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# INFLUENCE OF THE IRON METHIONATE AND FERROUS SULPHATE ON SOME PRODUCTIVE INDICES IN BROILER CHICKENS

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## **Abstract**

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Comparative study of iron methionate (Fe-methionate) and iron sulphate (FeSO<sub>4</sub>, Merck) in broiler chickens was conducted. The aim of the study was to establish the effect of two levels of iron methionate and iron sulphate (40 ppm and 300 ppm) on some productive indicators and iron depositing in liver. The rheological studies conducted with FeSO<sub>4</sub> (heptahydrate) and iron methionate were aimed to explore the opportunities to mix these products with the feed. The iron content in both products were analysed using atomic absorption spectroscopy (AAS), which showed that the iron methionate contained 13.3% iron and the ferrous sulphate contained 20% iron. We found that the rheological parameters (angle of repose, flow rate, Hausners ratio) was better in ferrous methionate compared to that of the ferrous sulphate (heptahydrate), which was a prerequisite for better mixing with the feed.

In terms of the chicken weight and feed convention at a basic iron in the mixtures of 119.5 ppm and additive of 40 ppm and 300 ppm ferrous methionate or ferrous sulphate, respectively the trends were in favour of the organic product, however the differences were not statistically significant. The ferrous methionate deposits in the chicken's liver were higher compared to that of the ferrous sulphate (p < 0.05).

*Key words:* ferrous methionate, ferrous sulphate, broiler chickens, liver deposit, productive indices, rheological parameters

## Introduction

The trace element iron is of great importance to fast growing animals, such as the broiler chickens. The sources of protein in the compound feed of the broiler chickens contain high levels of iron, which prevent them from iron deficiency anaemia (Marinov, 2011). It has been scientifically proven that the chickens need for iron varies from 75 to 80 ppm/kg of feed (Davis et al., 1968). Other researchers (Vahl and Klooster, 1987) recommend that the iron content in the compound feed of the broiler chickens should be 100 ppm. The

American standards (NRC, 1994) recommend that the iron content in the compound feed should be 80 ppm throughout the fattening period and that the dose of the ferrous inorganic salts should be 25-40 ppm (Surdzhiyska, 1996; Lyons, 2006). The increased iron content in the oral administration reduces its reabsorption (Edwards and Washburn, 1968) and the iron deposits mainly in the liver, followed by the spleen, chest and thigh muscles (Seo et al., 2008).

A number of inorganic iron compounds (ferrous sulphate, ferric oxide, etc.) which have positive effect on the health and growth of the broiler chickens are used in the feed production.

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However, the low level of absorption, poor bioavailability, low adaptability in the homogenisation with the compound feed and the incompatibility with the vitamin B group have been clarified. This requires that organic iron compounds, which may replace the inorganic salts, should be searched (Cao et al., 1996; Shinde et al., 2011).

The aim of this study was to compare the influence of the ferrous methionate and ferrous sulphate (heptahydrate) on some production traits and the deposit of iron in the chicken's livers.

#### **Materials and Methods**

Ferrous methionate synthesized from Department of Organic Chemistry, University of Chemical Technology and Metallurgy – Sofia, which contained 13.3% iron and 34% methionine, was studied. The product was analysed using atomic absorption and so its molecular weight was determined. It was compared to ferrous sulphate (FeSO $_4$ 7H $_2$ O) produced by the company Merck, which contained 20% iron.

Rheological studies were conducted with the following products: ferrous sulphate (FeSO<sub>4)</sub> and ferrous methionate (FeMet). The aim was to explore the opportunities to mix these products with the feed. The methods used were in compliance with the European Pharmacopoeia 7 (2.9.34. Bulk Density and Tapped Density of Powders and 2.9.36. Powders Flow). A test involving 55 broiler chickens of both genders (4-linear hybrid ROSS-IKOV) was conducted. The chickens were divided into five equal groups: 4 test groups and a control one. In the beginning of the test the chickens were two weeks old. The groups of birds were placed in cages equipped with galvanized mesh and automatic feeders and drinkers. The premise had adequate lighting, ventilation and options to regulate the temperature. The plan of experimental study is shown in Table 1. It shows that the chickens received the same compound feed throughout the growing period, as it differed only in the dose and form of the iron (as described above) in the various groups.

The compound feed was produced from our own recipe (Table 2) and with the of trace element premix (prepared

Table 1 Scheme of the experiment

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Groups	Scheme
I	Compound feed + Fe-methionate (40 ppm)
II	Compound feed + Fe-methionate (300 ppm)
III	Compound feed + FeSO4 (40 ppm)
IV	Compound feed + FeSO4 (300 ppm)
V (Control)	Compound feed without added Fe+

from our recipe without involving iron). The composition of the compound feed did not include undesirable substances. In the beginning of the experiment the chickens were divided into groups using the method of analogues upon initial live weight of 0.183 kg. Then samples of the compound feed used in all groups were taken and analysed for presence of Fe<sup>+</sup> using atomic absorption spectrophotometer (Spectra AA 800, Varian, Australia). The chickens were allowed to eat in groups without limitation. During the study the weight was controlled every 15 days and the food consumption was controlled on a daily basis.

On the 10<sup>th</sup> and 25<sup>th</sup> day the chickens of all groups were treated prophylacticaly with Bioseled® (Biovet, Peshtera, Bulgaria; with composition in 1 ml: Na-selenite 0.6 mg, Vitamin E 25.0 mg) through the water. The treatment continued two consecutive days and the dose taken was 0.2

Table 2
Composition of the compound feed for broiler chickens used in the study

Components	%
Corn	42.288
Wheat	15
Soybean meal (46% crude protein)	30
Sunpro (45% crude protein)	3
Sunflower oil	3
Endox (0.0125%)	0.0125
Limestone (37% Ca)	0.9
Dicalcium phosphate	1.8
Lysine 98%	0.2
Salt	0.25
Sodium bicarbonate	0.25
Coxidin	0.05
Trace element premix*	0.05
Vitamin premix	0.2
Nutritional value	
Crude protein, g/kg	213.31
Crude fat, g/kg	54.89
Crude fibre, g/kg	41.08
Crude ash, g/kg	61.43
Ca, g/kg	8.9
P, g/kg	7.54 (a date string represented with only 2 digits for the year)
Moisture g/kg	110.38
Na+CI, g/kg	4.08
<i>Metabolizable energy</i> – broiler chickens, kcal/kg	2717.2

<sup>\*</sup> Trace element mixture does not contain iron

ml/l of drinking water. The iron content in the basic feed was  $119 \pm 5$  ppm. During the study the health status of the chickens of all groups was good. There were no clinical symptoms or mortality.

At the end of the study (which continued 35 days) 5 chickens with an average weight for the group were selected. They were subjected to clinical analysis, which included: Body weight before slaughter, Weight of carcass (without giblets) and Weight of the liver. The samples of the liver were stored at a temperature of -18°C and then were thawed out and dried in a drying scale equipped with infrared heater. They were analysed with atomic absorption spectrophotometers Spectra AA 800 (Varian, Australia) and Spectra AA 220 Z (Varian, Australia) equipped with graphic cuvettes Varian.

Based on the controlled parameters the daily average growth, food and energy consumption and the Food consumption per kg/Gain (kg) were calculated. The results were processed using variation and statistical methods; the reliability of the differences was determined based on the Student-Fisher tables. In the calculation of the body weight the non-parametric method Mann-Whitney U test was used.

# **Results and Discussion**

The rheological characteristics of the comparable iron compounds are presented in Table 3. It shows that some of the main parameters (angle of rest, flow rate, Hausners ratio) in the organic products are similar, while the flow rate of the ferrous methionate is the most favourable. The ferrous sulphate does not flow freely and the indicators responsible for such behaviour (angle of repose and flow rate) can not be determined directly. These results are probably due to the high value of the indicator loss on drying (37°C).

The positive rheological parameters of the powder mixtures could ensure their even distribution when mixed with animal feed. There is a relationship of the rheological studies with the results of the published data on the pharmacokinetics and utilisation of the ferrous methionate compared to the ferrous sulphate (Arnaudova-Matey, 2013) and with the results of the iron deposit (Table 7).

In terms of feed consumption (Table 4) it may be concluded that the control group had the highest consumption and the lowest consumption was reported in the group, which received ferrous methionate in a dose of 40 ppm. The concentration of the iron compounds has various effects on the consumption. The differences in the feed consumption of the groups are insignificant. The results of the feed consumption and utilisation that we have obtained are similar to those obtained by other authors (Vahl et al., 1987; Cao et al., 1996). The results for the daily average consumption of feed from chickens were analog of the total consumption. The minor variations in the feed consumption of the test groups of chickens have led to minor variations in energy. The energy consumed per chicken during the treated groups varies within the range of 326-348 kcal. The accepted energy corresponds to the age of the chickens (Surdzhiyska, 1996). The iron products not influenced on the accepted energy.

Table 3
Comparable rheological characteristics of iron methionate and ferrous sulphate

Parameter	Ferrous sulphate	Ferrous methionate
Weight, g	50	50
Saturated volume, cm <sup>2</sup> /g	53.7	60.2
Saturated volume after jarring, cm <sup>2</sup> /g	50	52.6
Bulk density, g/cm <sup>2</sup>	0.93	0.83
Bulk density after jarring, g/cm <sup>2</sup>	1	0.95
Angle of repose, °	Not flow	30.65
Flow rate, g/cm <sup>2</sup> .s	Not flow	2.25
Hausner ratio	1.07	1.14

Table 4 Comparative data about fodder consumption per kilogram of gain in broiler chikens continuously fed with a compound fodder containing ferrous methionate (FeMet) or ferrous sulphate (FeSO $_4$ )

Groups	Food intake per chicken in the period, kg	Food intake per chicken at a daily average basis	Energy accepted by a chicken at a daily average basis, kcal	Food consumption, kg/Gain (kg)
I 40 ppm FeMet	4.2	0.12	326	2.826
II 300 ppm FeMet	4.32	0.125	340	2.7
III 40 ppm FeSO <sub>4</sub>	4.37	0.128	348	2.71
IV 300 ppm FeSO <sub>4</sub>	4.28	0.122	331	2.66
V Control group	4.45	0.135	367	2.99

The differences between the utilisation of the feed by the test groups are not statistically significant. The best utilization of feed during the test period was observed in the group receiving ferrous sulfate in a dose 300 ppm (with 2.66 kg), and the highest consumption was observed in the control group (2.990 kg/kg gain). It was found that the concentration of ferrous sulfate (300 ppm) positively influenced the feed efficiency by 2.5% (relative to 40 ppm), while with the ferrous methionate effect of the dose was 4.5% (in favor of the higher). The control group had the highest consumption of feed.

The next table (Table 5) presents the results for the body mass of chickens in the different periods. The data shows that initial weight of the chickens is 183-200 g for all groups, which indicates for good uniformity within groups and between them. Initial weight corresponded to the age of the chickens (10 days). On the 17th day of the experiment the body mass of chickens for all groups was in the range from 0.756 to 0.838, as the highest it was in birds receiving 300 ppm Fe-methionate (without any statistical significance in the difference found). The final live weight fluctuated in the range of 1.694 g (for control group) to 1.838 g (for first group). There was a statistically higher final live weight of chickens receiving iron as ferrous methionate compared with the control group, at p < 0.05. The effect was higher with 1.5 -2% in favor of the lower dose (40 ppm). Ferrous methionate is Bulgarian medicinal products consisting of organic chelate complex of iron with methionine, studied previously by other authors with a positive effect in sows and gilts (Petrichev, 2006), as well as and pharmacokinetics in broiler chickens (Arnaudova-Matey, 2013). At this stage the most commonly used compound as a source of iron in the micronutrient compounds for different animal species, including and in broiler chickens, is still ferrous sulphate. In the groups receiving iron as ferrous sulfate was observed lower final body weight compared to recipients of iron methionate and birds from the control group, but the differences are statistically unreliable. Increasing the dose of iron in the feed of the chickens did not contribute to higher productivity. Similar results were obtained by Vahl and Klooster (1987), and we also in previous studies (Arnaudova-Matey, 2013). Similar was the trend of the indicator average daily gain. With lowest final body weight and gain were chickens in the control group.

Table 6 presents data for some poultry slaughter indicators. Weight of the carcass followed the trend in weight development. The carcass weight was highest in the first group (1.157 kg) which received ferrous methionate, and lowest in the control (1.086 kg). The effect of the dose was more clearly pronounced in the groups (3 and 4) receiving ferrous sulfate. In all treatment groups was observed trend for enlargement of the liver, as the highest weight it was in chickens received ferrous sulfate in a dose of 300 ppm. Despite the differences in the carcass weight and liver in the experimental groups compared with the control was not established reliability.

The results of the iron deposited in the liver of the broiler chickens treated with ferrous methionate or ferrous sulphate are

Table 5
Comparative data about of the body weight of broiler chickens continuously fed with a compound fodder containing ferrous methionate (FeMet) or ferrous sulphate (FeSO<sub>4</sub>)

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Groups	Initial body weight, kg	Body weight on the 17th day of the study, kg	Body weight at the end of the study, kg
I 40 ppm FeMet	$0.195\pm0.002$	$0.781 \pm 0.027$	1.838±0.079
II 300 ppm FeMet	$0.188 \pm 0.082$	$0.838 \pm 0.026$	1.825±0.028
III 40 ppm FeSO <sub>4</sub>	$0.200\pm0.094$	$0.819 \pm 0.027$	$1.819\pm0.046$
IV 300 ppm FeSO <sub>4</sub>	$0.193\pm0.062$	$0.756 \pm 0.027$	$1.781 \pm 0.058$
V Control group	$0.188\pm0.013$	$0.775\pm0.019$	$1.694\pm0.060$

Statistically significant difference compared to the control group calculated through the non-parametric method Mann-Whitney U test;\* - at P < 0.05; \*\*-at P < 0.01

Table 6
Comparative data of the indicators obtained in the slaughter of broiler chickens continuously fed with a feed with a compound feed containing ferrous sulphate (FeSO<sub>4</sub>) or ferrous methionate (FeMet)

Parameters Groups	Body weight before slaughter, kg	Weight of carcass, kg	Weight of the liver,
I 40 ppm FeMet	1.830±0.025	1.157±0.012	35.7±3
II 300 ppm FeMet	$1.820 \pm 0.031$	$1.150\pm0.010$	34.8±5
III 40 ppm FeSO <sub>4</sub>	$1.810\pm0.035$	$1.140\pm0.015$	35.4±7
IV 300 ppm FeSO <sub>4</sub>	$1.775\pm0.034$	$1.105\pm0.014$	36.6±4
V Control group	1.690±0.029	$1.086 \pm 0.022$	32.8±7

presented in Table 7. It shows that in the liver of birds, received with feed ferrous methionate (at a dose of 40 ppm and 300 ppm Fe $^+$ ) for 35 days was deposited more iron as compared to those, which received ferrous sulphate in the same concentrations (p < 0.05). There are differences in the accumulation of the iron in chickens from treated groups compared with the control, but the data are statistically significant only in Group I (40 ppm Fe $^+$ ) against the V $^{th}$  group (Control).

As expected, higher concentrations of iron in the feed are not a prerequisite to significant deposits of Fe<sup>+</sup> in the liver of chickens. Was verified that the chickens in comparison with mammals deposited more iron in the liver and according Davis et al. (1968) a significant part of it is in the form of hemosiderin. Both the ferritin and haemosiderin are metabolised in the body in the same way. It is evident that the dose has no significant influence on the controlled parameters in both comparable medicinal products of the iron (Figure 1).

Table 7
Depositing of iron in the liver of broiler chickens continuously fed with a compound feed containing ferrous sulphate (FeSO<sub>4</sub>) or ferrous methionate (FeMet)

Groups	Number of birds	Mean	SEM
I 40 ppm FeMet	5	632.0**	76.3
II 300 ppm FeMet	5	699.4**	110.5
III 40 ppm FeSO <sub>4</sub>	5	391	51.15
IV 300 ppm FeSO <sub>4</sub>	5	421.12	38
V Control group	5	437.8	77.6

Statistically significant difference compared to the control group: \* - at P < 0.05; \*\* - at < 0.01

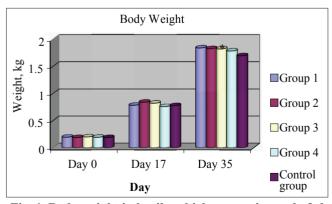


Fig. 1. Body weight in broiler chickens continuously fed with a fodder containing iron sulphate or iron methionate \* Statistically significant difference compared to the control group

# **Conclusions**

- The rheological performance (angle of repose, flow rate, Hausner's ratio) are better in the organic product compared to the ferrous sulphate (heptahydrate), which is a prerequisite for the better mixing with the feed.
- In terms of body weights of chickens and feed conventions in basic iron in the feed compounds (119.5 ppm) and the addition of 40 ppm and 300 ppm ferrous sulphate or ferrous methionate the trends are in favor of organic chelate product, but the differences are statistically unreliable.
- The ferrous methionate deposited more Fe $^+$  in the liver of chickens compared with ferrous sulphate, as the differences are statistically significant (p < 0.05).
- The dosage of used medicinal products has not a significant impact on the controlled parameters at the two comparable sources of iron.

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