

**Analele Universității *Spiru Haret***

**Seria Medicină Veterinară**

**Anul XII, nr. 12, 2011**

**EDITURA FUNDAȚIEI ROMÂNIA DE MÂINE  
BUCUREȘTI, 2011**

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ISSN: 1454-8283

Revistă cotate CNCSIS la categoria D

<http://www.spiruharet.ro/facultati/cercetare.phd>

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# ESOPHAGEAL-GASTRIC JUNCTION IN LABORATORY MAUSE

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## Abstract.

Aim of the study is to illuminate some data on gastric esophageal junction morphology in Guinea Pig brings explanations on mice unable to vomit. There are few literature data on the conformation and structure of the laboratory mouse esophagus and stomach. They try to explain why the mouse can not vomit. Deviating slightly to the left only in the cervical region the esophagus runs mainly in the cervical region the esophagus runs mainly midsagittally along the dorsal aspect of the trachea. Its length is about 30 mm (3, 4). Throughout its length the diameter is about 2 mm. Throughout its length the diameter is about 2 mm. The epithelium of the esophagus is moderately to extensively cornified. The submucosa is free of gland. Both layers of the muscular coat are made of bundles of skeletal muscle. The esophagus enters the middle of the lesser curvature. The **esophageal sphincter** is a circular muscle that surrounds the base of the esophagus. At its lower edge, it has muscle fibers that insert into the limiting ridge. So when the sphincter contracts, it not only constricts the walls of the esophagus, it also pulls the sides of the limiting ridge's "U" together, thus hiding and tightly closing the esophageal opening. Muscle layer is formed on the entire length of skeletal muscle fiber

**Keywords:** esophagus, stomach, Laboratory mouse

## Introduction

There are few literature data on the conformation and structure of the laboratory mouse esophagus and stomach. They try to explain why the mouse can not vomit. Dissection was performed on a total of 20 laboratory mice, using appropriate instruments. Histological preparations were processed by usual methods (Frossmann, W.G. L., Orzi, R., and col., 1969).

Deviating slightly to the left only in the cervical region the esophagus runs mainly in the cervical region the esophagus runs mainly midsagittally along the dorsal aspect of the trachea. Its length is about 30 mm (Knight, M. H. (1987). Throughout its length the diameter is about 2 mm. The epithelium of the esophagus is moderately to extensively cornified. The submucosa is free of gland. Both layers of the muscular coat are made up of loose bundles of skeletal muscle (Langer, Peter 2003) .

The stomach is oriented transversely and lies mainly caudal to the rib cage in the left cranial part of the abdominal cavity. The esophagus enters the middle of the lesser curvature. The stomach represents 0,5% of the body weight (Margaret J. (1965). The stomach consisted of two distinct parts: the left part (non-glandular, proventriculus or forestomach) was greyish, thin-walled and slightly transparent, and the right part (glandular or ventriculus) was white and thick-walled (Musser, G.G. and Carleton, M.D. 2005).

Anatomical textbooks on rats usually mention in passing that rats can't vomit. They tend to implicate the limiting ridge or the lack of striated muscle in the rat's esophagus, and sometimes both (Smith, E. M., and col., 1968, Stuart R.R., 1947), but these textbooks do not go into more detail about exactly *how* these features of a rat's anatomy prevent a rat from vomiting, or if there are any other features involved. Rats cannot vomit, but they do *regurgitate* occasionally (Spines, Robert L. 1982).

## Materials and methods

Dissection was performed on a total of 20 laboratory mice, using appropriate instruments. Formations resulting from anatomical dissection were photographed pictures are processed for better visibility. Esophagogastrica region pieces were taken to achieve histological preparations. Histological preparations were processed by usual methods. The topographical situation, omentum connections and arterial supply to the esophagogastric junction were examined macroscopically. Dried specimens were made to study the morphological form and internal structures.

## Results and discussion

The esophagus is a hollow body with laminated musculomembranoasa structure which connects farige and stomach. After the trajectory it can be systematized into three portions: cervical, thoracic and abdominal. Cervical portion is one that crosses the entire length of neck, it starts at the caudal end portion of the pharynx esophagus (esophageal vestibule) and ends in the right breast coming in to the first pair of ribs. Cervical portion has a length of about 15 mm initial portion is located in the median dorsal neck trachea and in the middle portion is located on the left side of the neck so that the caudal portion to be located again crosses over traheei. Portiunea thoracic mediastinum entire length with a length of about 20 mm. Abdominal portion of the laboratory mouse has a length of several mm are easily noticed in the abdominal cavity ( fig. 1). Layered wall of the esophagus consists of mucosa, submucosa, muscular and serous start. Stratified epithelial lining shows a floor disposed on the basement membrane. This separates the chorion epithelium. Epithelium is cornificat. Esophageal submucosal glands is lacking. Muscle layer is formed on the entire length of skeletal muscle fibers. The mause esophagus has two layers of striated muscle (outer longitudinal and inner circular), which become smooth near the attachment point with the stomach. The esophagus is closed off from the stomach by the **gastroesophageal barrier**, which consists of the **crural sling**, the **lower esophageal sphincter**, and the several milimeters of intraabdominal esophagus that lie between them. Humans also have a crural sling and an esophageal sphincter, but ours are placed right on top of one another. In mause, they are separated by several millimeters of intraabdominal esophagus (fig 1).

The **esophageal sphincter** is a circular muscle that surrounds the base of the esophagus. At its lower edge, it has muscle fibers that insert into the limiting ridge (Fig 2). So when the sphincter contracts, it not only constricts the walls of the esophagus, it also pulls the sides of the limiting ridge's "U" together, thus hiding and tightly closing the esophageal opening



Figure 1- portion of the thoracic and abdominal esophagus in mouse

The mouse stomach is a stomach a one compartment type compound. Recurbat the shape of a bag-shaped "U". The stomach consisted of two distinct parts: the left part (non-glandular, proventriculus or forestomach) was greyish, thin-walled and slightlytransparent, non-glandular section that receives the esophagus and serves as a holding chamber for food, (Its walls are similar to those of the esophagus) and the right part (glandular or ventriculus) was white and thick-walled. (Fig. 1,2) The mucosa of the glandular (right) part was lined by simple columnar epithelium. The lamina propria was occupied by simple tubular, gastric glands. The cells of these glands were discernible in 3 distinct zones. The chief cells,showing intense basophilia, occupied the lower third region of the gland; the parietal cells were predominantly present in the upper half of the glandular tubule. A few of them appeared to be admixtured with the chief cells in the lower third of the gland. The neck cells were evident near the openings of the tubular portion of the gland at the gastric pits. The lamina muscularis mucosae was thin. The tela submucosa consisted of loose connective tissue with blood vessels. The tunica muscularis was thick, consisting of an inner spiral and an outer obliquely circular layer. Ganglion cells were present between these muscle layers. The tunica serosa was thin. The nonglandular (left) part of the stomach was lined by keratinized, stratified squamous epithelium. The stratum granulosum was distinctly visible. The thickness of the tunica muscularis decreased from the nonglandular to the glandular stomach.



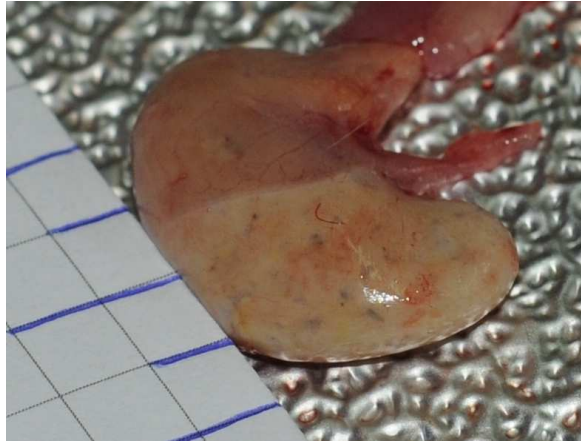


Figure 2 – conformatia externa a stomacului

The forestomach and the corpus are separated by a low fold of tissue called the **limiting ridge** (*margo plicatus*). The limiting ridge extends circumferentially from the large curvature of the stomach to the small curvature, just below the esophagus. At the esophagus, the course of the limiting ridge bends into a U-shape and almost surrounds the esophageal opening ( fig. 3).

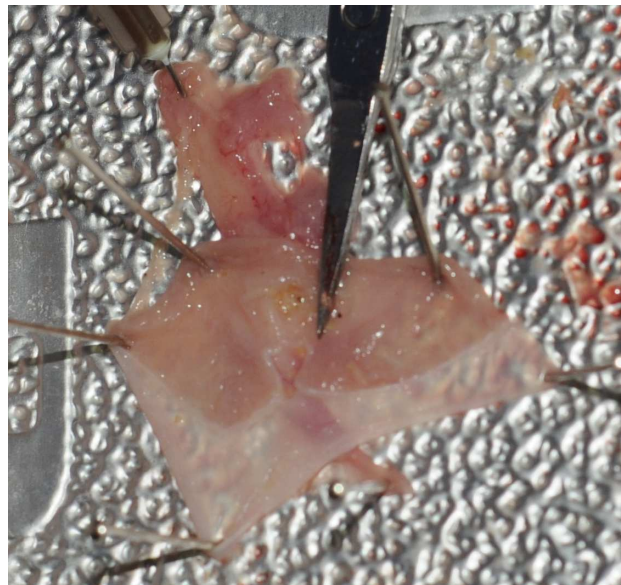


Figure 3 - limiting ridge (*margo plicatus*) and cardia orifice

Stomach in mice show the characteristic four compartment stomach tunics: mucosa, submucosa, muscular and serous.

On histological examination of gastric mucosa is found to continue occupying the esophageal mucosa type aglandulara portion of the stomach lining cardiale as being

located in the middle of the stomach cavity to limit the glandular portion astomacului. The left bow portion notice a gas storage area resulting from digestion stomach, forming diverticula. In this area of the esophageal epithelium is further ( fig 4). Epithelium cardiale area is soft layered pavement type, consisting of basal layer, intermediate layer and a layer pavement (fig 5). Further stratified pavement epithelium soft cardinal diverticula in the fold, the crossing is by simple prismatic epithelium stomach composed of secretory prismatic cells of the mucous type. Corion cardiei mucosal area is composed of connective fibers in the muscular lining is. Submucosa consists of connective tissue laxity and muscular skeletal muscle consists of fibers that are continuous muscular glandular region. Thus there is a double compartment stomach in mice which consists of a zone type aglandulara esophageal and glandular area (stomach itself). Boundary between two regions is the area that is narrow cardiale lining edge to form limiting ridge (*margo plicatus*).

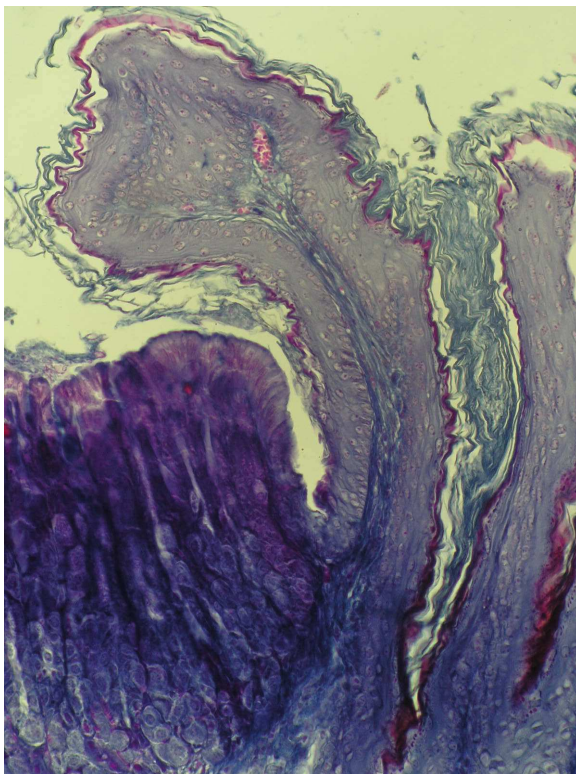


Fig. 4 - Mouse stomach - epithelial transition zone, Col. tricromică Mallory; Ob. 20x

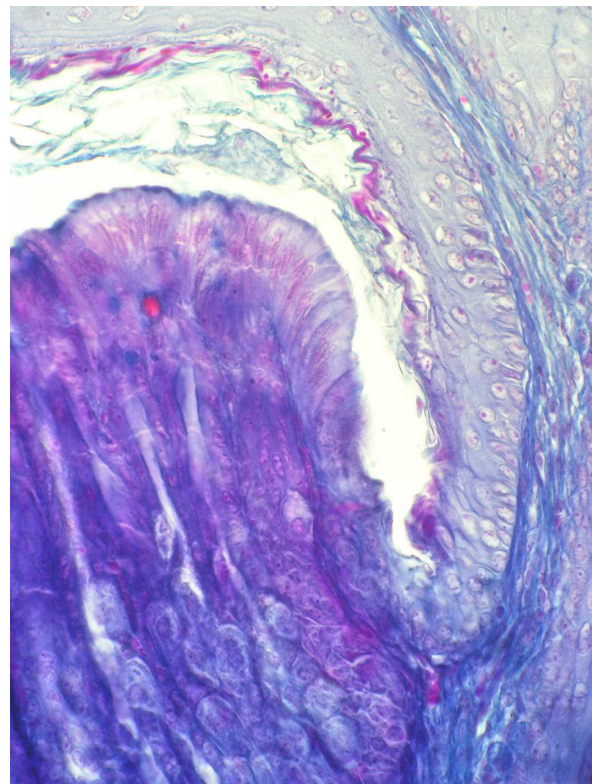


Fig. 5 – Mouse stomach - epithelial transition zone; Col. tricromică Mallory; Ob. 40x

Glandular mucosal area is characterized by the presence of a simple prismatic epithelium composed of epithelial cells with high basal pole located on the basal membrane, nuclei are located in the lower third and the apical pole contribute to mucosal surface.

Secretory epithelial cells are mucous cells of closed type, with a role in the development and elimination by merocriție mucinuous. Glandular stomach mucosa in the chorion, composed of connective tissue laxity, there are fibers and connective cells, blood and lymphatic vessels and nerve fibers. The thickness of the region corion background are obvious gastric glands with simple tubular part composed of basal cells, principal cells and marginal cells.

Muscularis level in the transition, it is composed of striated muscle fibers that are continuous longitudinal layer of smooth muscle fibers. The longitudinal layer of smooth muscle fibers are inserted through the connective fibers, skeletal muscle fibers from the region cardiei. In this area, is the original type muscular and striated muscle is an area of transition from a striated muscular type by a smooth muscular type. (fig 6).

În secțiunile histologice din această zonă se constată prezența fibrelor musculare striate de tip scheletic, ce au o dispunere longitudinală, prezentând caracteristic pe suprafața lor alternanța de discuri clare și întunecate

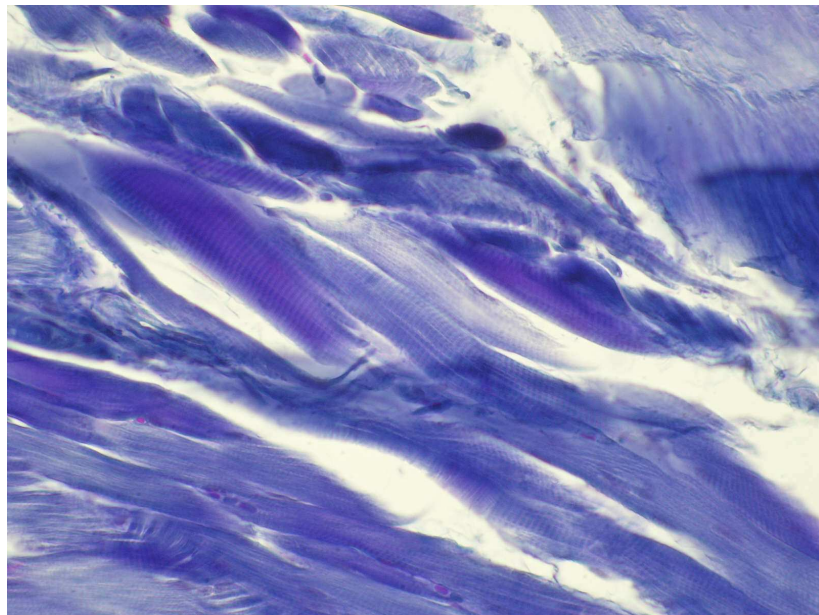


Fig.6 - Mouse stomach - an area of transition from striated muscular type in a smooth type; Col. tricromică Mallory; Ob. 40x

The section in Figure 3 can be seen crossing the zone presence of striated muscular type in the flat type, fiber type skeletal striated in longitudinal arrangement continues with a compact layer of smooth muscle fibers with longitudinal arrangement also.

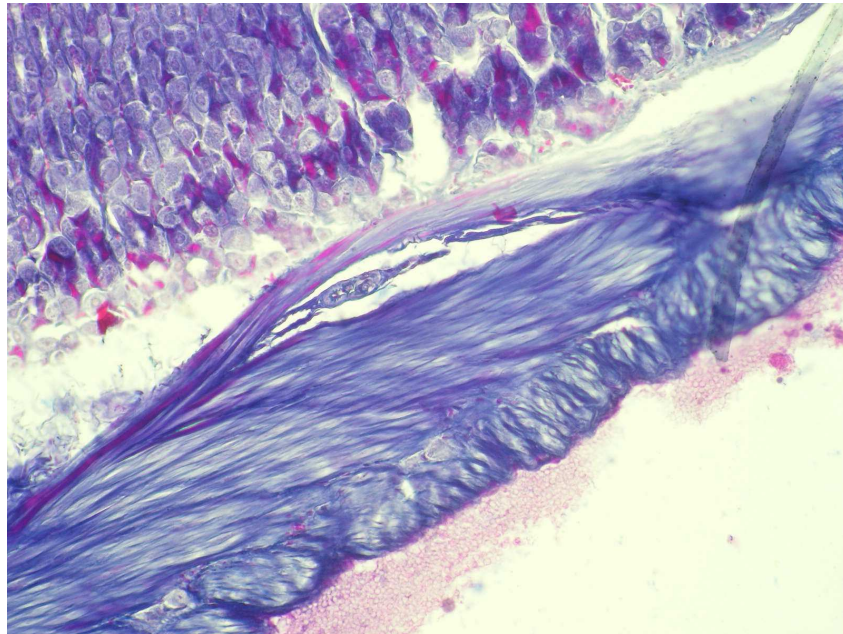


Fig. 7 - Mouse stomach - smooth muscular type in the glandular compartment; Col. tricromică Mallory; Ob. 20x

Muscular stomach, after more than cardiei area, is exclusively composed of smooth muscle fibers. In the right background and pyloric areas, muscled consists of smooth muscle fibers arranged in two levels: internal circular and external longitudinal between the two plans are being muscular plexus Auerbach vegetation with a small amount of tissue laxity. (fig 7)

### Conclusions

1. Esophageal mucosal is cornificated. Esophageal submucosal glands is lacking.
2. Muscle layer is formed on the entire length of skeletal muscle fibers
3. At the esophagus, the course of the limiting ridge bends into a U-shape and almost surrounds the esophageal opening
4. Esophageal submucosal glands is lacking
5. stomach shows a bottom region extends into the left dorsal diverticulum with a stomach that is part of the aglandular stomach portion
6. limit the portion of the stomach and aglandular portion is represented by limiting ridge
7. limiting ridge is in its structure in the right hole cardia skeletal muscle

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# IMMUNOLOGICAL DETERMINATIONS IN NONSPECIFIC MODULATORS ADJUVANT THERAPY IN SOME DISEASES IN DOGS

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## *Abstract*

*For carrying out the work have been taken under the observation more clinical cases (13 cases), with a different etiology in which adjuvant therapy was applied, using two products with nonspecific immunomodulatory role (Levamisol and Polidin). Treatments were applied to canine individuals with different breeds and ages.*

*The clinical evolution was monitored during treatment cases, the purpose of these treatments, and determination of immunological parameters (leukocyte formula). In this respect, two separate blood samples have been taken at the beginning of therapy and after 14 days from the first harvest.*

*The WBC final results chart has shown a significant increase (from 28.2% to 36.8%) of the percentage of lymphocytes in animals treated with Polidin, which demonstrates the quality of this product as non-specific immunomodulator. In the situation of Levamisol, the leukocyte formula has shown visible changes only at the level of eosinophils which have fallen to the final harvest (4.83%) from baseline (7.5%). One can say that after the experiment, the product Polidin, has a higher efficiency than the nonspecific immunomodulator Levamisole.*

**Key-words:** canine, immunostimulation, leukocyte formula.

## *Introduction*

Immunomodulation refers to the control of the immune response by medical techniques, methods and substances used in order to induce, stimulate, amplify, or on the contrary, to reduce or inhibit the immune response to the therapeutic or experimental needs(1,3).

In various situations of medical and veterinary pathology, therapeutic or prophylactic remedy can be only one acting on the immune response. This remedy, either stimulates and enhances immune responses in some infectious disease or inhibit the immune response to organ transplantation (5).

Defense capability is the result of a complex cellular and humoral factors, specific and non-correlated to the complex mechanisms of the immune system. Together with other mechanisms that contribute to maintaining the integrity of the morpho-cells, soluble mediators, immunoglobulins, immune system components, constitute a whole, a system equipped with wide possibilities of response to bacteria, viruses, parasites and other etiological agents (2, 4).

The evolution of clinical cases during the canine treatments, the result of these treatments, and determination of immunological parameters (leukocyte formula), after application of adjuvant therapy have been followed.

## ***Materials and methods***

For carrying out the work, 13 cases with different etiology have been taken under clinical observation and they were applied adjuvant therapy. Two products were used with non-specific immunomodulatory role (Levamisol and Polidin). Treatments were applied to canine of different breeds and ages. Polidin is a polibacterial immunomodulator, consisting of a mixture of 13 species of gram-positive and gram-negative bacterial suspensions, heat-inactivated, partially through ball lysates (1%) dilute in chlorinesodium (0.785%) with a standard concentration of 48 million inactivated bacteria/ml product.

It was run subcutaneously or intramuscularly one ampoule (1 ml) per animal for a period of 5-7 days.

Levamisol is a broad spectrum antihelmintic and an unspecific immunostimulating. The product is administered subcutaneously, the dose for all species is 1 ml/10 kg bodyweight.

As immunostimulating, it has been calculated one third of the antihelmintic dose for 3 days with booster in three days.

To make leucocyte formula two blood samples were developed: at baseline and after 14 days from the first harvest.

### ***Methods for diagnosing and evaluating the effectiveness of therapy:***

- clinical observation of animal
- bacteriological examination
- mycological examination
- coproparasitologic examination
- virusological examination
- WBC count

## ***Results and discussion***

Graph 1 shows that after Polidin immunostimulation the percentage of neutrophils (mean) decreased from 67.6% initially to 61.8 harvested after 14 days. The percentage of lymphocytes has a substantial increase from 28.2% to 36.8%. The other keyparameters are unchanged. The percentage of lymphocytes showed a significant increase in the final harvest, which shows the immunostimulating capacity of Polidin.

**Table 1**

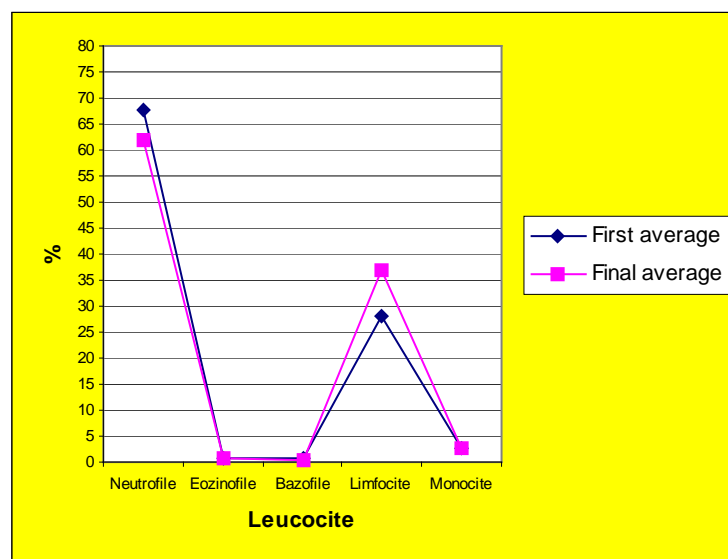
**The total number of leukocytes (%) - Polidin average inoculated cases original collection**

Case	Neutrophils	Eosinophils	Basophils	Lymphocytes	Monocytes
1	57	0	3	35	5
4	70	0	0	27	3
5	78	1	0	18	3
7	68	0	1	28	3
12	65	2	0	33	0
<b>Average</b>	<b>67,6</b>	<b>0,6</b>	<b>0,8</b>	<b>28,2</b>	<b>2,8</b>

**Table 2**

**The total number of leukocytes (%) – Polidin average inoculated cases - harvest after 14 days**

Case	Neutrophils	Eosinophils	Basophils	Lymphocytes	Monocytes
1	51	0	2	41	6
4	66	1	0	32	1
5	66	0	0	43	1
7	64	0	0	34	2
12	62	3	0	34	1
<b>Average</b>	<b>61,8</b>	<b>0,8</b>	<b>0,4</b>	<b>36,8</b>	<b>2,6</b>



*Graph 1. Polidin average inoculated cases*



After treatment with immunostimulating Levamisol (Graph 2) were observed only relevant amendments on the percentage of eosinophils, which decreased from 7.5% initially to 4.83 at harvest after 14 days.

Based on percentage growth of lymphocytes (from 28.2% to 36.8%) after treatment with Polidin, after immune treatments in the present work, one can deduce that Polidin has a greater immunomodulation ability than Levamisole.

**Table 3**

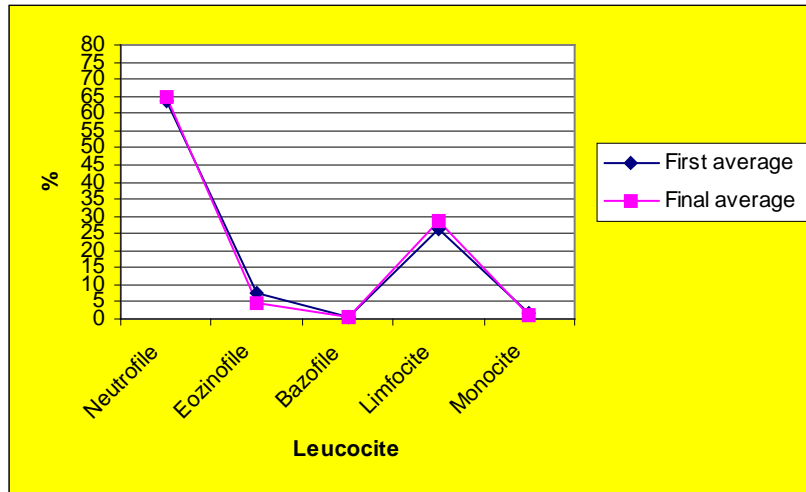
**The total number of leukocytes (%) – average Levimasol inoculated cases  
- original collection**

Case	Neutrophils	Eosinophils	Basophils	Lymphocytes	Monocytes
2	64	7	0	27	2
3	61	12	0	23	4
6	61	6	2	28	3
9	68	5	0	27	0
10	68	4	0	28	0
13	61	11	2	26	0
<b>Average</b>	<b>63,83</b>	<b>7,5</b>	<b>0,66</b>	<b>26,5</b>	<b>1,5</b>

**Table 4**

**The total number of leukocytes (%) –average Levimasol inoculated cases  
- harvest after 14 days**

Case	Neutrophils	Eosinophils	Basophils	Lymphocytes	Monocytes
2	59	6	0	34	1
3	64	6	1	27	2
6	63	5	1	30	1
9	67	3	0	29	1
10	72	2	0	25	1
13	63	7	1	28	1
<b>Average</b>	<b>64,66</b>	<b>4,83</b>	<b>0,5</b>	<b>28,83</b>	<b>1,16</b>



Graph 2. Levimasol average inoculated cases

### Conclusions

1. Out of the 13 cases studied, cure or improvement of clinical signs were achieved in 11 cases.

2. It was noted that after the Polidin immunostimulation the percentage of neutrophils (mean) decreased from the initial to final harvest. The percentage of lymphocytes showed a significant increase in the final harvest, which shows the immunostimulating ability of Polidin.

3. After treatment with immunostimulating Levamisol relevant changes were observed only in the percentage of eosinophils, which decreased significantly at the end, from the original collection.

4. Based on the percentage of lymphocytes increase after treatment with Polidin, after immune treatments in the present work, one can deduce that Polidin has a greater immunomodulating ability than Levamisol.

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## TRANSPORTATION RESEARCH ON STRESS IN BROILERS - BIOCHEMICAL REACTIONS

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### **Abstract**

*Serum and plasma biochemical reactions were analyzed in 2 groups of broilers in the age of 42 days subject to transportation stress (group 1 auto transport distance of 2 km, 2.56 hours for transportation during the summer and the group 2 transport distance of 65 km in 6.31 hours in summer).*

*For this purpose, blood samples were taken randomly from the axillary vein of 100 chickens in two stages: before and after transport. 10 biochemical parameters were determined: total protein, albumin, glucose, total lipids, glutamic oxalacetic transaminase (GOT), glutamic pyruvate transaminase (GPT), creatine phosphokinase (CPK), corticosterone, adrenaline and noradrenaline.*

*Between the two stages there were significant differences in the following parameters: glucose  $170 \pm 25$  mg/dl, stage 1,  $195 \pm 25$  mg/dl, stage 2, glutamic oxalacetic transaminase (GOT)  $125 \pm 30$  U/l,  $140 \pm 40$  U/l, creatine phosphokinase (CPK)  $270 \pm 45$  U/l and  $330 \pm 90$  U/l,  $3.53 \pm 0.45$  corticosteronul ng/ml stage 1,  $5.30 \pm 0.6$  ng/ml stage 2, adrenaline  $8.87 \pm 1.68$  ng/ml and  $12.2 \pm 1.3$  ng/ml, noradrenaline  $14.3 \pm 1.6$  ng/ml and  $17.8 \pm 2.1$  ng/ml. For the other parameters analyzed total protein, albumin, glutamic pyruvate transaminase (GPT) have not been significant value differences between the two phases.*

*Based on major findings, the main serum and plasma biochemical changes recorded after transport, on landing in both groups were: hyperglycemia, increased activity of GOT, CPK, increased corticosterone, adrenaline and noradrenaline. The data confirm other results from literature.*

**Keywords:** chicken meat, transport stress, biochemical reactions

### **Introduction**

Transport of animals, including chickens for meat not long ago was a much-debated issue internationally, only economically, unlike today when it became a wide audience concern, given the many public and political implications that it has on welfare. Mutation occurred on the approach to transport animals is a consequence of their different interests lie in contemporary society, ethical and moral question, on the one hand and economic and political on the other side. Meat chickens during transport according to the specialized work presents a high level of mortalities from fractures of the wings, the legs, the bruises, the bruises, the calou and biochemical and hematological changes. The period of preparation for the accession of our country into the EU in 2007 has led to knowledge and assessment of these indicators

during transport in our country's conditions for chickens for meat and its implementation based on the necessary measures to align with EU requirements .

### Materials and methods

Were analyzed in serum and plasma biochemical reactions two batches of broilers aged 42 days subject to transportation stress (group 1 auto transport distance of 2 km, 2.56 hours for transportation during the summer and group 2 transporter from a distance of 65 km in 6.31 hours in summer). For this purpose, blood samples were taken randomly from the axillary vein of 100 chickens in two stages: before and after transport. Samples, a quantity of 2 ml syringes were collected without anticoagulant for serum biochemistry and syringes with anticoagulant substance for plasma biochemical measurements were determined 10 biochemical parameters: total protein, albumin, glucose, total lipids, glutamic oxalacetic transaminase (GOT), glutamic pyruvate transaminase (GPT), creatine phosphokinase (CPK), corticosterone, adrenaline and noradrenaline. For determinations were formalized methods used in the laboratories of its kind in Romania and immunoenzymatic methods for hormones.

### Results and discussion

In Table.No 1 and Figures 1,2,3,4,5,6 are presented the 10 values of biochemical parameters in group 1 to determine your chickens for meat transported from a distance of 2 km in 2.56 hours in summer

**Table 1**

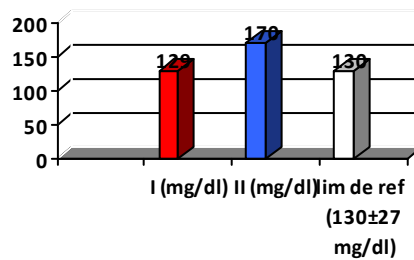
**The values of serum biochemical parameters in group 1**

Nr	Analyzed parameter	U/M	Values recorded		Values of the reference
			Disembark	After disembark	
1	Total protein	g/dl	3,6±0,4	3,65±0,6	3,5±0,5
2	Albumine	g/dl	1,57±0,6	1,67±0,5	1,5±0,5
3	Glucose	mg/dl	129,3±11	170±25	130±27
4	Total lipids	mg/dl	590±150	608 ±120	620±130
5	Glutamic oxalacetic transaminase (GOT)	U/L	85±25	125±30	70±40
6	Glutamic pyruvate transaminase (GPT)	U/L	10±9	11±10	12±8
7	Creatine phosphokinase (CPK)	U/L	155±30	270±45	150±100
8	Corticosterone	ng/ml	1,95±0,30	3,53±0,45	2,15±0,1
9	Adrenaline	ng/ml	6,17±1,7	8,87±1,68	6,1±1,4
10	Noradrenaline	ng/ml	10,7±1,6	14,3±1,6	11,7±1,4

Total protein, albumine, total lipids and glutamic pyruvate transaminase after landing one group determined to have been similar to the reference values.

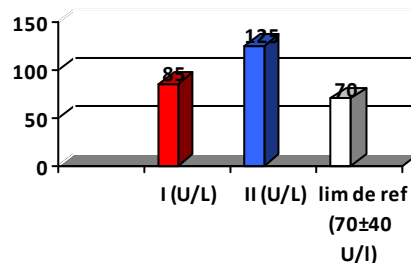
Glucose after landing, was higher ( $170 \pm 25$  mg / dl) than in the previous stage and the reference value (Fig. 1).

Data showed a statistically significant increase in blood glucose levels determined after landing, compared to the level determined before loading, ( $p < 0.001$  - highly significant), which demonstrates an early stress response even after transport distance of 2 km.



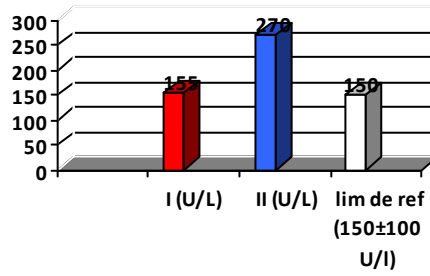
**Fig. No 1** Dynamics of average blood glucose values No.1 chickens for meat in group No. 1, before boarding and after landing

Glutamic oxalacetic transaminase (GOT) after landing, showed a value of  $125 \pm 30$  U / L (Figure No. 2) statistically higher than pre-boarding ( $85 \pm 25$  U / L). GOT activity increased significantly in value after landing from the value determined before loading ( $p < 0.001$  - highly significant), indicating an intense stress reaction transport.



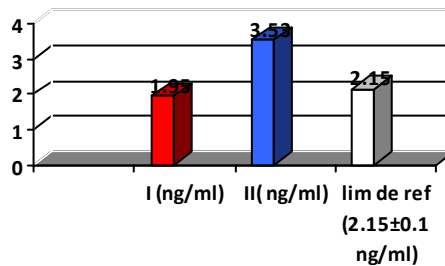
**Fig. No 2** Dynamics of mean values of SGOT activity in chickens for meat in group No. 1, before boarding and after landing

Creatine phosphokinase (CPK). After landing, CPK values were determined for  $270 \pm 45$  U / L (Fig No 3), being significantly higher ( $p > 0.001$ ) value obtained prior to boarding,  $155 \pm 30$  U / L. Increased CPK activity is correlated with that of TGO, as reactions to the synergistic action of stress factors during transport, and possible injuries possibly incurred during handling and muscle of the journey.



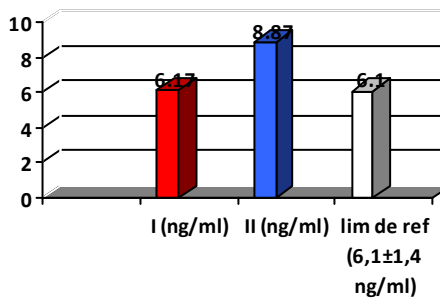
**Fig. No.3** Dynamics CPK activity in chickens from group 1 before and after transport (average)

Corticosterone after transport to the landing, the value was  $3.53 \pm 0.45$  ng / ml. Value as determined by plasma transportation landing was significantly greater than the value determined before loading ( $p < 0.001$  - highly significant)



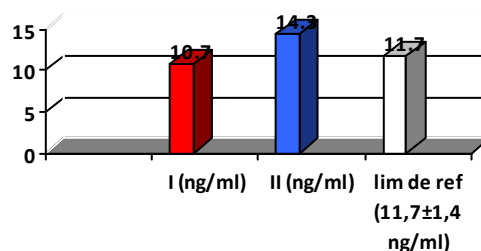
**Fig. No 4** Dynamics of the corticosterone average values for meat chickens in group 1 before boarding and after landing

Adrenaline caused by transport, land value has been increased plasma epinephrine  $8.87 \pm 1.68$  ng / ml greater than the value determined prior to boarding and the reference. After transport, the landing was a very significant increase ( $p < 0.001$ , Figure No. 5) the value of plasma adrenaline levels recorded prior to boarding.



**Fig. No.5** average dynamics of adrenaline for meat chickens in group 1 before boarding and after landing

Noradrenaline, after shipment, landing norepinephrine value was  $14.3 \pm 1.6$  ng / ml. After transport, the landing was a significant increase ( $p < 0.001$ , Figure No. 6) the amount of plasma norepinephrine, value determined prior to boarding.



**Fig. No.6** norepinephrine dynamics of the average meat chickens in group 1 before boarding and after landing

In Table.No 2 and Figures 7,8,9,10,11,12 are presented values the 10 biochemical parameters determined in group 2 of the chickens for meat transported from a distance of 65 km in 6.31 hours in summer

**Table 2**

**The values of serum biochemical parameters in group 2**

Nr	Analyzed parameter	U/M	Values recorded		Values of the reference
			Disembark	After disembark	
1	Total protein	g/dl	3,54±0,3	3,6±0,4	3,5±0,5
2	Albumine	g/dl	1,84±0,3	1,82±0,3	1,5±0,5
3	Glucose	mg/dl	132±14	180±20	130±27
4	Total lipids	mg/dl	680±11	700±120	620±130
5	Glutamic oxalacetic transaminase (GOT)	U/L	80±39	140±40	70±40
6	Glutamic pyruvate transaminase (GPT)	U/L	10±	14±7	12±8
7	Creatine phosphokinase (CPK)	U/L	165±40	330±90	150±100
8	Corticosterone	ng/ml	2,6±0,45	5,3±0,6	2,15±0,1
9	Adrenaline	ng/ml	6,8±1,1	12,2±1,3	6,1±1,4
10	Noradrenaline	ng/ml	10,6±1,1	17,8±2,1	11,7±1,4

Total protein, albumin, total lipids and glutamic pyruvate transaminase determined in group 2 after landing, had values similar to limit of reference.

Glucose after landing, was higher ( $180 \pm 20$  mg/dl) pretransport stage and the reference value (Figure No. 7). Individual minimum blood glucose value after landing, was 160 mg / dl, and a maximum of 200 mg / dl Data showed a statistically significant increase in blood glucose levels after transport compared with the level determined prior to boarding ( $p < 0.001$  - highly significant).

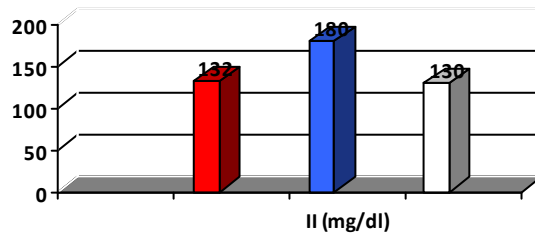


Fig.nr.7 Dynamics of average blood glucose values chickens for meat in group No. 2, before boarding and after landing

Glutamic oxalacetic transaminase (GOT) After landing, GOT activity was  $140 \pm 40$  U / L (Fig No. 8), significantly higher than the pre-boarding ( $80 \pm 39$ U / L). TGO increased activity of the very significant ( $p < 0.001$ ) after landing led to the samples before shipment.

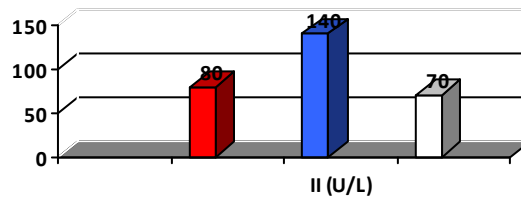


Fig.no 8 Dynamics of the mean values of GOT activity in chickens for meat in group No. 2, before boarding and after landing

Creatine phosphokinase (CPK) After landing, CPK activity was  $330 \pm 90$  U / l (Fig No. 8), significantly greater than the value obtained prior to boarding. After landing chickens was a statistically significant increase in CPK to the values determined before loading ( $p < 0.001$  - highly significant).

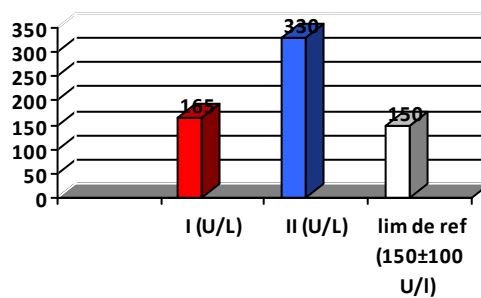


Fig. No.9 Value dynamics mean CPK values for meat chickens in group No. 2 finish, before boarding and after landing

Plasma Corticosterone shipment, landing value was  $5.3 \pm 0.6$  ng / ml. After transport, the landing was a very significant increase ( $p < 0.001$ , Figure No. 10) value corticosterone plasma level determined prior to boarding.



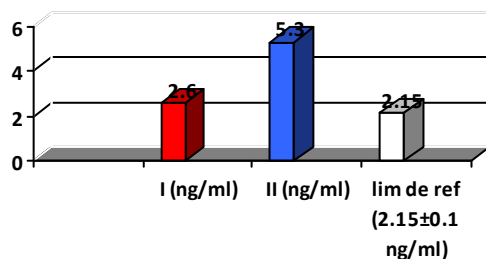


Fig. No.10 Dynamics of the average corticosterone meat chickens in group No. 2 before boarding and after landing

Adrenalin after transport, value was  $12.2 \pm 1.3$  ng / ml higher than the value determined before boarding. After transport, the landing was a very significant increase ( $p < 0.001$ ) plasma adrenaline value to the level determined prior to boarding (fig no.11).

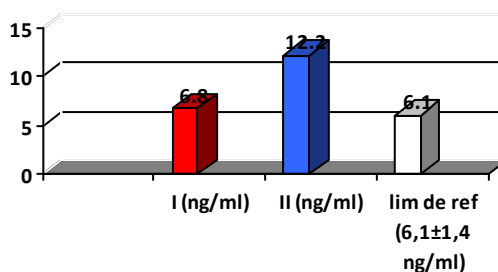


Fig. No.11. Dynamics of the average adrenaline meat chickens in group No. 2 before boarding and after landing

Noradrenaline as transport, was  $17.8 \pm 2.8$  ng / ml. After shipment, the landing was a very significant increase ( $p < 0.001$ , Figure No. 12) the value of plasma norepinephrine compared with the level determined prior to boarding.

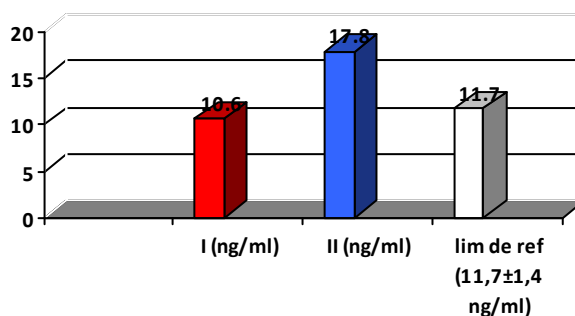


Fig. No. 12 Dynamics of norepinephrine mean values for meat chickens in group No. 2 before boarding and after landing

The analysis presented shows that: corticosteronul at No. 2 after the transport group, the landing has increased as a result of the action on the adrenal of physical and psychological stressors, catecholamines (adrenaline and noradrenaline), showed high values after landing, as a result of the action stresorilor (physiological and psychological) on medulosuprarenalei. Note that the value differences between the stages of determining hormone levels were higher in group than in group No.1 No.2In Table.3 and Figures 13,14,15,16,17,18 compared the values of are six biochemical parameters determined at group No. 2 and Lot 1

Analyzing the data in Table No.3 and figure No.13-18 resulting for the six parameters that determine: glucose in group 2 showed higher values compared with group No. 1, respectively  $180 \pm 20$  mg / dl compared to  $170 \pm 25$  mg / dl, the difference being significant ( $p < 0.2$ , Figure 13). The finished meat chickens in both groups after transport to an increase in glucose transport in response to stress. Differences between the two groups after transport of blood glucose was not statistically significant, which demonstrates that brutal handling operation did not significantly affect blood sugar levels. Thus, compared to the reference value of  $130 \pm 27$  mg / dl in group No. 1 was a value of  $170 \pm 25$  mg / dl, and in group No. 2  $180 \pm 20$  mg / dl.

**Table 3**

**Comparison of biochemical parameters in chickens for meat in batches lot. 1 and 2 after transport**

No.	Analyzed parameter	U/M	Values recorded		Values of the reference
			Lot 1	Lot 2	
1.	Glucose	mg/dl	$170 \pm 25$	$180 \pm 20$	$130 \pm 27$
2.	Glutamic oxalacetic transaminase (GOT)	U/l	$125 \pm 30$	$140 \pm 40$	$70 \pm 40$
3.	Creatine phosphokinase (CPK)	U/l	$270 \pm 45$	$330 \pm 90$	$150 \pm 100$
4.	Corticosterone	ng/ml	$3,53 \pm 0,45$	$5,3 \pm 0,6$	$2,15 \pm 0,1$
5.	Adrenaline	ng/ml	$8,87 \pm 1,68$	$12,2 \pm 1,3$	$6,1 \pm 1,4$
6.	Noradrenaline	ng/ml	$14,3 \pm 1,6$	$17,8 \pm 2,1$	$11,7 \pm 1,4$

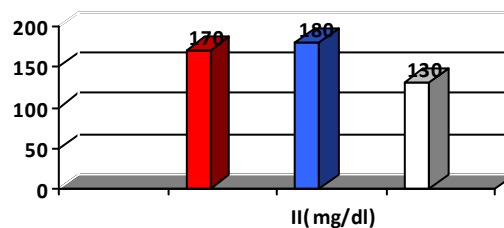


Fig. No. 13 Dynamics of average blood glucose values for meat chickens in group No. 1 to No. 2 after the transport group

Glutamic oxalacetic transaminase activity showed higher values in group no. No. 1 versus group 2, respectively  $140 \pm 40$  U / L, compared to  $125 \pm 30$  U / L, the difference being very significant ( $p < .001$ , Figure No. 14).

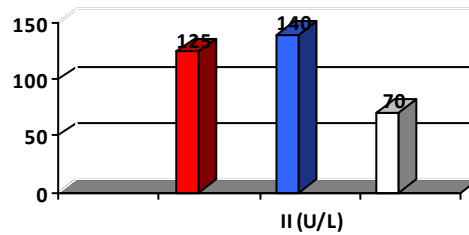


Fig. No. 14 Dynamics of average values for meat chickens GOT activity in group No. 1 to No. 2 after the transport group

Glutamic pyruvate transaminase showed slightly elevated in comparison with group 1 group 2, respectively  $14 \pm 7$  U / L vs.  $11 \pm 7$  U / L, the difference being significant ( $p < 0.2$ ).

Oxalacetic glutamic transaminase activity was significantly increased after transport group both No.1 and No.2 in the group. Thus, compared to the reference value of  $70 \pm 40$  U / L in group No. 1, after the transport has been a value of  $125 \pm 30$  U / L ( $p < 0.001$ , highly significant), and the lot. 2  $140 \pm 40$  U / L ( $p < 0.001$  - highly significant). Increased serum GOT activity is a result of changes in cell permeability, degenerative or inflammatory lesions of skeletal muscles (Mitchell 1998) and liver, caused by injuries during transport. Moreover, in group No. 2, increased GOT activity against consignments no. it was very significant ( $p < 0.001$ ) and  $140 \pm 40$  U / L compared to  $125 \pm 30$  U / L. Data shows that there is a positive correlation between increased activity of this enzyme and how to handle the chickens during transport and handling brutal grip caused intense changes in the skeletal muscle.

Glutamic pyruvate transaminase GPT presented values close to the baseline of  $12 \pm 8$  U / L, at  $11 \pm 10$  U / L in group No. 1 and  $14 \pm 7$  U / L in group No. 2. The activity of the creatine group showed higher values compared with group No.1 No.2 respectively  $330 \pm 90$  U / L to  $270 \pm 45$  U / L, the difference being very significant ( $p < 0.001$ ).

Creatine phosphokinase activity, skeletal muscle specific enzyme, had also increased after transport evident in both groups: in group 1 to  $270 \pm 45$  U / L to  $155 \pm 30$  U / L phase 1, while in group 2- $330 \pm 90$  U / L to  $165 \pm 40$  U / L, significant differences ( $p < 0.001$ , highly significant)

The increase in CPK activity may be correlated with changes in skeletal muscle during the action of stress factors during catching, loading and transportation. The intensity of action of these factors was correlated with increased activity by the enzyme. Thus in group 2, CPK was  $330 \pm 90$  U / L, compared with  $270 \pm 45$  U / L in group L, the difference being very significant ( $p < 0.001$ ).

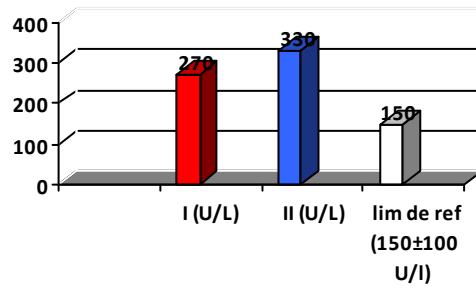


Fig. No. 15 Dynamics of average values of CPK activity in meat chickens in group No. 1 to No. 2 after the transport group

Corticosterone determined after landing the No. 2 group was  $5.3 \pm 0.6$  ng / ml and in group No. 1  $3.53 \pm 0.45$  ng / ml. The difference between the determined values is significant ( $p < .01$ , Figure No.16).

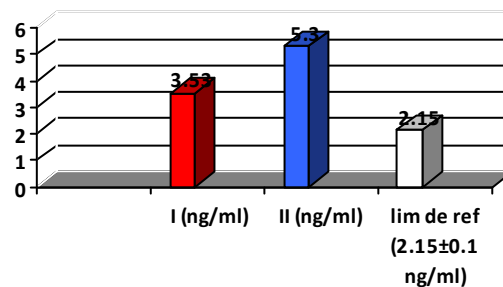


Fig. No.16 Dynamics of average values corticosterone chickens for meat after transport lots 1 and 2

Adrenaline was quite different, being  $12.2 \pm 1.3$  ng / dl in group No. 2 and  $8.87 \pm 1.68$  ng / ml. The difference between the determined values is very significant ( $p < .001$ , Figure 17)

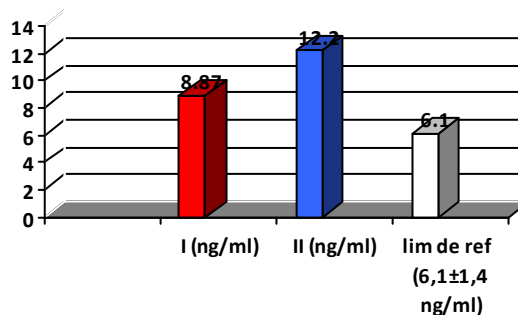


Fig. No.17 Dynamics of average values of adrenaline chickens for meat lots Nos. 1 and 2 after transport

Noradrenaline had different values, being  $17.8 \pm 2.1$  ng / dl in group No. 2 and to  $14.3 \pm 1.6$  ng / dl in group No. 1. The difference between the determined values is very significant ( $p < .001$ , Figure No.17).

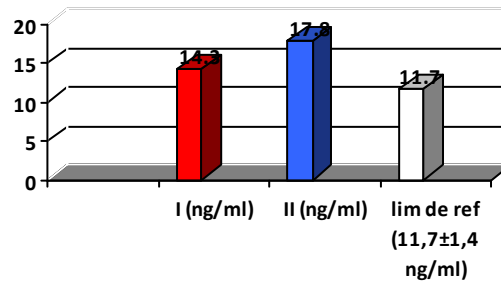


Fig. No. 18 Dynamics of average values for meat chickens noradrenaline lot. 1 and 2 after transport

### Conclusions

The analysis of the results of biochemical measurements, including hormonal for meat chicken flocks of lots Nos. 1 and 2, during transport, the samples taken before boarding and after landing, the distance of 65 km in 6.31 hours - Lot No. 2 and at a distance of 2 km in 2.56 hours - Lot No. 1, that:

1. The main serum biochemical changes, recorded by the transport, on landing in both groups were: hyperglycemia, increased activity of glutamic-oxalacetic transaminase (GOT) and creatine phosphokinase (CPK). Reactions recorded were very significant ( $p < 0.001$ ).

2 Changes in serum hormone parameters were the very significant increase ( $p < 0.001$ ) values corticosterone and catecholamines, adrenaline and noradrenaline respectively.

3. The biochemical changes caused by transport, the landing was intense, in group No. 2, to lot No. 1, the difference was significant between the values determined in blood sugar, GOT and CPK activity, corticosterone, adrenaline and noradrenaline which shows a positive correlation between transport duration and intensity of laboratory reactions.

4. He demonstrated a positive correlation between changes in serum hormone parameters analyzed, on the one hand and serum biochemical reactions of elsewhere.

5. Measurements show that the laboratory performed during the transportation of chickens for meat farms are home to slaughterhouses under stress whose intensity is related primarily to the duration of transport, calculated from boarding the first car phones in the holding cage to cage last landing mobile slaughterhouse.

7. Biochemical reactions are early, accurate and objective reason which can be used successfully in evaluating and monitoring the action of stress factors on the welfare of chickens kept for meat during transport.

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## MONITORING METABOLIC DISEASE OF DAIRY COWS IN BIOECONOMY CONTEXT

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### **Abstract.**

*Most of the metabolic diseases of dairy cows - milk fever, ketosis, retained placenta, and displacement of the abomasum - occur within the first two wk of lactation. In addition to metabolic disease, the majority of infectious disease experienced by the dairy cow, especially mastitis, but also diseases such as Johne's disease and Salmonellosis, become clinically apparent during the first two week of lactation. Metabolic disease is the most commonly recognized disease on dairy farms. While the pathogenesis is well known, metabolic disorders continue to occur. Metabolic diseases are associated, with one disease predisposing to another. Evidence suggests that metabolic disease affects host defence, and therefore, impacts the common infectious diseases of dairy cows. Risk for metabolic disease is affected by dietary formulation but is modified by cow behaviour and intake. Regardless of dietary formulation, the cow and management factors on a given farm may determine the impact of metabolic disease*

**Keywords:** metabolic disease, cow, and bio economy

### **Introduction**

Most periparturient abnormalities have some metabolic element as a component of the sufficient cause of clinical disease. Negative energy balance, fat mobilization and subsequent elevations in ketone body concentrations play a contributing role in the expression fatty liver syndrome, clinical ketosis, and abomasal displacement. A negative energy balance during transition may also increase the risk of retained placenta, metritis, and mastitis through impaired immune function. Subclinical disease incidence is far more common than clinical disease, frequently goes unnoticed and may be associated with significant clinical disease risks, impaired production and reduced reproductive performance. Prevention depends on several factors including proper transition cow nutrition and management, control of body condition; and may be helped through the use of certain feed additives such as propyleneglycol, rumen protected choline and ionophores [15,20,21].

The livestock sector plays a crucial role in the provision of global public goods and services. There are opportunities to alleviate many of the risks associated with the expanding sector and to develop its full potential in ensuring benefits for the poor with a gender equality perspective, and to encourage a more responsible use of increasingly scarce inputs and natural resources. This will require dynamic generation and adoption of new technologies, products and services as well as networks and institutional development within an enabling policy and regulatory environment. The vigorous growth of the livestock sector, its importance for income generation, food security, human

nutrition and health, and its impact on various public goods and services require careful attention by the international community [1]

#### ***Monitoring subclinical ketosis***

By most definitions, the theoretical testing period for transition cows would extend from 3 weeks prior to calving until 3 weeks after calving. Practically however, the most important time periods are: during the last week prior to calving and within the first 2 weeks after calving.

It is unusual for cows to develop subclinical ketosis precalving because the etiology of the condition depends on the homeorhetic drive for milk production. However, cows in an energy deficit precalving will start mobilizing energy reserves in the final week before parturition. This can be measured via serum or plasma non-esterified fatty acids (NEFA). The challenge for this precalving sample is predicting when the animal is going to calve. In the past, establishment of a serum bank and retrospective submission of samples relative to calving have been recommended [2,4]

#### **Serum BHBA**

In contrast to NEFA, serum BHBA should only be used post calving. The first two weeks are the primary risk period for sub clinical ketosis, defined by a serum concentration of 1400  $\mu\text{mol/L}$  BHBA or greater. Although BHBA is the most stable of the ketones, it is the most subject to variation associated with feed intake, thus all samples on a given farm should always be taken at the same time of day. In addition, hemolysis is known to artificially elevate values; therefore, haemolysed samples should be avoided. Other disadvantage of serum BHBA is the cost and the laboratory turn around time (minimum 24 hours). However, all things considered, serum BHBA analysis is the gold standard from which to compare cow-side tests. A reasonable goal is to have less than 2 cows per 10 with BHBA above 1400  $\mu\text{mol/L}$  in the first 2 weeks post-calving [8,12].

#### **Milk Ketone Tests**

Most milk ketone tests measure acetone and acetoacetate through a chemical reaction with nitroprusside that causes a colour change from white to either pink or purple. These tests in general are poorly sensitive in milk (<40%) but highly specific (>90%). One exception is the milk ketone test that measures BHB. It is marketed in Europe as "Ketolac BHB", in Japan as "Sanketopaper", and in Canada as "Keto-Test". This test has a much higher sensitivity in milk (>70%) and reasonably good specificity (>70%, up to 90%). This is a semi-quantitative test that allows choosing a lower threshold for screening to increase sensitivity, and a higher threshold for diagnosis to increase specificity.

#### **Urine Ketone Tests**

The urine ketone tablet tests are based on the same nitroprusside reaction as the milk powder ketone tests. These tests are highly sensitive (approaching 100%) but are poorly specific. Thus, they are great tests for ruling out sub clinical ketosis with a negative test result. However, their use overestimates a sub clinical ketosis problem because of a high probability of false positive reactions. If the urine test was used to evaluate the goal of less than 2 cows per 10 with BHBA above 1400  $\mu\text{mol/L}$  in the first 2 weeks post-calving, an adjustment of the goal to less than 5 cows per 10 with positive urine ketone tests would be required. However, recent works suggest that a 5 second interpretation using the Ketostix in urine is just as accurate as the Keto-Test in milk.



More work needs to be done to fully assess the utility of urine ketone tests sensitivity and specificity of the test must be incorporated into establishing goals, and intervention thresholds.

### **Herd Disease Records**

Herd records are important tools for monitoring the incidence of periparturient disease. Producers should set goals for the minimizing the incidence of metabolic disease. Herd consultants should periodically review herd performance relative to these goals. In addition, intervention levels should also be considered. Several diseases are associated with increasing age and this must be taken into account when assessing herd performance. For example, in monitoring and comparing herd incidence of milk fever and clinical ketosis, it is important to stratify this by parity. A high proportion of first lactation animals will likely give a herd a much lower incidence of milk fever and clinical ketosis, since risk increases with age.

### **Dry Matter Intake**

Clearly cows that are mobilizing NEFA precalving will have sub optimal dry matter intake. In a recently completed project, serum BHBA concentration in the first week post-calving was significantly associated with the average DMI in the week prior to calving. There was a significant increase in the risk of sub clinical ketosis (BHBA > 1400umol/L of blood serum) if the DMI was below 12 kg/d (OR=5.7, P=0.05) in the week prior to calving. If the DMI in the week prior to calving was below 11 kg/d, there was a greater risk of an animal developing sub clinical ketosis in the first or second week post-calving (OR=2.9, P=0.05). Thus measuring and monitoring the dry matter intake in the close-up group every week has utility. However, beware of group demographics relative to time of expected calving and parity, which can influence these parameters dramatically. Fresh cow intakes are generally less useful because we are primarily interested in the intakes of cows within the first three weeks post calving. If a fresh cow group exists, it is often composed of cows that may be several months post calving [5,13].

### **Identifying High Risk Herds**

Can herd incidence of certain diseases be used to decide whether a herd has a problem with sub clinical ketosis? Using data from a 25 herd study conducted in Guelph in 1995/1996, the median cumulative herd incidence of sub clinical ketosis was 41% in the first two months post calving, which crudely broke down into a threshold of 20% in week 1 and week 2 post calving. Summary data for each herd from each cows first DHI test post calving was used to assess the protein to fat ratio as a test at the herd level for classifying a herd as a high or low incidence herd for sub clinical ketosis. If more than 40% of cows in the herd at 1<sup>st</sup> DHI test had a protein to fat ratio of less than or equal to 0.75, those herds were likely to be problem herds.

This test had a sensitivity of 69%, and a specificity of 83%. Although more work needs to be done on herd level indicators of sub clinical ketosis, herd level protein to fat ratios appear to be better indicators of herd level issues than individual cow protein to fat ratios are of identifying cows with sub clinical ketosis problems. Additional analysis indicates that the herd incidence of displaced abomasum is positively associated with the probability of a herd having a high incidence (>20% in the first 2 weeks of lactation) of

sub clinical ketosis. In addition, if herds had greater than 10% of transition cows had a BCS > 4.0 at 3 wks precalving, that herd was extremely likely to have a problem with sub clinical ketosis.

### *Economics of Monitoring*

For the herd level monitoring interpretation, the savings achieved is in identifying a problem sooner rather than later, since nearly all problems will eventually be identified. A conservative estimate of the economics of a biweekly program suggests that a routine monitoring program would payback if one major problem was identified earlier than traditional means every 4 to 5 years. The economics of individual cow testing depends on the efficacy of treatment, accuracy of the test, cost of the therapy and prevalence of disease [1,18].

In the recent symposium: 8th WSEAS International Conference on Environment, Ecosystems And Development (ED'10) International Conference on Bioscience and Bioinformatics (ICBB '10) Advances in Biology, Bioengineering and Environment, 29-31 December 2010, organised by World Science and Engineering Academy and Society had been presented by Prof. dr. Lucian Ionita and col. (23) the article with title: „Pollution Control through Ecopathological Indicators for Nutritional and Metabolic Disorders by Dairy Cows from Farm Animals Ecosystems near Industrially Polluted Areas” in which was presented the following aspects:

- the ecopathological indicator regarding cattle farms located near polluting factories is represented by nutritional and metabolic disorders, that evolve asymptotically in animals, through low levels in Selenium or it's interference with other elements, including polluting ones.
- the cause/effect study can be explained by heavy metal and non-ferrous elements pollution discharged though industrial emissions that have interfered with Selenium in soil and plants also.
- another interference of Selenium is the one with water sulphates.
- the Selenium and vitamin E interfering can explain the health condition and reproductive status on milk cows.

### *Conclusions*

Metabolic diseases are interrelated, so that one disease increases risk for another. The energy-associated diseases include ketosis, displaced abomasum, fatty liver, retained placenta, metritis, and possibly mastitis.

Providing an environment for an adaptive cow response will remain key to health. Dairy advisors must take an active role in promoting quantitative monitoring to assist the producer. In addition to tracking average DMI, monitoring energy balance using milk or blood NEFA or ketone assays may be essential, and may provide an early warning of problems to come. Since disease represents failures (those cows who could not negotiate stress), analysis of disease incidence records must be conducted and compared to known risk factors, including BCS, DMI, pen moves, and concurrent disease. These areas are obvious points where nutritionists and veterinarians can interact in a cooperative relationship.

Subclinical ketosis is an important and common disease in lactating dairy cows. Prevention depends largely on effective dry cow nutrition and management. Given the cost of subclinical ketosis, the fact it is a common problem in early lactation, and the strong association with clinical disease, monitoring programs for subclinical ketosis during the first few weeks of lactation may be warranted. There are several cowside tests

for subclinical ketosis available; however, all of the current tests have their strengths and weaknesses. The design and frequency of a sub clinical ketosis-monitoring program will depend on the purpose of the program and the frequency of disease within the herd.

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## CANINE ADAPTATIVE CARDIAC HYPERTROPHY

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### **Abstract**

*The study sought to identify changes in adaptive compensation and the remodeling of heart occurred in 20 dogs with degenerative valvular chronic and acute, which were presented at the clinic of the Faculty of Veterinary Medicine and Clinical Doctor's Vet Universe, with clinical signs dyspnea, cough, syncope, exercise intolerance, weakness, and altered heart sounds.*

*Echocardiography were found: concentric hypertrophy with shortening fraction initially increased by 6% of normal, dilatation of four chambers of the heart with shortening fraction decreased by 10% of normal. ECG were found: sinus tachycardia, atrial extrasystole, cardiac hypertrophy, cardiac ischemia.*

*For large mitral stenosis, left ventricular pressure load leads to the appearance and adaptive concentric hypertrophy, but in cases of moderate mitral regurgitation, volume overload causes adaptive eccentric hypertrophy. In severe cases, complicated with cordage tendon rupture or papillary muscle, these forms have become maladaptive cardiac remodeling is associated with symptoms of heart failure syndrome.*

**Keywords:** dog, adaptive changes, cardiac remodeling

### **Introduction**

Cardiac remodeling is an adaptive mechanism that improves their heart function. To install it, it is necessary for pathogenic factors to act on a long period and moderate intensity. In heart disease an progressive decrease in cardiac performance install, by ensuring the flow of organ failure and loss of cardiac contractile efficiency. Adaptation of cardiac volume overload and pressure is achieved by compensatory functional mechanisms of hypertrophy and dilation (4).

Relationship between ventricular distension pressure, end diastolic volume and stroke volume was discovered by Starling (*Starling's law of the heart*). Small increases pressure distension causes large changes in ventricular end diastolic volume, on which will increase in stroke volume and cardiac output by default. End diastolic pressure increases gradually, leading to an increase in cardiac output up to a maximum, corresponding to a maximum end diastolic pressure after that the flow begins to decrease as a result of limiting ventricular compliance. On the other hand, the increase in stroke volume (cardiac output by default) is also achieved by increasing ventricular contractility, which leads to greater systolic emptying, respectively telesystolic volume reduction (3). Increased systolic parietal tension (wall-stress) is the factor which triggered the process of concentric hypertrophy, while intraventricular end diastolic volume remains unchanged. The

consequence of myocardial hypertrophy is reduced the coronary flow and the hypoxic installation, which leads to the synthesis process development by myocardial anaerobic, while reducing the diastolic (and therefore coronary perfusion), by triggering sinus tachycardia (2, 4).

Ventricular volume overload leads to increased end-diastolic diameter (by myocardial fiber elongation) leading to an increase in stroke volume to a maximum (limit physiological adaptation), after which any further increase in volume causes a reduction in cardiac output (*the law Frank-Starling*). Eccentric hypertrophy adds force of contraction, the participation of all the epicardial muscle layers from endocardium (4).

Therefore, to maintain cardiac output is achieved through the cardiac ventricular concentric hypertrophy (increase in size of mass, volume and surface respectively myocytes but also by adding parallel sarcomeric) and eccentric hypertrophy (cavity dilatation with myocardial sarcomere elongation from 1.5 to 2.3 microns and adding sarcomeric units in series) (2, 3, 4).

These compensatory mechanisms able to increase cardiac output, respectively ventricular performance at a given time is exceeded, by setting up or global left ventricular failure with symptoms caused by the impossibility of ensuring the flow of the body even at rest (11).

The purpose of this study was to identify and characterize structural adaptive changes of heart (heart disease matched) and maladaptive (decompensated heart disease) in mitral valvular disease.

### ***Materials and methods***

The study was conducted during 2010-2011, the Faculty of Veterinary Clinic and veterinarian's office, Doctor's Vet Universe, a total of 20 dogs aged between 7 and 15 years, which were identified by clinical examination, echocardiography, electrocardiograms, mitral valvular cardiac disease and different cardiac arrhythmias.

In these dogs were recorded ECG changes on contractility, excitability and cardiac rhythm were then correlated with their adaptive efficiency. Also, with echocardiographic indicators were established structural and functional changes of the heart, which made the difference between the heart adaptive mode and the critical level of his decompensation. Electrocardiography were performed using an ECG machine type Veterinary VE 100, using the standard bipolar method and working parameters were: speed 25 mm / sec and amplitude 10 mm / mV. The cases of cardiac arrhythmia determination was made following the interpretation of ECG changes reported to regular sinus rhythm characteristics (8). Echocardiogram were achieved with a device type MINDRAY, DP 2200Vet microconvex probe, M and B mode. Following indicators were: shortening fraction (FS%), left ventricular end diastolic diameter (DTDVS), left ventricular diameter telesystolic DTSVS), left ventricular free wall thickness systole and diastole, systolic and diastolic septal thickness, mural thickening rate, general appearance of morphologic cardiac dimensions, valvular and subvalvular endocardium integrity and functionality, parietal endocardium and pericardial effusion presence.

### ***Results and discussions***

The onset of valvular mitral valve with moderate injuries, occurring physiological mechanisms of adaptation of the heart, which retains the functionality of the heart, this compensatory mechanisms produce changes detectable by echocardiography and electrocardiography (1, 10). Eccentric hypertrophy of mitral valve insufficiency ultrasound

differs from concentric hypertrophy (which occurs in aortic stenosis or increased afterload), by the presence of changes in the mitral apparatus, and by gradually increasing the diameter of the left ventricular end diastolic and systolic. Compared with normal echocardiographic appearance of the heart (figure 1), is observed by left ventricular hypertrophy and concentric remodeling (figure 2) and eccentric hypertrophy (figure 3).

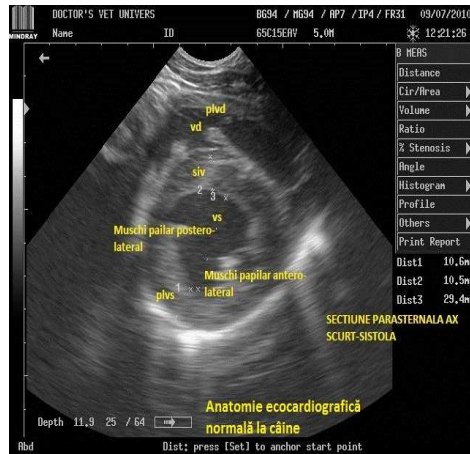


Fig. 1. Normal echocardiographic appearance



Fig. 2. Concentric hypertrophy in dogs



Fig. 3. Eccentric hypertrophy in dogs in chronic mitral insufficiency

In the case of mitral valvulopathy, these structural changes were accompanied by changes of hemodynamic parameters, left ventricular shortening fraction initially increased by 6% of normal (normal 25 - 45%). In the 20 cases of mitral valve insufficiency identified by us were found: the lesions localized degenerative valve apparatus, mitral valve prolapse (septal cusps), left atrial dilatation, left ventricular remodeling (figure 3).

Decreased cardiac output and altered electrical driving intramiocardice triggered increased sympathetic tone and the appearance of ectopic outbreaks. Thus, of the 20 cases of mitral valve insufficiency we investigated, 12 cases were found ECG changes, sinus tachycardia and atrial extrasystoles (figures 4 and 5).



Fig. 4. Sinus tachycardia

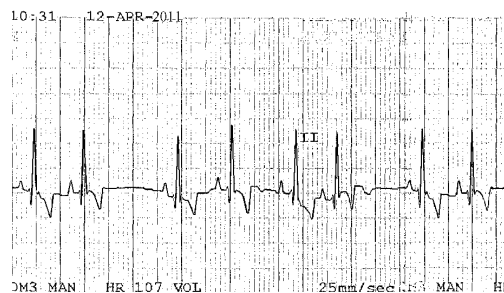


Fig. 5. Atrial extrasystole



If the development of mitral regurgitation, the blood from the ventricle, the high pressure (140 mm Hg systolic pressure), passes into the atrium, low (8 mm Hg systolic pressure). If regurgitated blood volume is low or incrementally over a period of time, then gradually increase atrial compliance, it can get to relax for a greater blood volume with a small increase pressure within the atrium (1, 2, 4). As a result, the adaptive mechanisms developed asymptomatic cardiomegaly (heart disease matched).

Aggravation of degenerative lesions valve apparatus (septal and parietal cusps) and subvalvular (cordage tendon and papillary muscles) leads to an increase in the volume of blood regurgitated with left ventricular volume overload and overcome of atrial distensibility, heart failure by setting up left. Bioelectric speaking, there have been changes in P wave morphology (P mitral), T wave (T high and negative) of the R wave (R □ amplitude 2.5 mV) and ST segment (depression or elevation) (Figure 6). In 40% of investigated cases of cardiac hypertrophy, the ECG changes were accompanied by clinical signs (dyspnea, cough, syncope, exercise intolerance, weakness, and altered heart sounds).

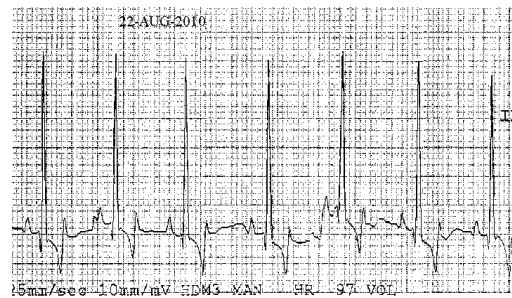


Fig. 6. Cardiac hypertrophy

Complicating mitral valvulopathy with similar lesions in the tricuspid valve (starting as mitral valve damage occurring, but are better tolerated), leads to the onset of global heart failure. Echocardiographic features were: FS% decreased by 10% from normal value, dilatation of four chambers of the heart, pericardial effusion, low systolic thickening. At this stage of cardiac remodeling, chronic valvular disease is distinguished from dilated cardiomyopathy (Figure 7) by the presence of valvular and subvalvular lesions heart device, which mitral regurgitation is produced by the dilation of mitral annulus.

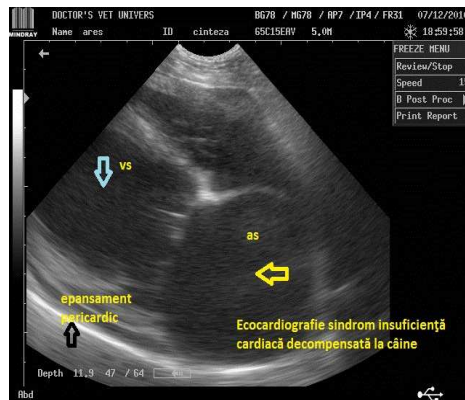


Fig. 7. Dilated cardiomyopathy with congestive heart failure syndrome

Hypertrophy and dilation are the result of adaptive cardiac remodeling. In the short term they are beneficial for the body that improves cardiac output, myocardial function support, and increase cardiac labor.

However, these adaptive mechanisms may be overcome by setting up heart failure, in which case hypertrophy and dilation are pathological significance.

Thus, during concentric myocardial hypertrophy, coronary blood flow is decreased (myocardial hypoxia). The continued growth of intraventricular blood volume (end diastolic volume), will determine the limit of the physiological adaptive dilation (eccentric hypertrophy). The result is decreased cardiac mechanical efficiency, decreased systolic ejection and cardiac output and other compensatory mechanisms that trigger adaptive, that activation of the renin-angiotensin-aldosterone system (RAAS) and release of antidiuretic hormone (5, 9). In the long term and these adaptive mechanisms are overcome by setting up as congestive heart failure (decompensated heart disease).

For the assessment of compensated and decompensated heart failure have made reference to the classifications made by NYHA (New York Heart Association) as (7) and ISACHC (World Small Animal Cardiac Health Council) (6).

### *Conclusions*

1. Hypertrophy and dilation are the result of adaptive cardiac remodeling.
2. In the short term they are beneficial for the body that improves cardiac output, myocardial function support, increase cardiac labor.
3. Exhaustion of these physiological mechanisms, adaptive, leading to installation of decompensation of heart disease and onset of heart failure due to primary lesions and pathophysiological mechanisms (maladaptive).
4. Monitoring series morphological changes, hemodynamic and electrical heart determines the time evolution of heart disease.

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## THE CONSUME OF FOODS WITH HIGH MYCOTOXICOLOGICAL CONTAMINATION RISK AND OCHRATOXIN INCIDENCE ON HUMANS

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### Abstract

Mycotoxins are secondary metabolites of mycotoxigen fungi that affect both human and animal health. The man is exposed through direct consume of vegetal origin products, as well as through animal origin products consumption that can contain mycotoxin residues. Research consisted in the immunoenzymatical mycotoxicological analysis of urine samples gathered from human patients. At the same time, a nutritional enquiry has been done, in order to ascertain the frequency of some aliments consumption at the investigated patients, some of which having a high risk in mycotoxicological contamination.

The obtained results have revealed a daily and weekly consumption of high contamination risk products, like cereals, condiment mixes, tee and coffee.

The results of the mycotoxicological analysis have shown the presence of ochratoxin in 10 out of 20 analysed samples, having values between 201,26 ppt – 643,70 ppt.

**Key words:** nutritional enquiry, human patients' urine, ochratoxin

### Introduction

OTA is suspected to be involved in human nephropathy and in creating tumours at the level of the urinary organs. The level of halving of OTA in blood is high, reaching almost 35 days after a single oral ingestion, due to the unfavourable toxic kinetic elimination [5]. The high level of halving of OTA along with a high frequency of human exposure to food contaminated with OTA, drives to a presence of high levels of OTA in samples of human blood collected from around the world [4] OMS, 2001. Hult and Fuchs (1986) states that the peculiarities of metabolism of this mycotoxin consists in the fact that it is not stored in the body, but numerous studies *in vivo* and *in vitro* showed that it is distributed through blood especially in the kidneys, having an undeniable nephrotoxic effect.

However, so far have not been confirmed through research any clear links between human nephropathy and exposure to OTA by the population after eating (OMS, 2001). Many studies imply a possible Balkan endemic nephropathy in the etiology of OTA (NEB) which is mentioned as pathology in some regions of Bosnia-Herzegovina, Bulgaria, Croatia, Serbia and Romania [3]. Investigations commonly performed worldwide are involving an analysis of OTA in food and biologic fluids, especially plasma in both healthy population and groups of patients with nephropathy. Specialty literature shows conflicting results of these tests. Thus, analysis conducted by Abid et al. (2003) from 1991 to 2000 in Tunisia on a significant number of

volunteers have confirmed the presence of major levels of OTA in plasma at patients with NIC living in similar conditions to those in the Balkans and who consumed food contaminated with OTA [1]. In contrast, Gross et al. (2003) concluded that OTA levels were not very large in the patient's blood hospitalized with kidney problems.

The concept that not just OTA is involved in producing nephropathy, but its association with other factors is confirmed by more recent research. Hassen et al. (2004) have found high levels of OTA in blood and of  $\beta$  2-microglobulin (B2M) at the patients with NIC [2]. Beta 2-microglobulin is a peptide which is a constituent of the human leukocyte antigen complex (HLA), whose values increase in affections where the rate of production or destruction of lymphocytes is increased. Because B2M is excreted at glomerular level and then partially absorbed within renal tubules, by comparing serum levels of this peptide in urine, it is possible to determine the cause of renal dysfunction (glomerular or tubular).

## **Materials and methods**

A nutritional survey model was drawn up which aimed at investigating the alimentation of the target group chosen for study and, in particular, the frequency with which they consume foods with increased mycotoxicological contamination risk. This aimed at obtaining information on the consumption frequency for different types of food, some of which had an increased risk of contamination with mycotoxins. Have been selected five food categories: cereals (corn meal, flour, bread, toast bread, breakfast cereals, biscuits, corn puffs), seeds, nuts (sunflower seeds, pumpkin seeds, peanuts, hazelnuts, walnuts, pistachio), spices (mixture condiments, blend of spices for steaks, paprika, cinnamon, vanilla), beverages (coffee, tea, beer, wine) and animal products (meat, milk, cheese). For all the above food was monitored the frequency of consumption (daily, weekly, monthly, seldom or never) by the investigated subjects.

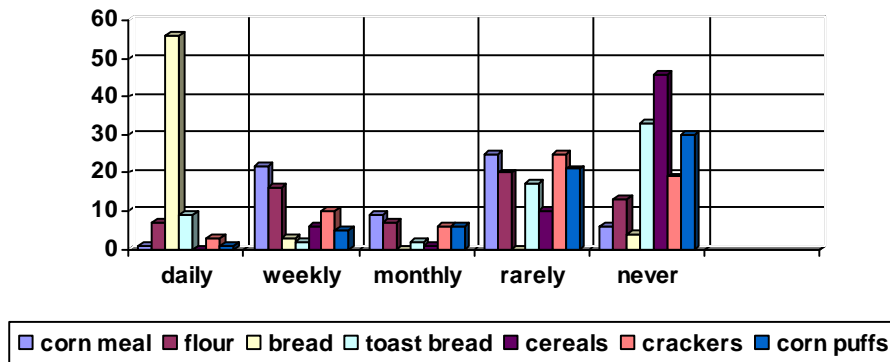
Mycotoxicological analysis of 20 urine samples was performed by the ELISA immunoenzymatic method, effectuated in a human medical clinic. Reading samples was performed using the Dynex apparatus and the analysis of the results using this software.

## **Results and discussion**

The survey was conducted on a group of 63 patients with medical problems in the urinary system. The results are summarized below.

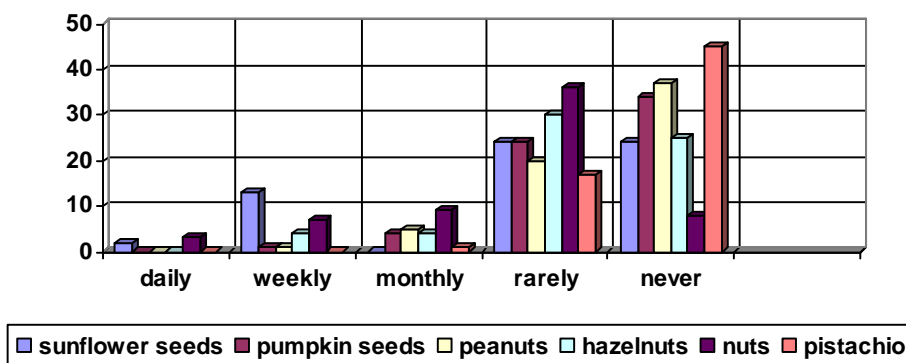
## Summary of the nutritional survey for the investigated group

### 1. Cereals



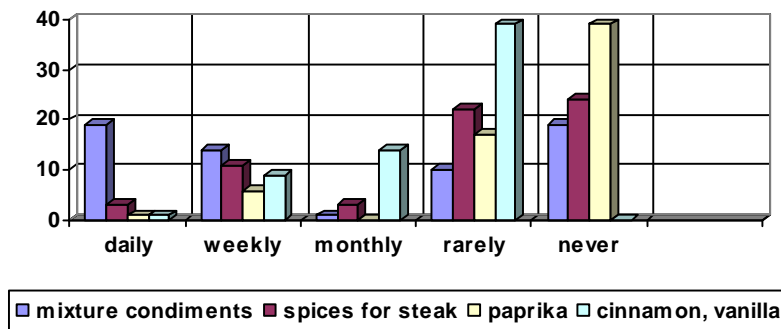
The data and figure above shows that the highest frequency of the daily consumption was at the white bread followed by toast bread and flour. More than half of these patients rarely or never consume these products (the conclusion is justified due to the pathological conditions).

### 2. Seed, nuts



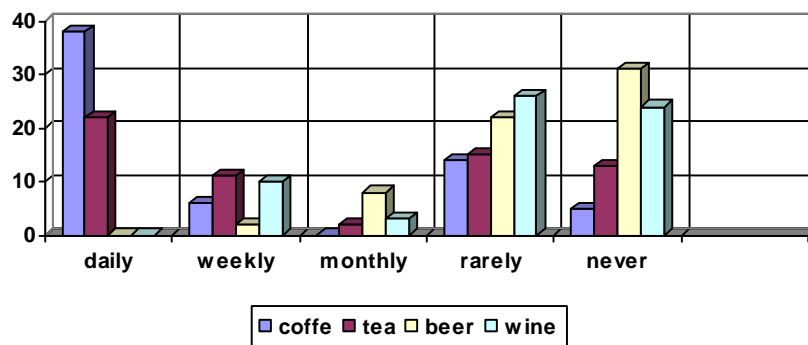
In terms of consumption of seeds and nuts (food with increased risk of contamination by mycotoxins) is observed a low rate of consumption at the majority of patients investigated. Of these, sunflower and pumpkin seeds are the most commonly consumed.

### 3. Condiments



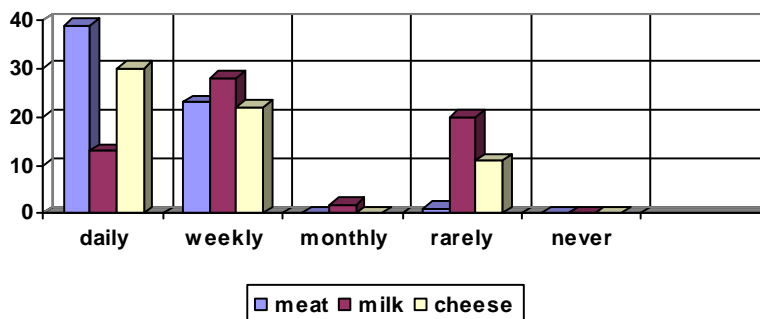
The data presented shows that although most patients consume rarely or not at all these foods, there are people who use daily vegetable mixtures of condiments, foods also with increased risk of contamination.

### 4. Drinks



It is observed that also on these types of foods there is an increased daily consumption of coffee and tea at more than half of the investigated patients, as where as a low or nothing at all at the rest of the patients.

### 5. Animal products



These foods are consumed on a daily or weekly basis by the most of the investigated patients. The risk of contamination with mycotoxins is relatively small for this product given that they are well controlled Sanitary-Veterinary, stored in refrigerated conditions and sold relatively rapid from the date of manufacture.

Mycotoxicological analysis of 20 urine samples was performed using the immunoenzymatic method ELISA, realised in a human medical clinic.

Reading the samples has been done using the Dynex device (Photo 1) and the interpretation was realized using the RIDAWIN/FOOD/211010.MET software.



Photo 1. Analysing samples with Dynex

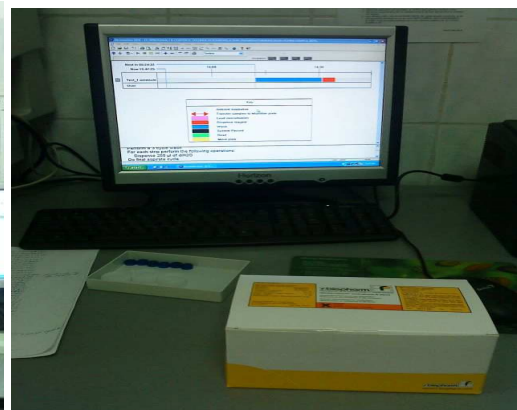


Photo 2. Analysis kit R-Biopharm, Diamedix Company

Urine samples have been directly analyzed without prior processing, as specified by the analysis kit (R-Biopharm) for ochratoxin, provided by the Company Diamedix (Photo 2).

The results obtained in the analysis have been positive for 10 out of 20 samples analysed, having the following values: 315.76 ppt, 205.86 ppt, 644.42 ppt, 244.82 ppt, 553.31 ppt, 451.88 ppt, 206.52 ppt, 498.64 ppt, 240.38 ppt and 201.26 ppt, at the rest of the analysed samples there are negative results.

### **Conclusions**

In terms of the daily and weekly consumption of the products with a high risk of contamination is high for cereals, spice mixture of condiments, tea and coffee.

The incidence of ochratoxin in the analysed urine samples was 50%, having a range of value between 201.26 ppt and 643.70 ppt.

Frequent consumption of foods with high risk of contamination by mycotoxins could be a predisposing factor in the production of mycotoxicosis at the human population.

### **Acknowledgement**

This work was cofinanced from the European Social Fund through Sectoral Operational Programme Human Resources Development 2007-2013, project number POSDRU/89/1.5/S/63258 "Postdoctoral school for zootechnical biodiversity and food biotechnology based on the eco-economy and the bio-economy required by eco-san-genesys"

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## COPPER CONCENTRATION IN THE CATTLE'S HAIR – REFERENCE VALUES AND NORMS FOR DIAGNOSIS

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### *Abstract*

*Epidemiological, clinical and drawing of the cattle's hair samples from 3 farms were performed: farm B (standard )with optimum production, reproduction and sanitary veterinary parameters, farm C (nonpolluted area), farm Z (industrial polluted area, especially with heavy metals). In these 3 farms 70 cattle's hair samples were drawn and tested in order to establish the reference values of the copper (Cu), the influence of some factors – season, physiological statement of the cows, effects of the pollution with heavy metals on the hair's copper concentration. Reference values established for hair's copper varied between 10-18 ppm. Hair's copper concentration was significantly reduced in grazing period (June) versus stabling period (March), respectively  $8,21 \pm 1,04$  ppm versus  $10,79 \pm 1,68$  ppm ( $p < 0,005$ ). Hair's copper values were not influenced by the physiological statement of the cows. Copper concentration was significantly reduced in the cattle's hair from an industrial polluted area, especially with heavy metals, respectively  $5,45 \pm 1,41$  ppm (farm Z) and  $10,84 \pm 1,45$  ppm (farm C) ( $p < 0,001$ ). This indicate secondary hypocoprosis by copper interference with some polluted heavy metals. The study demonstrate that copper determination in the cattle's hair may be a useful marker for the evaluation of copper statement and for hypocoprosis or hypercoprosis diagnosis of the cattle.*

**Key words:** *cattle, hair, copper*

### *Introduction*

Diagnosis of dismineralosis in cattle can be realised by complex paraclinical methods (4, 6, 7, 8, 9, 11). Primary or secondary hypocoprosis is frequently found to the cows and calves having usually subclinical evolution; it is necessary to use multiple and complex norms for diagnosis. One of the method for diagnosis also simple and noninvasive, is determination of copper statement in the cows hair (2, 5, 10). The hair is able to reflect real content of the minerals transported by the plasma. Anke (1) printed out the correlation between the content of some minerals in fodder and in animal's hair respectively; he proposed „hair test” to be used in order to diagnosis mineral deficiency in the cattle. It was also demonstrated that the statement of minerals in the hair may be a good indicator for the lacting cows supply with some mineral constituents (I, Fe, Mn, Zn) (13). This paperwork was intended to pointed out reference values of the copper in the cow's hair, the influence of some factors (season, physiological statement,

industrial pollution of the ecosystem) on the cattle's copper hair in order to establish some norms for the diagnosis of hypocprosis (primary or secondary) or hypercprosis.

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### ***Materials and methods***

The study was performed in 3 farms of cattle: *farm B* (standard) with optimum production, reproduction and sanitary-veterinary parameters, *farm C* (found in a nonpolluted industrial area but with some problems of reproduction and sanitary-veterinary) and *farm Z*, found in an industrial polluted area with important implicates on production, reproduction and health of the cows and calves. 70 hair's samples were drawn from the cows: 10 in the farm B, 50 in the farm C and 10 in the farm Z. Drawing, conditioning and processing of the samples were performed after an officialised methodology for sanitary-veterinary laboratories. Copper was determined by a colourimetric method with natrium dietilditiocarbamat.

### ***Results and discussions***

Results were centralized and statistically procesed by the test t-student in the tables 1-3. In order to established the reference values for hair's copper, in the farm B (standard) was randomly constitutes a group of 10 cows (5 with advanced pregnancy and 5 in an optimum lactation period). The values of hair's copper varied between 10 and 18 ppm; no important diferences were found between the two groups of cows. The results obtained bordered in reference values interval which reflects a normal statement of the copper in the body. The reference values recorded are in general comparable with those recorded by others authors in the cattle's hair (1, 2, 5, 6). Registering of the hair samples with low values of copper, under minimal limits of 10 ppm, may be considered as marker for a copper deficiency statement and increased values of hair's copper over 18 ppm may indicate hypercprosis (eventually copper intoxication).

In the tables 1 and 2 were centralised copper's values in the cattle's hair according to the season (table 1), respectively physiological statement of the cows (table 2) from the farm C.

Table 1

**Values of the hair's copper from the lacting cows according to the season**

<b>Period</b>	<b>Group</b>	<b>Farm</b>	<b>n</b>	<b>Cu (ppm) X ± s</b>
<b>Stabling (March)</b>	<b>1</b>	<b>C</b>	<b>10</b>	<b>10,79 ± 1,68</b>
<b>Grazing (June)</b>	<b>2</b>	<b>C</b>	<b>10</b>	<b>8,21 ± 1,04</b>
<b>T test</b>				<b>p &lt; 0,005</b>

Table 2

**Values of the hair's copper from the cows according to the physiological statement**

Physiological statement	Group	Farm	N	Cu (ppm) $\bar{X} \pm s$
Nonpregnant cows	1	C	10	13,10 $\pm$ 4,77
Advanced pregnant cows	2	C	10	10,30 $\pm$ 1, 61
Lacting cows	3	C	10	10,84 $\pm$ 1,45
T test p	a $\leftrightarrow$ b a $\leftrightarrow$ c b $\leftrightarrow$ c			p < 0,1 (NS) p > 0,2 (NS) p < 0,5 (NS)

In the group was recorded a hair's copper value significantly decreased versus the group 1 – respectively 8,21  $\pm$ 1,04 ppm versus 10,79  $\pm$ 1,68 ppm ( $p < 0,005$ ). The results recorded may explain a hypocprosis by fooder deficiency in the copper in the pasture period (table 1).

Results centralised in the table 2 demonstrates that physiological statement of the cows does not influence significantly values of the hair's copper.

It was noted that physiological statement of the cows obviously influences the level of calcium, phosphorus and magnesium in the hair; the lower values was recorded from these elements in the hair samples from lacting cows compared to the pregnant ones because these minerals, calcium particularly, are consumed in large amounts during lactation and may disturb mineral homeostasis (12). These aspects did not observed in the case of copper in our study.

Table 3

**Values of the hair's copper from the lacting cows in an industrial polluted area (farm Z) comparatively to anonpolluted one (farm C)**

Farm	Group	Area	n	Cu (ppm) $\bar{X} \pm s$
Z	1	Industrial polluted	10	5,45 $\pm$ 1,41
C	2	nonpolluted	10	10,84 $\pm$ 1,45
T test p				p < 0,001

The farm Z is found in an industrial polluted area with emission of residues of sulphides, molybdenum, lead, cadmium, chromium and so on These residues polluted air, water and fooder plant which disturbed the ecosystem in the area.

In the group 1 of cows values of hair's copper was significantly lower versus the control one (group 2) – 5,45  $\pm$  1,41 ppm and 10,84  $\pm$  1,45 ppm respectively ( $p < 0,001$ ). The results demonstrate a secondary hypocprosis in the cows following of copper interferences with polluted residues in the area.

The study demonstrated that copper determination in the cattle's hair represent an usefull marker for the copper statement evaluation and for the diagnosis of hypocopprosis or hypercopprosis in the cattle.

### *Conclusions*

1. Reference values of the cows hair for copper were established on the standard cows farm as beeing 10-18 ppm.
2. In grazing period the values of the cows hair copper were significantly lower than the values in stabling period, respectively  $8,21 \pm 1,04$  ppm and  $10,79 \pm 1,68$  ppm ( $p < 0,005$ ); that demonstrates a primary hypocopprosis.
3. The values of the cows hair copper were not influenced by the physiological statement of the cows, respectively advanced pregnancy, nonpregnancy and lactation.
4. The values of copper in the hair were significantly lower to the lacting cows found in an industrial polluted area versus a nonpolluted one:  $5,45 \pm 1,41$  ppm and  $10,84 \pm 1,45$  ppm respectively ( $p < 0,001$ ); that demonstrate a secondary hypocopprosis by interferences of copper with polluted residues in the area.
5. Determination of copper in the cows hair represent an useful marker for the copper statement evaluation and for diagnosis of hypocopprosis or hypercopprosis in the cattle.

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## INFLUENCE OF HEAT STRESS ON THE PRODUCTIVE PERFORMANCE OF MANGALITSA PIGS

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**Abstract.** The study aimed to establish the influence of high temperatures on the productive performance of Mangalitsa pigs raised in the alternative system and exposed to 30-36<sup>0</sup>C. The experimental period was between 86 and 100 kg liveweight. The animals had free access to standard, isoprotein and isocalory diets, with 13.5% crude protein (CP) and 3100 kcal/kg metabolizable energy. Feed intake was measured on a daily basis. The energy-protein balance was calculated on the basis of comparative slaughter made at the beginning and end of the experiment. The metabolizable energy (MEc) was estimated by chemical analysis (feed and excreta) using mathematical modelling and the Whittemore's formula. The metabolizable energy utilization efficiency was 0.61 at Large White and 0.53 at Mangalitsa.

**Key-Words:** Mangalitsa, energy metabolism

### *Introduction*

Mangalitsa is a breed with a valuable genetic background which can be used in the current context of organic livestock, considering the natural resistance and adaptation to growing conditions in intensive, extensive and alternatives systems.

Mangalitsa is one of the most popular breeds of pigs in Europe, because meat has superior properties, such as taste, marbling and low cholesterol content. Many Americans Farmers have imported the Mangalitsa breed and the technology of its raising.

The efficiency of nutrients use is a major step to develop strategies for profitable growth of farm animals; the determination of energy metabolism is a relevant stage in assessing the efficiency of feed conversion to animal production.

In particular context of ecophysiology, the environmental factors have major influence on productive performances, energy metabolism and feed use efficiency. For example, exposure of Large White fattening pigs at heat stress by high temperature lowers the average daily gain with 23% [5]. The negative implications of long-term exposure are often obvious in the critical production stages, as they are threatening and negatively impact the animal's welfare [1,2]. The climate conditions have a major role in expressing the genetic potential of races and the metabolizable energy utilization efficiency [6]. To assess the changes caused by environmental factors, it is necessary to know the normal aspects of energy metabolism in fattening pigs, depending on race.

To ensure a sustainable economy, this must become an eco-bio-economy, environmentally friendly and based on a livestock biodiversity [3].

The studies aimed the determination of energy metabolism in Mangalitsa pigs exposed at thermic neutral temperature, compared with Large White pigs.

## *Materials and Methods*

The experiments were conducted on each of 22 Mangalitsa pigs, raised in intensive (G1) and alternative system (G2). The initial average weight was 86.6 kg. The group G1, the animals were housed in air-controlled rooms and exposed at  $20^{\circ}\text{C} \pm 1^{\circ}\text{C}$ . The relative humidity was 72%. The group G2, the animals were housed in alternative system and exposed at  $30\text{-}36^{\circ}\text{C}$ . The stocking density was identical in the two farming systems.

The experiment ended when the animals reached 100 kg body weight.

The animals had free access to water and to standard, isoprotein and isocalory diets, with 13.5% crude protein (CP), 4.9% lysine and 3100 kcal/kg metabolizable energy. Feed intake was measured on a daily basis.

Feed, excreta and biological samples were analyzed according the Weende scheme. The crude protein was determined by **Tecator – Kyeltec Auto Analyze**. The ether extract was determined by **Soxtec System HT**, starch by the polarimetric method and sugar by Bertrand's method.

The digestibility coefficients for the nutritive substances were established as a result of 3 specific experiences, seven days each.

The gross energy was calculated using standard equations. The digestible energy and digestible protein were determined by the specific experiments, when there recorded ingesta and excreta. The energetic and protein balance were calculated using the method of comparative slaughtering and mathematical modelling. The metabolizable energy (MEc) was calculated using Whittemore's formula [7]:

$$\text{MEc} = \text{DE} - (\text{Eu} + \text{dE} + 6.8 \text{ BFM} + 1.4 \text{ S})$$

where:

MEc = corrected metabolizable energy

DE = digestible energy

Eu = energy urine = 7.2 DP

DP = deaminated protein, g

dE = deaminated energy = 4.9 (DP – Pm)

Pm = protein for maintenance, g (estimated by mathematical modeling)

BFM = bacterial fermentable matter, g = (DCF + NDS) – (St + S)

DCF = digestible crude fiber, g

NDS = non-nitrate digestible substances, g

St = starch, g

S = sugar, g

The productive performances data were ANOVA statistically processed.

## *Resultats and Discussions*

The productive performances are presented in Table 1.

Table 1. The productive performances of pigs

Specification	G1	G2
Initial weight, kg	86.4	86.7
Final live weight, kg	101.5	100.6



Experimental period, d	34	43
Average daily gain, g	442.3	324.1
Feed intake, kg	4.32	3.57
Warm yield, %	78.3	78.1
Cold yield, %	75.4	75.1
Fat thickness, mm	36.3	27.7

At 20°C, in intensive system, 100 kg weight was achieved after 34 days, while at 30-36°C, in alternative system, this weight was obtained after 43 days (Table 1). The differences were significant ( $p \leq 0.05$ ). The daily exposure to heat stress increased the fattening period with 9 days.

The average daily weight gain was 442.3 g at G1 and 324.1 g at G2. The average daily weight gain decreased with 26.7% in group 2 compared to group 1 ( $p \leq 0.05$ ). The differences were significant ( $p \leq 0.05$ ).

The feed intake was 4.32 kg at G1 and 3.57 kg at G2, with 17.3% less. The differences were significant ( $p \leq 0.05$ ).

The warm slaughtering yield was 78.3% (G1) and 78.1% (G2), while the cold slaughtering yield was 75.4% and 75.1%, respectively. The exposure to heat stress did not influence the slaughtering yield, the average values being statistically similar.

The fat thickness was 36.3 mm at G1 and 27.7 mm at G2. The exposure to heat stress induced a 23.7% depression of this parameter if compared to group 1.

The following coefficients resulted from the digestibility experiments: organic matter (OM) 87.34% (G1) and 87.10% (G2); crude protein (CP – apparent digestibility) 89.25% (G1) and 89.96% (G2); energy 86.35% (G1) and 86.57% (G2). The exposure to heat stress did not influence significantly diet digestibility in neither of the two groups. This results are consistent with those presented in literature [4].

The energy balance (Table 2) shows values of 1025 kJ/G<sup>0.75</sup> corrected metabolisable energy (MEc), 635 kJ/G<sup>0.75</sup> metabolisable energy for production (MEp) and 62% efficiency of MEc utilisation as MEp for group 1. The corresponding values for group 2 were 945 kJ/G<sup>0.75</sup> MEc, 520 kJ/G<sup>0.75</sup> MEp and 55% efficiency of MEc utilisation as MEp.

The energy retained as gain (RE) was 16.7 kJ/G<sup>0.75</sup>, of which 8 kJ/G<sup>0.75</sup> in protein (PEr). The corresponding values for group 2 were 13.6 kJ/G<sup>0.75</sup> RE, of which 7 kJ/G<sup>0.75</sup> PEr. The efficiency of MEc utilisation as PEr was 0.78% in-group 1 and 0.74% in-group 2 ( $p \leq 0.05$ ). The efficiency of MEc utilization as LEp (energy retained in fat) was 0.85% in-group 1 and 0.70% in-group 2, with 17.5% lower ( $p \leq 0.05$ ).

Table 2. Energy balance (by G<sup>0.75</sup>)

Specification	L1	L2
MEc, kJ	1025	945
MEp, kJ	635	520
RE, kJ	16.7	13.6
PEr, kJ	8.0	7.0
LEr, KJ	8.7	6.6
ME p/MEc	62	55

PEr/ MEc	0.78	0.74
LEr/ MEc	0.85	0.70

### ***Conclusions***

The productive performance decreased significantly in pigs exposed to heat stress.

The efficiency of metabolisable energy utilization as energy retained in fat decreased by 17.5% in the animals exposed to heat stress if compared to exposure to 20°C.

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### ***Acknowledgement***

This work was cofinanced from the European Social Fund through Sectoral Operational Programme Human Resources Development 2007-2013, project number POSDRU/89/1.5/S/63258 "Postdoctoral school for zootechnical biodiversity and food biotechnology based on the eco-economy and the bio-economy required by eco-san-genesys"

# EVIDENCE HISTOCHEMICAL CELLS ENTEROCROMAFINE OF THE SMALL INTESTINE FROM CHICKENS (GALLUS DOMESTICUS)

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## ABSTRACT

*Diffuse neuroendocrine system cells belong enterocromafine system, including widespread cell types in different organs of the digestive, respiratory, urinary, etc..*

*Secretory cells are cells enterocromafine containing chemical mediators stored in granules into the cytoplasm. Besides the capacity to store biogenic amines, enterocromafine cells develop local acting hormones polypeptide.*

*Research has sought of recognizing the description of a histochemical techniques enterocromafine cells, their morphology and their location in the histological structures of the small intestine to elucidate their functions.*

*Intestine samples collected were fixed in Bouin fluid and processed after several attempts by reaction with silver-hexametilentetraamina Lillie-Burtner version.*

*In the small intestine, cells are located at the base enterocromafine intestinal villous, the crypts of the villi separated, but in depth in the epithelium lining the glands secreting Lieberkühnn until the muscle lining.*

*Besides components of intestinal epithelium cells (enterocytes, goblet mucous cells, stem cells, etc.) were identified with the prism-shaped cells enterocromafine black granules arranged Intracytoplasmic brown, who come into contact with the basal membrane but do not reach the apical pole in the lumen organ.*

**Keywords:** *enterocromafine cells, histochemical reaction, small intestine.*

## INTRODUCTION

Enterocromafine cells are cells of digestive tube neuroendocrine system belonging to diffuse (APUD system). APUD system (Amine Precursor Uptake and Decarboxylation) cell types including nerve cells and endocrine features, from different organs of the digestive, respiratory, urinary, etc capable of producing peptides with capacity of decarboxylation (1, 6).

Experimental embryology research showed ectodermal embryological origin of these cells, they derive from neural crests cells or neural placode localized in mesenchymal or primitive digestive tract epithelium (3).

The existence of a particular subpopulation of cells, distinct from other parts of the gastrointestinal tract, was mentioned as early as 1870 by Heindenheinn. These cells were subsequently called cells or *cells Kultchitsky enterocromafine*.

In 1907 Oberndorfer showed granulations in the cytoplasm of chromaffin cells enterocromafine. In 1924, Masson made the first complete description of histo-and histogenetics enterocromafine digestive cell, which is called „*cell-Masson Kulchitsky*”. Its main feature is the presence of cytoplasmic grains, which can reduce ammoniacal silver nitrate (6).

In 1953, Lembeck extract from cells enterocromafine serotonin and serotonin metabolism. The biochemical results and highlights 5-hydroxyindoleacetic acid. Enterocromafine cells have the capacity to produce serotonin (5-hydroxytryptophan), which is metabolized in the liver to *5-hydroxy-acid-indolacetic* which is eliminated urinary and he can be dosed. These cells produce *specific neuron usually enolaza (NSE)* and *cromogranina A (CgA)*, both of these can be used as tumor markers, if hyperplastic proliferation (carcinoid tumors) (2).

APUD system cells are called, „clear cell”, name given because of their histological behavior (topographic colorations are ineffective, so the cells are stained with silver salts are called, „argirofile”). Since affinity for silver salts they were divided into different cells and cell argentafine argirofile or „enterocromaphin like” (4, 5).

Argentafine cells detain ammoniacal silver salts in secretory granules while the presence of reducing substances argirofile requires (4, 5).

## MATERIAL AND METHOD

The purpose of the research carried out aimed at highlighting enterocromafine cells by histochemical technique, their location and their morphology in histological structures of the small intestine to elucidate their functions.

Histological parts of small intestine taken from slaughtered poultry (*Gallus domesticus*) were fixed in Bouin fluid being further processed for paraffin inclusion. Paraffin blocks were sectioned at 6 microns and colored by reaction with silver version hexametilentetraamina Lillie-Burtner, interpreted and microphotography.

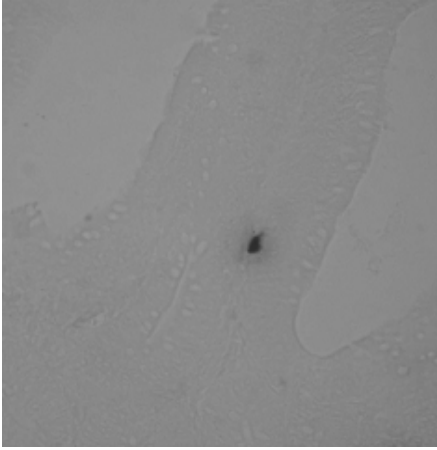
## RESULTS AND DISCUSSION

Histological structure of the small intestine in *Gallus domesticus* is similar to that of mammals, consisting of the 4 characteristic tunics: mucosa, submucosa, muscular and serous.

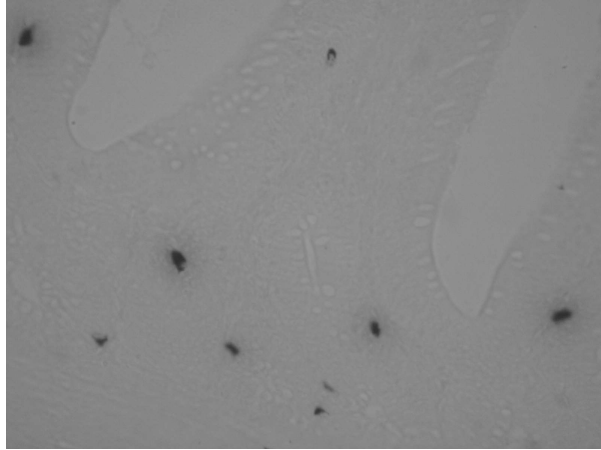
Intestinal villi are scattered all over the intestine, intestinal epithelium is simple prismatic, consisting of: primary cells (enterocytes), goblet mucous cells, undifferentiated stem cells and cells enterocromafine. In the intestinal villous epithelial cells are plentiful, which shows an intense mitotic activity of undifferentiated stem cells. Location is observed in the cells enterocromafine intestine crypts on intestinal epithelial basal membrane cells among absorptive, goblet and undifferentiated stem (fig. 1).

**Lieberkühn glands are well developed and branched into the small intestine. They have a simple tubular structure, sinuous and open the crypts of intestinal villi at the base.**

**In the intestinal mucosa chorion cells are arranged enterocromafine disseminated intestinal glands of the basal membrane (Lieberkühnn) (fig. 2).**



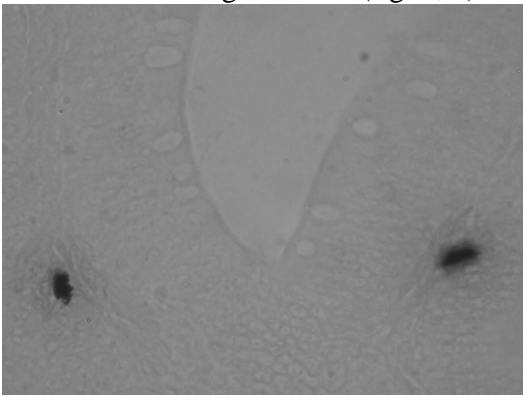
**Fig. 1 – Location enterocromafine cells in the lining of intestinal reaction with silver – hexametilen tetraamina; Ob. 40x**



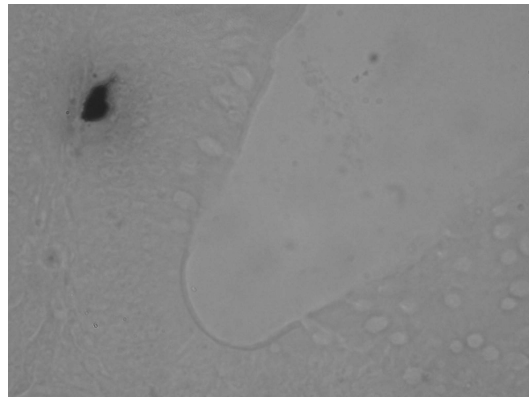
**Fig. 2 – Location chorionic cells in the intestinal mucosa enterocromafine; reaction with silver-hexametilentetraamina; Ob. 40x**

Enterocromafine cell morphology outlined by reaction with silver histochemical hexametilentetraamină prismatic or pyramidal in shape, with intracytoplasmic secretory granules blackish brown color, a colorless or yellowish background tissue.

The small intestine crypts enterocromafine cells are arranged on the basal membrane and not in contact with organ lumen (fig. 3, 4).

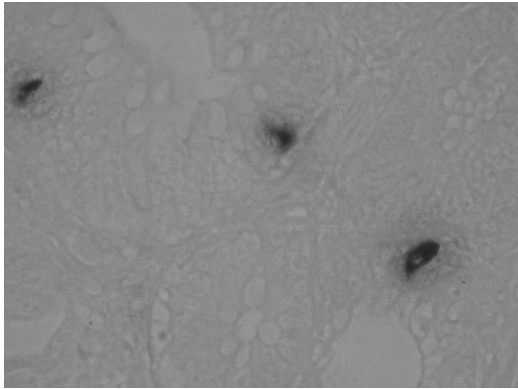


**Fig. 3 – Enterocromafine cell morphology in the lining of intestinal reaction with silver-hexametilen tetraamina; Ob. 100x**

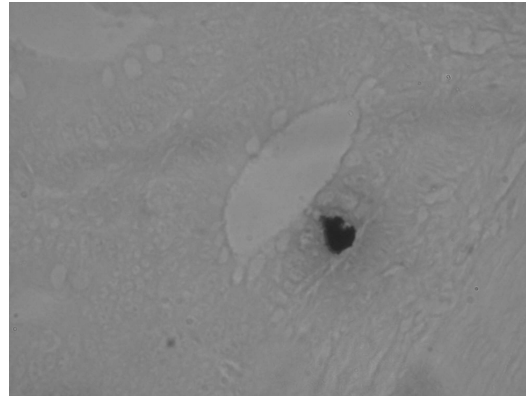


**Fig. 4 – Enterocromafine cell morphology in the lining of intestinal reaction with silver-hexametilentetraamina; Ob. 100x**

The intestinal glands (Lieberkühnn) there are two types of cells: some are in contact with surface epithelium apical pole and the second type are the cells that do not come in contact with the organ lumen, and the product of secretion is eliminated by basal membranes (fig. 5, 6).



**Fig. 5 – Enterocromafine cell morphology intestinal glands located in the structure, reaction with silver-hexametilen tetraamina; Ob. 100x**



**Fig. 6 – Enterocromafine cell morphology intestinal glands located in the structure, reaction with silver-hexametilentetraamina; Ob. 100x**

## CONCLUSIONS

1. By reaction with silver hexametilentetraamină Lillie-Burtner version, were found in the small intestine in *Gallus domesticus* enterocromafine cells, integrated chemical mediators and neuroendocrine by neurotransmițătorii whom develop to influence the synthesis of exocrine cells of the digestive tract and intestinal smooth muscle.

2. Enterocromafine cells are prism forms or pyramidal cells in contact with basal membrane, scattered among enterocytes in the intestinal epithelium or intestinal glands including secretory cells of chorionic intestinal mucosa.

3. The intestinal epithelium is a single cell type, located on the basal membrane not in contact with the organ lumen.

4. The intestinal glands are two types of cells: some are in contact with the surface epithelium and the second type, which do not come in contact with body lumen. Both types of cells removed by biogenic amines and neurotransmitters basal membranes or intercellular communication proteins located on the sides of the cells produced in the capillary or directly reaching the target cells nearby.

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# MYCOTOXICOLOGICAL SCREENING OF FEED USED IN DIARY CATTLES NUTRITION FROM INDIVIDUAL FARMS

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## **Abstract**

*Mycotoxins are fungi secondary metabolites which affect health of all animal species.*

*The research consisted in the investigation by means of a mycotoxicological exam of fooder (straw, alfalfa) administered to cattles exploited household system, in order to determine the contamination degree with some mycotoxins.*

*Have been collected and analyzed a total of 16 feed samples from eight households from Bucharest, resulting in a total of 32 final determinations. The mycotoxicological analysis was performed by means of the immunoenzymatic test ELISA for the determination of the mycotoxins aflatoxin and ochratoxin A.*

*Of the 32 samples taken in preparation, 25 samples were contaminated with OTA and AF, and 7 samples were lacking, according mycotoxicological examination, contamination with fungi that produce OTA or AF. So 78.12% of the total samples were positive mycotoxicological exam subject for the two mycotoxins.*

*Depending on forage harvesters, it was found that all three types of contaminated feed, regardless of their origin, but different values recorded in the OTA and AF concentration.*

**Key words:** *fooder, cattles, aflatoxin B1, ochratoxin A*

## **Introduction**

Mycotoxins are fungi secondary metabolites, extremely aggressive for humans and animals health. Fungi can develop in the substratum represented by the fodders used as ingredients, especially cereal grains, or in the product itself, during the manufacture process, during shipping or in the storage period.

The research consisted in the investigation by means of a mycotoxicological exam of fooder (straw, alfalfa) administered to cattles exploited household system, in order to determine the contamination degree with some mycotoxins. Mycological and mycotoxicological examinations were



performed in laboratory for determining concentrations of the two most common mycotoxins, aflatoxins and ochratoxins.

Aflatoxins (AFB1, AFB 2, AFG1,AFG2) are mycotoxins produced in nature by species of mychetes from the geneders *Aspergillus* (*A. flavus*, *A. parasiticus*) and rarely by some species from the geneder *Penicillium* (*P.variable*, *P. puberulum* etc) and *Rhizopus spp.*

Ochratoxins represent a group of compounds (A, B,C,D) in whose chemical structure L – fenilalanine is combined by means of amidical bond with an isocumarinic derivate. The production of these ochratoxins is encouraged by the presence of oligoelements in the environment, a temperature of 20-20 °C and humidity of 18-20% at wheat (2).

### **Material and methods**

Have been collected and analyzed a total of 16 feed samples (straw, alfalfa and mixed fodder) from eight households from Bucharest, resulting in a total of 32 final determinations. The mycotoxicological analysis was performed by means of the immunoenzymatic test ELISA for the determination of the mycotoxins aflatoxin and ochratoxin A.

5 g were taken from each analysed assay witch was processed by grinding: these 5 g were extracted with 25 ml of methanol 25%; the obtained essence was filtered using filter paper. Standards, assay essence and the mycotoxins combined with the enzyme werw mixed together and then were added in the reservoirs coated with antibodies. After wash, the enzymatic substratum was added, the intensity of the blue colour thus obtained in inverse ratio to the concentration of the mycotoxins from the assay or from the standard. After adding the stopping solution, blue turned into yellow, the intensity of colour was measured by spectrophotometricals means using a microplates reader having a filter of 450 nm.

The optical densities (DO) of the assays were compared to those the standards, thus determining the concentrations of the assays.

### **Results and discussions**

After analyzing 32 of fodder administered to cattles (straw, alfalfa and mixed fodder), respectively 16 samples of food duplicate samples, aflatoxin and ochratoxin A (OTA) were identified as shown below.

#### **1. Results obtained from analysis of aflatoxin in straw, alfalfa and mixed fodder**

After the analysis of the fodder administered to cattles in order to identify AF, the following results were obtained, as shwon in table 1 and chart 1.

Table 1

Results obtained at the analysis of mycotoxin AF

No. assay	Furage	Source (household)	DO	Concentration AF (ppb)
1.	Straw	G1	0.376	15 ppb
2.	Alfalfa		0.256	35 ppb
3.	Alfalfa	G 2	0.321	15 ppb
4.	Straw		0.424	5 ppb
5.	Straw	G 3	0.460	5 ppb
6.	Alfalfa		0.459	5 ppb
7.	Alfalfa	G 4	0.277	35 ppb
8.	Straw		0.273	35 ppb
9.	Fodder	G 5	0.387	15 ppb
10.	Alfalfa		0.380	15 ppb
11.	Fodder	G 6	0.498	5 ppb
12.	Alfalfa		0.521	5 ppb
13.	Straw	G 7	0.576	1.7 ppb
14.	Alfalfa		0.550	1.7 ppb
15.	Alfalfa	G 8	0.820	0 ppb
16.	Straw		0.620	0 ppb

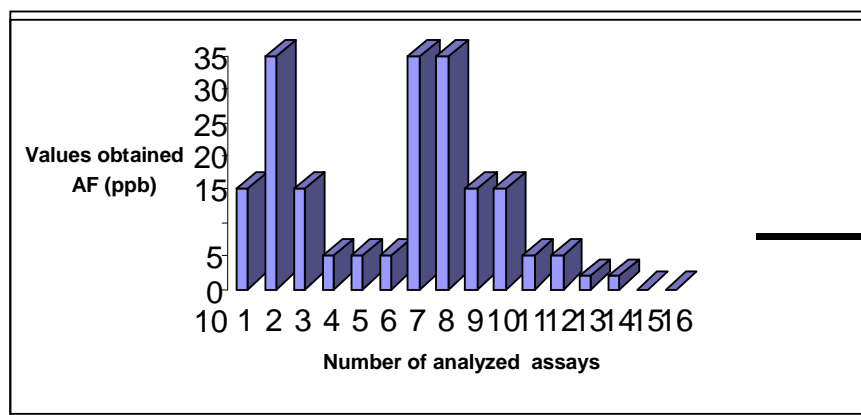


Chart 1. The representation of the contamination degree with AF of the analyzed assays

- From the 16 assays analyzed for aflatoxin, in 12 analyzed assays the results were over maximum limit (4 ppb), with variation contained in 5-35 ppb representing a percentage of 75%.

- In the 4 assays analyzed the results was under detection limit for aflatoxin (0-1.7 ppb). In this case a percentage of 25% assays analyzed was contaminated with species of mychetes which producing aflatoxins.
- According to the mycotoxicological analysis, found that 14 assays analyzed were contaminated with fungi that could cause bovine aflatoxicosis (a percentage of 87.50%).

Aflatoxicosis is a disease caused by consumption of aflatoxins, wich are metabolized in ruminants by the liver and excreted in the bile. Aflatoxin B<sub>1</sub> is the most potent mycotoxin (toxic substance produced by a mold). Aflatoxin B<sub>1</sub> increases the apparent protein requirement of cattle and is a potent cancer causing agent (carcinogen). When significant amounts of aflatoxin B<sub>1</sub> are consumed, the metabolite M<sub>1</sub> appears in the milk within 12 hours. Research suggests M<sub>1</sub> is not as carcinogenic or mutagenic as is B<sub>1</sub>, but it does appear to be as toxic as its parent compound.

The recommended aflatoxin feeding level is 0 parts per billion (ppb). The level of aflatoxin an animal can tolerate depends on age and sex of the animal, its health status, and overall management on the farm. To avoid contamination of milk, do not exceed 20 ppb aflatoxin in the total ration of lactating cows (1).

## 2. The analysis of OTA in the assays of straw, alfalfa and mixed fodder

The analysis of the ochratoxin A in the assays, reveald its presence in varied quantities as they shwon in table 2 and chart 2.

Table 2

Results obtained at the analysis of mycotoxin OTA

<b>Nr. Sample</b>	<b>Type of forage</b>	<b>Origin (household)</b>	<b>DO</b>	<b>Concentration OTA (ppb)</b>
<b>1.</b>	Straw	<b>G1</b>	<b>1.389</b>	<b>0 ppb</b>
<b>2.</b>	Alfalfa		<b>1.258</b>	<b>10 pp</b>
<b>3.</b>	Alfalfa	<b>G 2</b>	<b>1.268</b>	<b>10 ppb</b>
<b>4.</b>	Straw		<b>1.377</b>	<b>0 ppb</b>
<b>5.</b>	Straw	<b>G 3</b>	<b>1.292</b>	<b>5 ppb</b>
<b>6.</b>	Alfalfa		<b>1.110</b>	<b>10 ppb</b>

7.	Alfalfa	<b>G 4</b>	<b>0.756</b>	<b>20 ppb</b>
8.	Straw		<b>1.021</b>	<b>15 ppb</b>
9.	Fodder	<b>G 5</b>	<b>1.271</b>	<b>5 ppb</b>
10.	Alfalfa		<b>1.218</b>	<b>5 ppb</b>
11.	Fodder	<b>G 6</b>	<b>1.345</b>	<b>0 ppb</b>
12.	Alfalfa		<b>1.374</b>	<b>0 ppb</b>
13.	Straw	<b>G 7</b>	<b>1.362</b>	<b>0 ppb</b>
14.	Alfalfa		<b>0.045</b>	<b>30 ppb</b>
15.	Alfalfa	<b>G 8</b>	<b>0.043</b>	<b>30 ppb</b>
16.	Straw		<b>0.045</b>	<b>30 ppb</b>

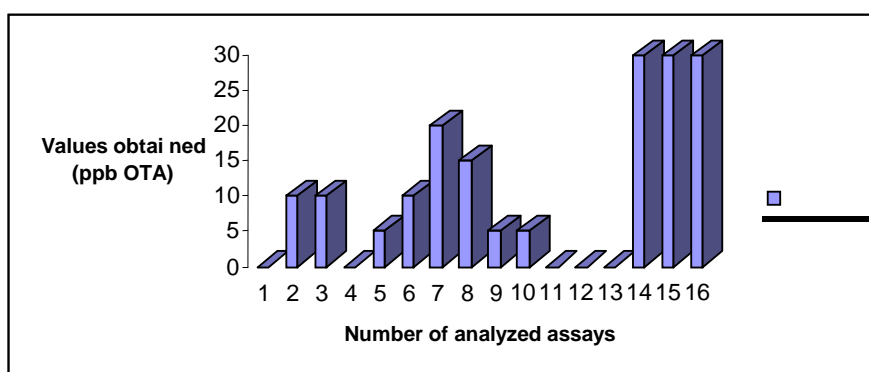


Chart 2. The representation of the contamination degree with OTA of the analyzed assays

- From the 16 assays analyzed for OTA, at 5 analyzed assays the results were undetectable (under the detection limit of the kit of 1  $\mu$ /Kg) representing a percentage of 31.25%.
- At the other 8 assays representing a percentage of 50 % from the analyzed assays, results had values over the detection limit, meaning between 10-30 ppb.
- At 3 analyzed assays, the obtained value exceeded the maximum allowed limit for this mycotoxin in fodder (5 ppb).
- According to the mycotoxicological analysis, found that examined fodder (straw, alfalfa and mixed fodder) were contaminated with fungi that could cause bovine ochratoxicosis (a percentage of 68.75%), but only 50% of feed samples can trigger ochratoxicosis (concentration in OTA  $\geq$  5 ppb).
- Lowest OTA concentration had mixed fodder samples (0-5 ppb).

Ochratoxin A is a mycotoxin that causes many adverse effects in domestic livestock and humans. The production of OTA in grain, particularly when stored under high-moisture conditions, can be readily

controlled. The toxic effect of OTA can be reduced by treatment of OTA contaminated grain (perhaps ammoniation and enzymatic hydrolysis of OTA to OA in ruminants and nonruminants), by reducing its absorption from the gastrointestinal tract (cholestyramine), or by feeding diets that will reduce its toxic effects (vitamin C) (2).

### Conclusions

After the test that were made and after the results that were obtained, regarding the analysis of the AF and OTA of the 16 assays of cattle's fodder, the following conclusions resulted:

► Of the 32 samples taken in preparation, 25 samples were contaminated with OTA and AF, and 7 samples were lacking, according mycotoxicological examination, contamination with fungi that produce OTA or AF. So 78.12% of the total samples were positive mycotoxicological exam subject for the two mycotoxins.

► Depending on forage harvesters, it was found that all three types of contaminated feed, regardless of their origin, but different values recorded in the OTA and AF concentration.

► Quantitative determination performed using ELISA method, direct competitive immunoenzymatic test - fast Ridascreen - led to a result set that assessment objections can be made to feed cattle given feed quality, traditionally reared in households in an urban area. Whether because of how the storage of feed, either because of inappropriate conditions in which animals are exploited, in many cases, animals develop various pathologies that may be associated with micotoxicosis.

► The certainty diagnosis of bovine mycotoxicosis is achieved only by collating data from clinical history, epidemiological investigation, clinical examination and completed the mandatory toxicology laboratory exams.

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# RESEARCH ON SWINE HOUSING MICROCLIMATE WITH THE INTENSIVE EXPLOITATION

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## Abstract

The works were carried out during 2009 in two private units for growth and the intensive exploitation of pigs, one of them with computer-automated workflow technology, the other unit having mechanical microclimate control.

Since each unit has a total of 10 samples taken to assess the microclimate in housing and was made the determinations of the following parameters: particulate matter, total bacteria and total fungi in the air. Also were isolated and identified bacterial and fungal species in air shelters.

Research has shown that computer-automated control system of the microclimate in housing makes it possible to keep within the parameters specified by the veterinary rules.

**Key words:** microclimate, swine housing, intensive exploitation

## INTRODUCTION

A unit located in the Calarasi county has a headcount of approx. 2500 head swine and was founded in 1998 by European investment funds, is intended for pig fat consumption. The farm is divided into sections as follows: pregnancy, maternity, baby piglets, fattening youth (over 25 kg), pigs for fattening in time I finish cabinet (35-65 kg) and fattening pigs in the finishing period II housing (65-105 kg) delivery platform. Flow is fully computerized and automated - feeding, watering, taking manure, microclimate control.

Unit B by Ilfov County was the second unit of intensive swine operations under study. The farm has a herd of about 100,000 heads of swine intended for fattening and selection. B unit structure is the same as in A. At the time of the research unit growth and operating system was partially automated pigs, feeding and watering being done manually. The takeover work with manure scrapers, water cushion, and carry out mechanical ventilation. For each unit were taken a number of 10 samples to assess the microclimate shelters, performed the measurements the following parameters: particulate matter, total plate count and total number of fungi in the air. Also were isolated and identified bacterial and fungal species in air shelters.

## METHODS

Air samples were taken from both units, compartments: pregnancy, maternity, fattening youth (over 25 kg), fat pigs during the finishing of the case I (35-60 kg), fat pigs in the finishing period II the case (65-105 kg). Evaluation parameters of microclimate in housing for pigs. Particulate matter in air shelters were taken with electric pump with flow rate 4 l / min on glass fiber filters and were expressed in mg / m<sup>3</sup> air. The pumps were placed in the center housing, corridors of speakers, the head of the animal. Total bacteria and total fungi were determined by

sedimentation method of passive air Petri dish with solid medium for bacteria and fungi respectively.

Results were expressed as NTG / m<sup>3</sup> air and NTF / m<sup>3</sup> air. Bacterial and fungal species in air shelters were isolated and identified by bacteriological and mycological respectively.

## RESULTS

In Table no. 1 presents the results of the evaluation parameters of microclimate in housing for pigs in unit A.

Table no. 1. Evaluation parameters of microclimate in housing for pigs

No	Compartment of swine	Particles in suspension (mg/m <sup>3</sup> aer)	NTG/ m <sup>3</sup> aer	NTF/ m <sup>3</sup> aer
1.	Gestation	3,2	200.000	<12.500
2.	Gestation	3,0	200.000	<12.500
3.	Maternity	3,1	210.000	<12.500
4.	Maternity	3,2	220.000	<12.500
5.	Youth for fattening (25kg)	3,9	240.000	<12.500
6.	Youth for fattening (25kg)	3,8	250.000	<12.500
7.	Fat pigs (35-60kg)	3,9	250.000	<12.500
8.	Fat pigs (35-60kg)	4,3	250.000	<12.500
9.	Fat pigs (65-105kg)	4,9	260.000*	<12.500
10.	Fat pigs (65-105kg)	4,7	260.000*	<12.500

\* values outside the veterinary hygiene standards

The bacteriological examination were isolated and identified from the air shelters nehemolitic strains of *Escherichia coli* spp *Stapylococcus*

The mycological examination were isolated and identified strains of *Candida albicans*.

In Table. no. 2 are presented the results of the evaluation parameters of microclimate in housing for swine unit B.

Table no. 2. Evaluation parameters of microclimate in housing for pigs

No	Compartment of swine	Particles in suspension (mg/m <sup>3</sup> aer)	NTG/ m <sup>3</sup> aer	NTF/ m <sup>3</sup> aer
1.	Gestation	5,2	250.000	12.500

2.	Gestation	5,1	250.000	12.500
3.	Maternity	5,1	250.000	12.500
4.	Maternity	5,1	250.000	12.500
5.	Youth for fattening (25kg)	5,7	260.000*	12.500
6.	Youth for fattening (25kg)	5,5	260.000*	12.500
7.	Fat pigs (35-60kg)	5,9	270.000*	12.500
8.	Fat pigs (35-60kg)	6,3	260.000*	13.000*
9.	Fat pigs (65-105kg)	6,8	280.000*	14.000*
10.	Fat pigs (65-105kg)	6,9	280.000*	14.500*

\* values outside the veterinary hygiene standards

The bacteriological examination were isolated and identified from the air shelters gram-negative endotoxin potential producers represented by *Escherichia coli* and *Pseudomonas aeruginosa*, with gram-positive *Staphylococcus aureus*, *Bacillus subtilis*, *Proteus spp.* The mycological examination were isolated and identified: *Candida albicans.*, *Aspergillus fumigatus*, *Aspergillus niger*.

It is noted that the unit B was isolated compartments and identified flora bacterial and fungal more varied than unit A. Also, in unit B have been analyzed parameter values outside the limits recommended veterinary departments in six of the 10 investigated.

***Sanitary veterinary rules for the parameters analyzed in this study (1, 3):***

- Particulate matter in air shelters for animals: under 15 mg / m<sup>3</sup> air
- NTG / m<sup>3</sup> air: under 250,000 bacteria CFU
- NTF / m<sup>3</sup> air: under 12 500 CFU fungi

**CONCLUSIONS**

**Unit A**

- Fine particulate matter has ranged from 3.0 to 4.9 mg / m<sup>3</sup> air, values that are below the recommended veterinary 15 mg / m<sup>3</sup> air
- Total number of germs easily exceeded the value of 250,000 CFU / m<sup>3</sup> air compartments for fat pigs (65-105kg) in both determinations
- Total number of fungi was found to be below the allowable of 12,500 CFU fungi / m<sup>3</sup> air
- computer-automated system control micro-compartments made it possible to maintain unity within the parameters specified by the veterinary rules

**Unit B**

- particulate matter in air compartments of pigs had values ranging from 5.1 to 6.9 mg / m<sup>3</sup> air, values that fall within 15 mg / m<sup>3</sup> air, being more elevated than those of unit A
- NTG / m<sup>3</sup> air is above the allowable values in six compartments



- NTF / m<sup>3</sup> air is above the permitted levels in three compartments, and the other seven departments were found slightly above the limit allowed (12 500 CFU fungi / m<sup>3</sup> air)
- Lack of computerized automated system, growth and exploitation of pigs is as microclimate parameters to be harder on the way, recorded values above the recommended limits of veterinary

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## STUDY REGARDING ORHIDECTOMY METHODS ON DOGS

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### **Abstract**

*Represents a major percentage orchidectomy within genitalia surgery in dogs. This is requested by the owners very often, both because of the aggressive male behavioral disorders, and because of problems (most commonly prostate or testicular).*

*Observations were made on a total of 30 medium and large dogs over 20 kg of different races, brought FMV USH clinic for neutering and focused comparison of surgical techniques. The classical techniques aimed to improve both shortened convalescence, and avoidance of postoperative complications. They were usually translated by swelling of the scrotal region or disposal sutures superficial anatomical layers. The comparative study looked at three different surgical techniques for each of them were seen in 10 animals each.*

**Keywords:** orchidectomy, dog neutering

### **Introduction**

This paper aims to make a difference in efficacy between surgical methods and orchidectomy in dogs present a statistics of 30 cases, the advantages or disadvantages of each method.

Study were subjected to 30 dogs of various breeds, all over 20 kg for each method by studying the evolution, after surgery, 10 animals.

### **Materials and methods:**

- a) The first method is become almost traditional, single scrotal incision, on the interdartoic septum, an incision is made as small testicles being shown by taxis.
- b) The second method is the removal of scrotal bags, an incision is made periscrotal, elongated, after removing the remaining testis is sutured wound edges, all in separate points, with absorbable thread, possibly after a wound by suturing subcutaneous tissue run. The method is more laborious and has proven it can not be replaced in case of testicular tumor mass, adherent to surrounding tissues. Assume a more abundant bleeding (lowered, obviously, by the use of electrocautery).
- c) The third method is that the 'prescrotal incision' orchidectomy. Consists in making an incision in the region prescrotala right or left paramedian and incision care all anatomical layers up to the testicle which is "made" in this area through a vigorous taxis. Testicle is evidenced by congener interdartoic septum incision. Increased need for medical attention than not extrapelvina incision and urethra. Restoration is still in anatomical layers separate points with absorbable thread and the dermo-dermal skin suture.

## Results and discussion:

The dogs were subjected variant) showed that the postoperative period is relatively long, averaging 8 days. Also, an obvious two cases of scrotal swelling was the object of daily care (local use of antibiotic sprays, etc.). In two cases because of discomfort caused by licking abundant locally, and gave up 2 or 3 points of suture.

If the variant is b) it appears that this method is quite laborious, but it is indispensable when acceding testicular tumors and testicular tissue to be removed immediately adjacent. Suturing can sometimes be difficult, and discomfort caused by resulting voltage, and the fact that the tension is increased by walking an animal, involves the healing. Sometimes they run, during surgery, small incisions are located near by the first incision , just to relieve the tension zone. This will further scar smoothly, with minimal local care.

Postoperative treatment was the duration, all 10 cases, an average of 9-10 days and involves daily antibiotics and Betadine. In two cases, very old dogs, 12 and 14 years, treatment after surgery that lasted 15 to 16 days (up to removing sutures).

If the variant is c), with prescrotal incision, was observed that the postoperative period in all 10 cases was on average 2 days. Single incision, small prescrotal located, and because the technique allows the superficial dermal-dermal suture, made this method to advance their net on the other two, both in terms of duration of surgery, and postoperative treatment . Not to mention that the technique can be used in testicular tumors adherent to the tissue .

The table below is presented, in comparison, the average recovery of orchidectomy according to the method chosen, as well as observations related to each method:

Orchidectomy method	Postoperative Period	Observations average
Orchidectomy incision	7-8 days	recovery is relatively long (7-8 days) involved daily repeated care
Orchidectomy with excision scrotum	9-10 days	recovery difficult, prolonged up to 15-16 days, depending on the size of the wound
Orchidectomy with prescrotal incision	two days	recovery was rapid and complete in all 10 cases

## Conclusions:

Prescrotal orchidectomy could be a successfully method , with single incision on the interdartoic septum . Recovery is significantly reduced, and the incision that allows for easy suture, dermo-dermal superficial anatomical layers, creating a comfort for animal.

Orchidectomy with total ablation requires scrotal bags, because large scale incised tissue, creating the necessity of micro-incisions adjacent to the main wound, whole animal surgery creates a major upset, but the technique can not be replaced when they meet testicular tumors massive adhesions surrounding tissue.

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# METABOLIC DISEASE OF BOVINS AND RISKS FACTOR

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## Abstract

Most of the metabolic diseases of dairy cows - milk fever, ketosis, retained placenta (RP), and displacement of the abomasum - occur within the first two wk of lactation. In addition to metabolic disease, the majority of infectious disease experienced by the dairy cow, especially mastitis, but also diseases such as Johne's disease and Salmonellosis, become clinically apparent during the first two wk of lactation. Metabolic disease is the most commonly recognized disease on dairy farms. While the pathogenesis is well known, metabolic disorders continue to occur. Metabolic diseases are associated, with one disease predisposing to another. Evidence suggests that metabolic disease affects host defense, and therefore, impacts the common infectious diseases of dairy cows. Risk for metabolic disease is affected by dietary formulation but is modified by cow behavior and intake. Regardless of dietary formulation, the cow and management factors on a given farm may determine the impact of metabolic disease.

## Rezumat

Majoritatea bolilor metabolice ale vacilor de lapte: febra laptelui, cetoza, retentia placentara (RP) și deplasarea abomasumului – apar în primele doua săptămâni de lactație. În plus față de bolile metabolice, majoritatea bolilor infecțioase de care suferă vacile de lapte, mai ales mastita, dar și alte boli cum sunt, boala lui Johnes și salmoneloza, devin evidente din punct de vedere clinic în primele două săptămâni de lactație. Boala metabolică este cel mai adesea observată în fermele de lapte. Cu toate că se știe bine patogeneza lor, tulburările metabolice continuă să apară. Bolile metabolice sunt asociate, una predispunând la alta. Dovezile sugerează că bolile metabolice afectează sistemul de apărare al gazdei și deci cele mai obișnuite boli infecțioase ale vacilor de lapte. Riscul pentru bolile metabolice este influențat de alcătuirea rațiilor și este modificat de comportamentul vacilor și de consumul acestora. Indiferent de formularea rațiilor, vaca și factorii de management dintr-o anumită fermă pot determina impactul bolilor metabolice.

## Introduction

Metabolic diseases are those associated with the chemical processes necessary for maintenance of life. In cattle, metabolic diseases include errors in electrolyte / mineral metabolism, of which parturient hypocalcemia (milk fever) is most common, or errors associated with energy metabolism, including ketosis and displaced abomasum. This review will make two assumptions before reviewing studies correlating metabolic disease with changes in infectious disease resistance. One of those assumptions is that mammary gland infections are less likely in animals with a *strong* immune system and that periparturient immune suppression exists and predisposes cows to mastitis and other infectious disease. Metabolic diseases are associated in that the occurrence of

one increases the risk of another. These associations tend to leverage the impact of disease on the animal (Correa et al., 1993).

Parturient hypocalcemia and ketosis can present in either clinical or subclinical states. Clinical disease implies that cows exhibit physical abnormalities. Subclinical disease is one where cows do not exhibit clinical signs, but the biochemical condition is present. Most producers have been content to estimate the impact of metabolic disease as a function of occurrence of clinical disease. While clinical disease occurs at a modest rate, subclinical disease has become recognized as common.

### **Occurrence of Metabolic Disease**

Clinical parturient hypocalcemia affects an average of 6% of cows and has been associated with a 3-fold increased risk of dystocia, retained placenta, and displaced abomasum, and a nearly 9-fold increased risk for clinical ketosis and mastitis (Curtis et al., 1983; Kelton et al., 1998). Subclinical hypocalcemia, defined as plasma calcium of 5.5-8.0 mg/dl within 48 hours of parturition, has been preliminarily reported to occur in 25.3, 43.9, and 57.8% of lactation 1, 2, and 3+ cows (Reinhardt et al., 2004).

Clinical ketosis is estimated to affect about 6% of cows (Kelton et al., 1998). However, subclinical ketosis, defined by postpartum serum beta hydroxybutyrate, affected 59% of cows (Duffield et al., 1998). Ketosis is associated with a decrease in milk production and increased risk of other postpartum diseases (Rajala-Schultz et al., 1999). It is known that the risk of displaced abomasum is increased as a consequence of subclinical ketosis in lactation (Geishauser et al., 1997) or in the 2 weeks leading up to calving (LeBlanc et al., 2005).

These data may be interpreted several ways. They do suggest that there are a high proportion of cows very near "the edge" of clinical disease. This further suggests that any limited stressor, acting to tip the balance in favor of disease, may cause a very considerable proportion of cows to be clinically affected.

In the most parsimonious terms, metabolic disease, both electrolyte related and energy related, may be considered a problem associated with diet formulation, diet consumption, and/or individual (i.e., genetic) factors. Of these, diet consumption is probably the most variable. Therefore, if a single risk factor "root cause" of metabolic disease is to be considered, that "root cause" would focus on the factors associated with dry matter intake (DMI) in late gestation/early lactation cows. This is particularly and directly the case for the energy related diseases.

### **Energy Associated Disease**

Ketosis, fatty liver disease, and displaced abomasum are the common energy related metabolic diseases. Energy related disease is generally thought to occur as a result of excessive lipolysis (fat breakdown) that leads to ketosis/fatty liver. Lipolysis is stimulated when energy output exceeds intake. Endocrine drivers of lipolysis include decreased insulin (low insulin allows lipolysis to continue), increased glucagon (which increases lipolysis), increased glucocorticosteroids (cortisol -

which increases lipolysis), and catecholamines (epinephrine/norepinephrine -the so called "fight or flight" hormones that are powerful lipolytics). While some of these mediators are beyond direct control, the glucocorticosteroids and catecholamines are important mediators that are, to at least a partial degree, dictated by and within control of management.

### **Risk Factors for Altered DMI**

Body condition, social interaction, and concurrent disease are a few of the many factors affecting DMI. It is well known that over-conditioned cows [body condition score (BCS) • 4.0] have a greater decline in DMI around calving, putting them in a position of susceptibility to energy related disease. It has been suggested that adipose cells of over-conditioned cows are more sensitive to signals to initiate fat breakdown, and fat cows may exhibit insulin resistance. Over-conditioned cows tend to have increased fat breakdown, increased liver lipid concentration, and a shift toward ketogenesis. It appears that cows near calving with BCS 4.0 have a marked propensity toward lipid mobilization, and cows with BCS 3.0 have little propensity to mobilize fat (Duffield et al., 1999). Therefore, the recommendation that late dry cows be in a BCS range of 3.25 to 3.75 probably represents a good tradeoff between subsequent milk production and risk of metabolic disease. However, careful managers may be able to maintain health and gain high production in cows with greater BCS if environmental conditions are optimal and energy distress is avoided (Contreras et al., 2004).

Social (or grouping) stress can result in alterations of cow behavior and may affect energy balance. The effects may be mediated through decreased feed intake or through the stress induced lipolysis pathways. Pen moves result in observed social disorder for 2 days, with a milk yield depression of 2 to 5% for the average cow (Hasegawa et al., 1997). While this is a modest effect, social stress can effect the non-dominant cow to a much greater degree. Dominate cows (usually older, larger, more senior, and gaining weight) are largely unaffected by a group change. However, non-dominant cows (typically younger, smaller body size, and/or cows losing weight) may be targets of aggressive social behavior, with resulting less opportunity for feed and rest. Clinical ketosis and fat infiltration of the liver in late pregnant cows has been observed following feed restriction of 30 to 50% or fasting for 4 to 6 days (Gerloff and Herdt, 1984). Therefore, coupling the natural decline in DMI with social stress lasting more than two days, especially in non-dominant animals entering a marginal housing situation, a significant proportion of animals could be placed in acute negative energy balance leading to energy distress and clinical disease.

Social effects are accentuated in larger cow groups/herds, so they assume more importance as herds grow in size. The ability to measure cow interaction, and the effect it has on feeding behavior, is only beginning to be addressed. Social interaction is dependent on the constitution of the group, as well as housing, feeding, and other environmental factors. Therefore, the relationships can become complex and difficult to predict. In general, minimizing re-grouping at key times has been under investigation. These times include the period of 5 days prior to calving and 1 to 10 days after calving (Cook and Nordlund, 2004).

### **Relationships of Energy, Disease, and Host Defense**

Three other related diseases, retained placenta, endometritis, and mastitis, are prevalent conditions that have been putatively associated with energy deficiency in cows. Endometritis and mastitis affect 17% and 13 to 45% of lactations, respectively, and are infectious in origin, but the bacterial agents are considered opportunists so that these diseases are largely determined by cow defense (Hoganetal., 1989; Epperson et al., 1993; USD A, 1996; LeBlanc et al, 2002). Neutrophils are very important in bacterial defense, and it was shown that neutrophil function declines in late gestation, reaching a nadir near calving (Kehrli et al., 1989). Additionally, neutrophils are important in placental release, and cows with retained placenta had a deficiency in neutrophil function in the prepartum period (Kimura et al., 2002). Ketone bodies appear to decrease neutrophil response (McMurray et al., 1990; Sartorelli et al., 1999). Cows that exhibited hepatic lipidosis, a lesion consistent with energy distress, took longerto clear experimental intramammary infection and had blunted response to vaccination (Hill et al., 1985;Wentiketal., 1997). In addition, *invivo* work suggests that improvements in energy balance in late gestation tended to decrease retained placenta (Duffield et al., 2002). While it is unclear how negative energy balance affects host defense, it is important to recognize that diseases of the mammary gland and uterus may be associated with energy distress. Energy balance should be considered a potential contributor to these energy related diseases if antioxidant vitamins and minerals are adequate.

## Summary

Metabolic diseases are interrelated, so that one disease increases risk for another. The energy associated diseases include ketosis, displaced abomasum, fatty liver, retained placenta, metritis, and possibly mastitis.

Providing an environment for an adaptive cow response will remain key to health. Dairy advisors must take an active role in promoting quantitative monitoring to assist the producer. In addition to tracking average DMI, monitoring energy balance using milk or blood NEFA or ketone assays may be essential, and may provide an early warning of problems to come. Since disease represents failures (those cows who could not negotiate stress), analysis of disease incidence records must be conducted and compared to known risk factors, including BCS, DMI, pen moves, and concurrent disease. These areas are obvious points where nutritionists and veterinarians can interact in a cooperative relationship.

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# UTILIZATION OF VARIOUS PRE-ANESTHETIC DRUGS IN SEVOFLURANE ANESTHESIA IN DOGS

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## ABSTRACT

*The study was performed on 40 cases, aged between 2 and 16 years, of different breeds. Atropine has been administered to all animals which underwent surgery.*

*In the sevoflurane anesthesia we used 3 different pre-anesthetic protocols:*

*1) propofol administered i.v. – 28 cases (70 %);*

*2) diazepam-lidocaine-thiopental administered i.v., separately – 6 cases (15 %);*

*3) ketamine-midazolam-butorphanol administered i.v., in two stages – 6 cases (15 %);*

*Surgery involved: ovariohysterectomy, mamectomy, orchiectomy, enterotomy and splenectomy.*

*In the instance of Propofol use, the recovery was rapid (3-25 minutes) according to the age and the general state of the patient. When the combination of diazepam-lidocaine-thiopental was used, the recovery of the patients was slower (20-35 minutes), since lidocaine reduces the amount of thiopental required and stabilizes the myocardium (these drugs are useful in animals with central nervous system and/or cardiovascular depression). When the combination of ketamine-midazolam-butorphanol was used, there occurred no excitation phenomena and the recovery was faster (10-20 minutes).*

*The average concentration of sevoflurane to maintain anesthesia was 3.5 % (1-6 %).*

**Keywords:** *pre-anesthesia, monitoring, sevoflurane.*

## INTRODUCTION

In order to conduct surgery within the best conditions, anesthesia needs to influence the respiratory and cardiovascular system as little as possible. Moreover, it needs to induce analgesia, optimal miorelaxation and a most rapid and quiet recovery of the animal (2).

In the specialized literature, the most frequent pre-anesthetic protocol used in the Sevofluran anesthesia in dogs is the Propofol one.

The ideal inhalation anesthetic agent has to induce hypnosis, reduce reflexes and grant analgesia. Furthermore, it has to be easily controlled, grant a fast onset and offset of anesthesia, and have minimal adverse reactions (3). Sevofluran is a clear, colorless, volatile liquid, nonflammable in the air or oxygen. Sevofluran is one of the inhalation agents recommended to induce and maintain general anesthesia in animals.

Sevofluran is recommended in animals with cardiac ailments and in old patients and given its low solubility in the adipous tissue it can be used in obese patients (1).

## MATERIAL AND METHOD

The research involved 40 dogs, aged 2 – 16. Surgery covered: ovariohysterectomy, mamectomy, orchiectomy, enterotomy and splenectomy.

The animals undergoing surgery were pre-medicated with atropine sulphate 1% administered subcutaneously in a dose of 0.5 mg/10 kg live wieght.

Three protocols were used to induce anesthesia:

- propofol (6 mg/kg) administered i.v. – 28 cases (70%);
- diazepam (1 mg/kg) - lidocaine (2 mg/kg) - tiopental (4.5 mg/kg) administered i.v., separately – 6 cases (15 %);
- ketamine (7.5 mg/kg) – midazolam (0,1 mg/kg) - butorfanol (0,1 mg/kg) administered i.v., in two stages – 6 cases (15 %).



Fig. 1 – Inhalation narcosis device



Fig. 2 – Maintaining inhalation anesthesia with Sevofluran in dogs

The anesthesia was maintained, all along the duration of surgery, with Sevofluran in a concentration of 1-6% according to the age, the general state of the animal and the duration and type of surgery.

The main functions (cardiac frequency, respiratory frequency) as well as the body temperature and color of the mucous membranes were monitored during surgery.

## **RESULTS AND DISCUSSIONS**

The average duration of anesthesia was 50 minutes (30-180 minutes), in relation with the type of surgery.

Apnea was the most significant adverse effect following the use of Propofol, which however did not require medicine or mechanical intervention (assisted ventilation).

Propofol administration initially induced in the majority of the instances, an increase of the cardiac frequency which subsequently resumed the values characteristic to the breed and age.

The reduction of the respiratory frequency was noticed in all the monitored patients. This was due to the effects of the Sevofluran anesthesia.

The body temperature decreased by a mean of 1.5°C when the three pre-anesthetic protocols were used. The decrease in the body temperature occurred because of the skeletal muscles tonus reduction, of the vasodilatation and the diminishment of the capacity for thermoregulation self control.



Fig. 3 – Monitoring the main functions during the inhalation anesthesia with Sevofluran

The recovery of the palpebral reflex in the final part of surgery was faster when Propofol was used as a pre-anesthetic - an average of 5 minutes - as compared to the other protocols, which recorded an average of 12 minutes.

When Propofol was used, the recovery from anesthesia was fast (3-25 minutes), depending on the age and general state of the animal. When the combination of diazepam-lidocaine-thiopental was used, the recovery of the patients was slower (20-35 minutes), since lidocaine reduces the amount of thiopental required and stabilizes the myocardium (these drugs are useful in animals with central nervous system and/or cardiovascular depression). When the combination of ketamine-midazolam-butorphanol was used, there occurred no excitation phenomena and the recovery was faster (10-20 minutes).

### CONCLUSIONS

1. The most efficient pre-anesthetic protocol was the one involving administration of Propofol, as compared to the administration of the combinations diazepam-lidocaine-thiopental and ketamine-midazolam-butorphanol, which recommends it especially for the anesthesia of old animals.

2. The cardiac frequency was not significantly modified within the pre-anesthetic protocols used.

3. The body temperature did not vary significantly within the pre-anesthetic protocols used.

4. The recovery from anesthesia was much faster when Propofol was used as compared to the other two protocols.

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