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Synthesis and study of arginine-containing manganese and zinc chelates

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Abstract

Synthesis conditions have been established and arginine-containing manganese and zinc chelate compounds have been synthesized with general formula: Me (Arg)n$(CH_3COO)_2 \cdot mH_2O$, where Me=Mn, Zn; Arg- neutral molecule of arginine; n=1÷3; m=1÷4. Synthesized chelates have been studied using a number of physical-chemical researches. In particular, composition has been identified via trace element analysis, while the individuality has been established through fusion temperature measurement and diffractometric method. According to qualitative solubility research, chelates are highly soluble in water, but poorly soluble in organic solvents. Dissociation constant for chelate compound-containing solutions has been calculated using conductometric study method.

With the purpose of biological activity research, on the basis of preliminary trial tests an opinion is expressed that entry of arginine-containing manganese and zinc chelates into composition of broilers' combined feed premixes has had a positive effect on birds' live weight gain and their preservation. There is expressed an opinion on the reasonability of main tests conduction in order to identify the optimal doses of arginine-containing manganese and zinc chelate compounds for broilers.

Keywords: Manganese; Zinc; Chelate; Arginine; Egg-laying bird; Combined feed

1. Introduction

Receipt of ecologically safe agricultural production (poultry and animals' meat) with high qualitative and quantitative indicators is one of the most relevant problems today. Provision of their feed with a definite composition, amount and optimal ratio of essential microelements is an indispensable condition of this problem solution. Microelements can be added into poultry premixes composition in two:inorganic or chelate forms. Salts being in inorganic form are characterized by high toxicity that is caused by formation of poorly soluble and not easily assimilable substances in gastrointestinal tract of animals and birds. On the contrary, microelements being in chelate form have virtually very low toxicity, high digestibility and increased degree of efficiency when used in small doses that in its turn will cause ecological safety of microelements use in this form. Worth noticing that in 2003 the European countries have adopted the legislative act on as low as possible permissible doses of microelements in poultry and animal manure. As we have already mentioned, this problem can be solved through the use of chelate forms of microelements only. This fact is confirmed by the results of researches carried out for years by both foreign scientists [1-16] and our team [17-27]. Exactly these advantages explain the scaling-up of production of premixes containing microelements in chelate form compared to non-chelate ones, and this growth has an increasing character. For a number of reasons, amino acids have been used for receipt of chelate-form microelements from organic substances. Among amino acids arginine has been selected. It is a basic amino acid, in living organisms it converts into nitrogen oxide, which promotes blood vascular system elasticity that is so relevant during angina attacks, substantially increases blood supply to the brain and heart

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muscles, has a positive effect on genital system. Scientific studies have confirmed a beneficial effect of nitrogen oxide on arterial blood pressure control, immunity and central nervous system activity [28-29].

Among microelements, manganese and zinc are selected as a research subject. Manganese is among the most important biometals, enters into composition of enzymes, which perform crucial functions in the organism: participate in the neuromediators synthesis and exchange in the nervous system, secure normal operation of muscle tissue, take part in thyroid gland hormone (thyroxine) exchange processes, increase hypoglycemic effect of insulin, promotes the development of connective tissue, joints and bones etc.

Zinc is a cofactor of a large group of enzymes, it is necessary for normal progress of many biochemical processes. It participates in collagen synthesis, cell differentiation, pancreatic glands insulin functioning; performs the important role in skin regeneration processes, nail and hair growth processes, promotes vitamin E absorption and its normal blood concentration maintaining. Based on the above mentioned, the goal of our research is a synthesis of arginine-containing manganese and zinc chelate compounds, their physical-chemical research and study of their biological active.

2. Computational Method

- Trace element analysis– for determination of composition of arginine chelate compounds.
- Fusion temperature determination – for establishment of individuality of chelates.
- Solubility – for study of qualitative solubility of chelate compounds in different solvents.
- Conductometric study – for determination of dissociation constant of chelate compound- containing solvents.
- Weighing method – for determination of poultry weight gain.
- Counting technique – for establishment of poultry preservation.

3. Results and discussion

With the purpose of synthesis of arginine-containing manganese and zinc chelates there is studied a system with general formula:

$$Me(Arg)n(CH_3COO)_2 \cdot mH_2O$$

Where,

Me=Mn, Zn;
Arg –neutral molecule of arginine;
n=1÷3;
m=1÷4.

Metal acetate and arginine mixtures in1:1; 1:2 and 1:3 molar ratio were prepared for synthesis, mixtures were dissolved in minimal volume of water under conditions of heating and intensive stirring. True solutions are hold above water vapor bath with the purpose of concentration, obtained compounds are treated several times with a water aimed to removal of acetic acid residues. Finally, the compounds are washed with a small amount of water, ether and are dried at a room temperature. The individuality of synthesized chelates is established via fusion temperature measurement, at the fusion determining device SMP10. The qualitative solubility of compounds in different solvents is determined as well, according to which they are characterized by high solubility in water and dimethyl formamide, and by relatively poor solubility in alcohol and acetone (Table 1).

In order to determine the dissociation constant for arginine-containing chelate compounds, a conductometric study is carried out at the pH and Conductivity Sensor LE703 device. For these purposes, there were prepared solutions with concentrations within the limits of 0.025N-0.0006503N. Experiment was conducted in the thermostat at 25°C. Results of experimentsaregiven in Table 1.R^2– regression assessment indicator, which shows how close are the experimental data with the respective function of the diagram, is quite high and varies within the limits of 0,9868-0,8337. Dissociation constants of initial salts, in particular $K_{[Mn(CH3COO)2\cdot4H2O}$]=0.07581, and $K_{\{Zn(CH3COO)2\cdot2H2O]}$= 0.02581are measured under the same conditions.

As is seen from the Table, dissociation constants of chelates and their initial salts are of the same order (in the same limits). Proceeding from this fact we can assume that under conducted experiment conditions, chelate dissociation proceeds with detachment of the second (external) sphere (acetate-ions), and dissociation has no place in the inner

sphere. This fact can be explained by formation of the stable heterocyclic five-membered cycles between metal and organic ligand, with Me-Arg bond strength.

Table 1 Some physical characteristics of chelate compounds

SN	The formula of the compound	Molar mass g/mol.	Sliding t⁰c	Solubility				Conductometric survey results	
				water	Alcohol	Acetone	Dmfa *	R^2	K_D
1	$[Mn(Arg)](CH_3COO)_2 \cdot 4H_2O$	419.48	144	+	-	-	-	0.9330	0.05766
2	$[Mn(Arg)_2](CH_3COO)_2 \cdot 4H_2O$	593.96	136	+	-	-	-	0.9359	0.04963
3	$[Mn(Arg)_3](CH_3COO)_2 \cdot 4H_2O$	767.54	123	+	-	-	-	0.9598	0.06290
4	$[Zn(Arg)](CH_3COO)_2 \cdot 4H_2O$	429.45	58	+	+	Small soluble	+t	0.8860	0.03793
5	$[Zn(Arg)_2](CH_3COO)_2 \cdot 4H_2O$	603.93	114	+	Small soluble	-	Small soluble	0.8337	0.06156
6	$[Zn(Arg)_3](CH_3COO)_2 \cdot 4H_2O$	778.13	63	+	+t	-	Small soluble	0.9868	0.03310

DMFA * - Dimethylformamide; +: soluble; -: Insoluble

Theindividuality of compounds: $[Mn(Arg)](CH_3COO)_2 \cdot 4H_2O$(1), $[Mn(Arg)_2](CH_3COO)_2 \cdot 4H_2O$(2), $[Zn(Arg)](CH_3COO)_2 \cdot 4H_2O$(3),$[Zn(Arg)2](CH_3COO)_2 \cdot 4H_2O$(4) is established using diffractometric method. X-Ray-diffractometric study was conducted on $Cu_{k\alpha}$(λ=0.154184 nm) radiation using ДРОН-4.07. During exposition, samples were rotated around an own plane using special device – ГП-13. In order to compare, the diffractograms of the initial compounds are taken, as well. Based on the analysis of diffractograms, one may say that location and intensities of diffraction maximums of the 1, 2, 4 chelate compounds differ from both each other and from location and intensities of diffraction maximums of initial compounds (acetates and arginine), and each chelate compound is characterized by its distinctive maximums and intensities (Fig. 1, 2, 4). As for zinc chelate compound (3), as is seen from diffractogram, it is an amorphous solid substance, while the diffractograms of initial compounds (zinc acetate and arginine) are characterized by clearly expressed peaks (Fig. 3).

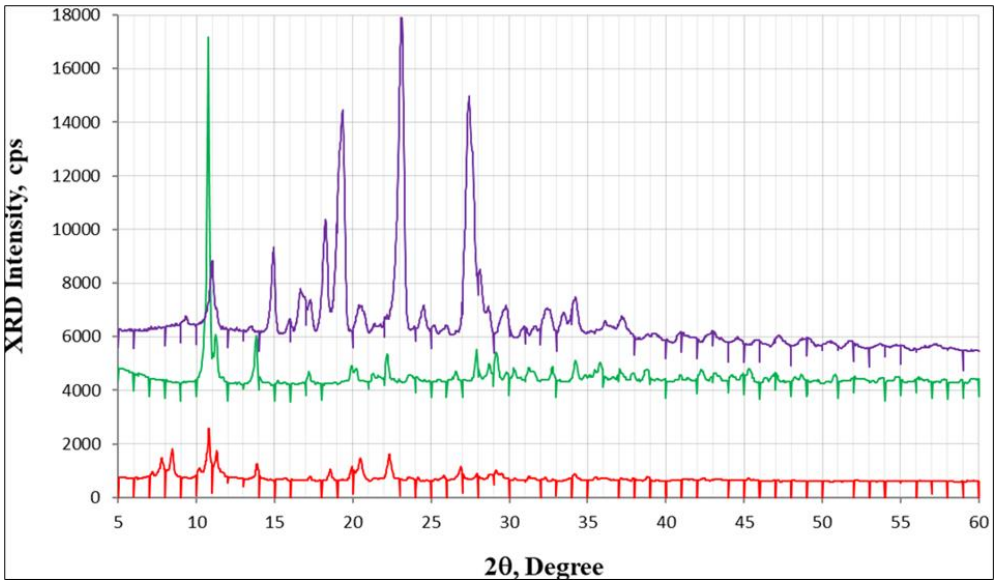

Figure 1 ■■ $[Mn(Arg)](CH_3COO)_2 \cdot 4H_2O$, ■■$Mn(CH_3COO)_2 \cdot 4H_2O$, ■■Arg

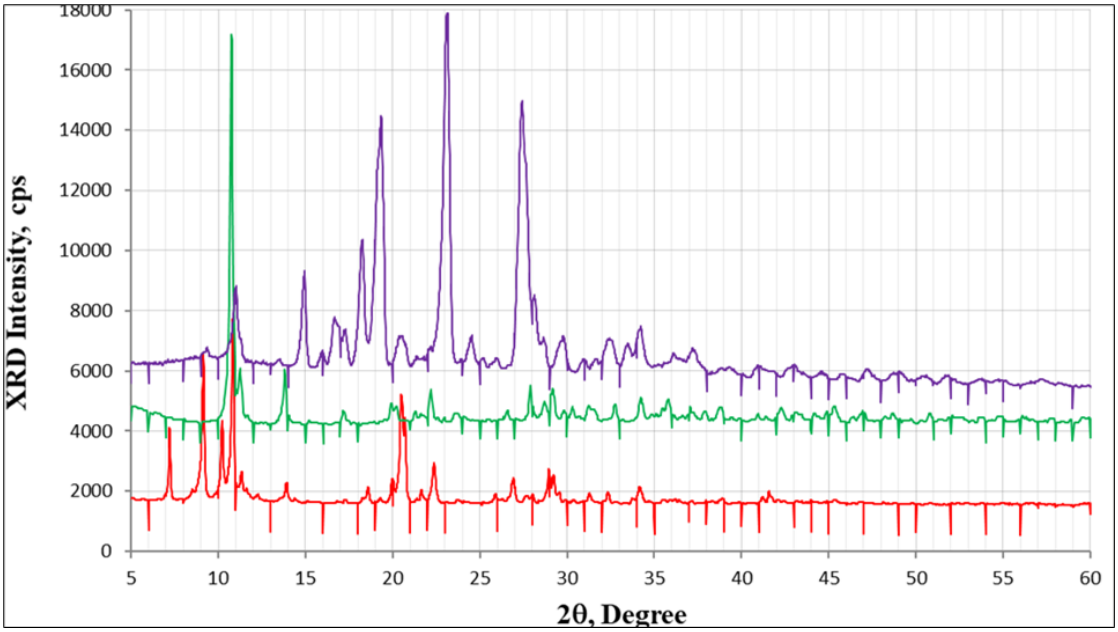

Figure 2 ■■[Mn(Arg)$_2$](CH$_3$COO)$_2$·4H$_2$O, ■■Mn(CH$_3$COO)$_2$ ·4H$_2$O,■■Arg

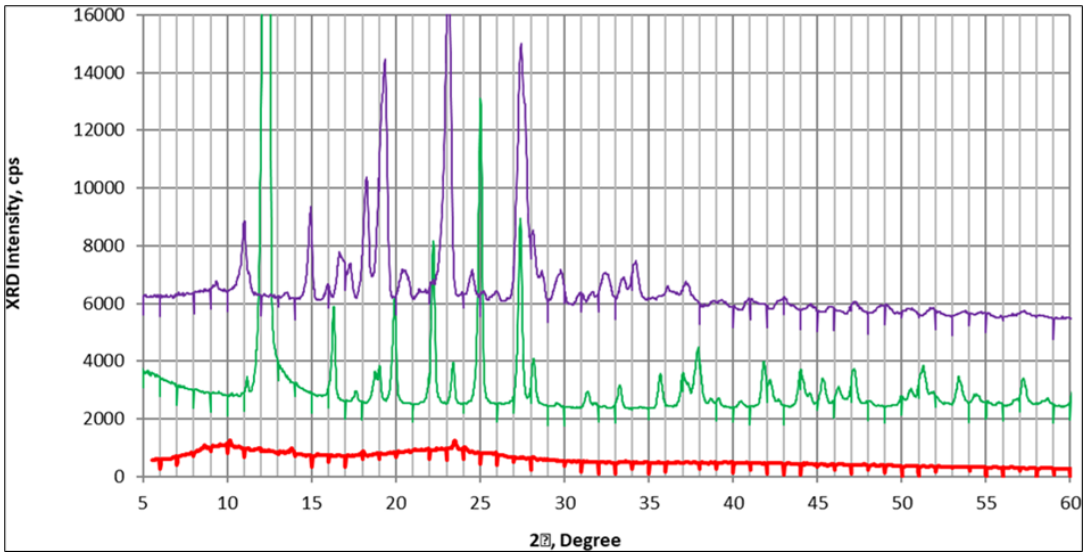

Figure 3■■ [Zn(Arg)](CH$_3$COO)$_2$·4H$_2$O, ■■Zn(CH$_3$COO)$_2$·2H$_2$O,■■Arg

Table 2 Composition of the mixture

Z Mixture	Znorm(g)	Zmax(g)
[Mn(Arg)](CH$_3$COO)$_2$·4H$_2$O	13.24	15.23
[Mn (Arg)$_2$](CH$_3$COO)$_2$·4H$_2$O	9.27	10.66
[Zn(Arg)](CH$_3$COO)$_2$·4H$_2$O	14.50	17.14
[Zn(Arg)$_2$](CH$_3$COO)$_2$·4H$_2$O	10.72	12.33

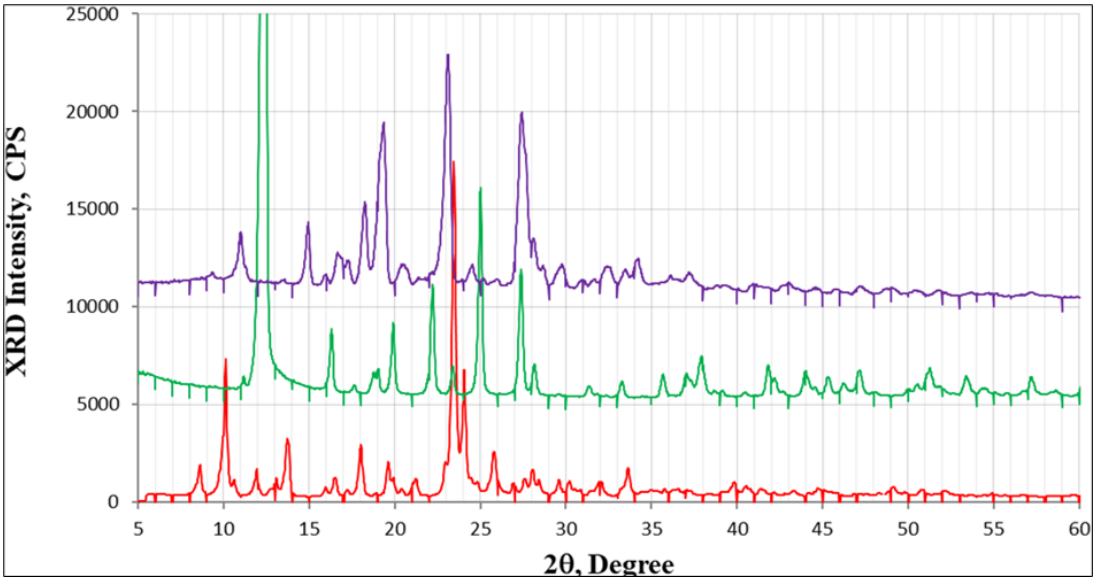

Figure 4 ■■[Zn(Arg)$_2$](CH$_3$COO)$_2$·4H$_2$O,■■ Zn(CH$_3$COO)$_2$·2H$_2$O, ■■Arg

Thus, based on the analysis of conducted diffractometric studies one can conclude that obtained chelate compounds are individual substances.

Trial tests on broilers have been carried out with the purpose of study of synthesized compounds biological activity. Chelate mixtures for 100 kg combined feed premixes intended for broilers have been prepared (Table 2).

Twenty wings eachfor two pilot and one control group have been selected for a test according to zootechnic analogues principle. Experiment has lasted for one month. In the test period we have studied poultry live weight in the beginning and in the end of experiment. Results are given in Table3.

Table 3 Live weight change during the test period

Group	Live mass change (g.)					
	The beginning of the experiment			End of trial		
	Mini	Max	M+m	Mini	Max	M+m
Control	1510	1660	1585±75	1510	1670	1590±80
Znorm	1500	1650	1580±68	1520	1660	1600±76
Zmax	1500	1670	1580±65	1530	1690	1600±68

Poultry preservation is studied as well (Table 4), according to which the maximum preservation is registered in the test group.

Table 4 Keeping the bird during the trial period, %

Group	Number of birds, (wing)		Maintenance %
	The beginning of the experiment	End of trial	
control	20	18	90
Znorm	20	20	100
Zmax	20	20	100

Based on preliminary trial tests we can assume that entry of arginine-containing manganese and zinc chelates into broilers' combined feed premixes composition has had a positive effect on live weight gain and poultry preservation.

4. Conclusion

On the basis of conducted researches, the following conclusions can be drawn:

- Synthesis conditions have been established and arginine-containing manganese and zinc chelate compoundshave been synthesized.
- Chelates are individual compounds characterized by high solubility in water and poor solubility in organic solvents.
- Dissociation constants of compounds have been calculated using conductometric study method.
- Based on preliminary trial tests conducted with the purpose of biological activity study one can assume that entry of arginine-containing manganese and zinc chelates into composition of broiler's combined feed premixes composition has had a positive effect on broilers' live weight gain and poultry preservation.
- There has been expressed an opinion on the reasonability of main tests conduction in order to determine optimal doses of arginine-containing manganese and zinc chelates for broilers.

Compliance with ethical standards

Acknowledgments

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Disclosure of conflict of interest

No conflict of interest.

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