereas cottonseed meal constituted 50%. In Sample 2, the proportion of cottonseed meal was reduced to 35%, while the proportion of the biomaterial obtained through biotechnological treatment increased to 10%. The wheat bran content

also increased by 10 percentage points compared with Sample 1. During the experiment, the proportion of wheat bran was varied from 20% to 50%, while the proportion of cottonseed meal was correspondingly reduced from 50% to 20%. At the same time, the proportion of the biomaterial obtained through biotechnological treatment was increased from 5% to 10%.

The developed compound feed formulations were analyzed at the State Center for Animal Disease Diagnostics and Food Safety of Fergana Region. The analyses included sanitary and bacteriological and microbiological assessments, determination of heavy metal concentrations in the collected product samples, and measurement of residual organochlorine pesticide levels. The following table presents the characteristics of compound feed samples prepared with the addition of 10% and 20% biomaterial.

Table 5. Sanitary, bacteriological, and microbiological characteristics of compound feed samples and their heavy metal and organochlorine pesticide residue levels

№	Parameter	Regulatory Limit (mg/kg)	Sample 1 (10%)	**Sample 2 (20%)**
Sanitary and bacteriological parameters				
1	Sanitary and Bacteriological Parameters		Compliant	Compliant
2	Total viable microbial count (MAFAM), in 1.0 g		Compliant	Compliant
3	Coliform bacteria (BGKP)		Compliant	Compliant
4	Salmonella		Compliant	Compliant
5	Staphylococcus aureus, in 1.0 g		Compliant	Compliant
6	Moulds, in 1.0 g		Compliant	Compliant
Microbiological Parameters				
7	Microbiological Parameters	10^7	10^3	10^5
8	Streptococcus thermophilus and Lactobacillus delbrueckii subsp. bulgaricus	10^7	10^3	10^5
9	Bifidobacterium	10^7	10^3	10^5
Heavy Metal Content (mg/kg)				
№	Heavy Metal	GOST 33824-2016	Sample 1 (10%)	Sample 2 (20%)
10	Heavy Metal	5,0	0,083	0,069
11	Lead (Pb)	1,0	0,05	0,05
12	Cadmium (Cd)	0,5	0,04	0,04
13	Arsenic (As)	0,1	0,0050	0,005
Residual Organochlorine Pesticide Content (mg/kg)				

15	Organochlorine Pesticide Residues	0,5 mg/kg		
16	α-HCH	0,5 mg/kg		
17	γ-HCH	0,01 mg/kg		
18	DDE	0,01 mg/kg		
19	DDD	0,01 mg/kg		

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