MINISTRY OF EDUCATION, RESEARCH AND INNOVATION

"ION IONESCU DE LA BRAD"

UNIVERSITY OF AGRICULTURAL SCIENCES AND VETERINARY MEDICINE

Aleea M. Sadoveanu nr. 3, 700490 – IAŞI, ROMÂNIA

Tel. +40-232-213069/260650 Fax. +40-232-260650

TAX CODE: 4541840

E-mail: rectorat@uaiasi.ro, http://www.uaiasi.ro

Report on the research activities accomplished during 2009, in the project entitled:

"RESEARCHES RELATED TO THE PHYSIOLOGICAL AND CIRCADIAN SECRETOR INVOLVEMENTS OF THE PINEAL GLAND IN THE REPRODUCTION STATUS OF THE LAYING HENS "

no. 1645/2008

PROJECT MANAGER:
Prof.univ.dr.ing. Paul Corneliu Boisteanu

Epiphysis plays a controlling role in the daily and seasonal rhythms in most species, the secretory rhythm of melatonin mammalian pineal is controlled by the suprachiasmatic nucleus of the somatic nervous system pathways. In birds the pineal gland contains a circadian oscillator self-supportive and numerous input channels that synchronizes the internal biological clock, including sensitivity to direct sunlight.

Some physical input channels (light, temperature, magnetic field) and Biochemical (VIP, norepinephrine) that influence the release of melatonin, are functional in vitro, representing a good model for the study of circadian biological clock. Synthesis of melatonin in birds and other indole hormones in the pineal gland and retina occurs, the same biosynthetic pathways exist in the lateral eyes, which shows that the primary function of melatonin is related to the photoreceptor. Pineal gland of birds contains a circadian oscillator that has a major role in temporal organization of brain function.

Pineal indoles secretion has a high regularity that seems to be essential endogenous alternation modulated L / D and temperature. Circadian clock that regulates the transcription of each pinealocyte avian melatonin biosynthesis in the cycle is expressed rhythmically and L / D and D / D, both in vivo and in vitro. The body's adaptation to external environmental conditions, pineal, through its hormones, does not act directly on the gonads or the endocrine system, but acts on the brain and brain functions contribute to adaptation to environmental conditions, reproductive activity is only one aspect of this process. Pineal gland through melatonin biosynthesis, is a transducer that mediates photoperiodic seasonality of reproduction function. Involvement of melatonin in the functional axis of hypothalamus-pituitary-gonadal is demonstrated by increased levels associated with low secretion of gonadotropins, and decreased concentrations of melatonin activate the hypothalamic-pituitary-gonadal axis.

The researches have been run on 100 laying hens belonging to ALBO SL – 2000 hybrid, reared on permanent litter, at ground. The individuals were allocated in 3 experimental groups:

- L_1 – photic pinealectomy through exposure at 150 lux light, 24 hours daily;
- L_2 – lighting schedule of 12L/12D;
- L_3 – lighting schedule of 16L/8D.

In all 3 experimental groups, the feeding was made using mixed feed having M.E. 2700Kcal/kg; CP 14.5%; Lysine 0.65%; Methionine 0.3%; Ca 3.5%; P 0.55%; Na 0.14%; Cl 0.13%; Linoleic acid 0.85%; Total xenophiles 24ppm.

Feed consumption was 127g/hen/day, water was administered ad libitum.

Under the first objective of the research project were pursued following indicators hematology (WBC, RBC, Hb, HCT, MCV, MCH, MCHC), biochemical (glucose, total protein, total fat, cholesterol, phospholipids, triglycerides, ion sodium, potassium ions, calcium ions, SGOT, SGPT, urea).

Table 1

Variations in haematologial parameters

Parameters	Measurement units	L_1	L_2	L_3
WBC	$10^3/mm^3$	20.45	26.18	25.2
RBC	$10^6/mm^3$	2.45	2.85	2.33
Hb	g/dl	7.5	9.7	9.5
HCT	%	28.6	29.4	27.4
MCV	μm^3	129.4	130.6	126.6
MCH	pg	36.6	34.9	38.8
MCHC	g/dl	40.5	23.4	44.6

In physiological conditions, blood erythrocyte count variation due to the continuous adaptation of the minimum rate at the rate of erythropoiesis - erythrtrolisis. The average life span of erythrocytes is 28 days, while they represent 2.4% from whole figurate blood compounds . The studio made lots met oligocytemia phenomenon characterized by low blood cell count below 2.4 x $103/mm^3$, a phenomenon often observed in laying hens.

A decrease in the number of erythrocytes was observed in group L3, 2.33 x$103/mm^3$, drop that is associated with low levels of erythrocyte constants.

The derived erythrocyte traits provided data related to the size, shape and loading of erythrocytes with hemoglobin. The amount of hemoglobin varies depending on the laying intensity and feed structure.

Table 2

Variations of the main biochemical traits

Studied traits	Measurement units	L1	L2	L3
Glucose	mg/dl	260.4	320.2	200.1
Whole proteins	g/dl	5.2	2.3	5.3
Whole lipids	mg/dl	2562	2250	2112
Cholesterol	mg/dl	147	125	118
Triglycerides	mg/dl	216	195	186
Sodium ions	mg/dl	396	360	348
Potassium ions	mg/dl	6	7	4
Calcium ions	mg/dl	31.4	28.3	36.8
TGO	U/L	159	134	133
TGP	U/L	18	12	10
Urea	mg/dl	6.2	5.5	5.8

Total Lipids indicated values of 2250mg/dl in L1, 2562 in L2 and 2112 in the group L3. The small differences noted between the 3 groups, but inclusion within the physiological values obtained. Excess fat in laying hens can produce liver damage, followed by increase of up to 30% lipid content in liver.

Cholesterol resulted in photic pinealectomised group (L1) was 147 mg / dl compared with 125 mg / dl obtained from L2 and 118 mg / dl registered with L3.

Triglycerides in the blood of hens in L1 values were 195mg/dl, 216mg/dl in group L2 and L3 group that exposure to light was recorded 16L/8D value was 186mg/dl.

Blood hypoglycemia blood can be reflected in a higher quality of eggs, with implications for increased survival of offspring. Low blood sugar levels were recorded in group L3, 200.1 mg / dl, hypoglycemia indicating a deficiency of glucose metabolism and disruption of energy metabolism leading to disorders of the nervous system and parenchymatous dystrophy. Group L1 have values of 260.4 mg / dl and group L2 a value of 320.2 mg / dl.

In carbohydrate metabolism, pinealectomy increases glucose and raises impaired glucose tolerance.

A diet deficient in protein ratio leads to lower production of eggs, respectively hypoproteinemia.

It was found that the formation of physiologically on egg protein is more important than energy report. Plasma protein level is very important throughout the year, but period is critical in laying.

Plasma protein levels in birds can be influenced by hormones, including estrogen during follicular development.

Also the amount of protein may be due to low and low concentrations of albumin and globulin. L1 values were obtained at 5.2 g / dl in group L2 2.3 g / dl and 5.3 in group L3.

In plasma ions determined, values were as follows:

Sodium ions showed a plasma concentration of 396mg/dl at L1 to L2 360mg/dl, 348 mg / dl at L3.

Blood potassium ions had a concentration of 6 mg / dl in L1 to L3 respectively 4mg/dl 7mg/dl to L2.

In laying birds calcium deficiency causes osteopathy, which in addition to etching processes are reflected on the eggshell and extend the skeleton, through a process of replacement of mineralized tissue with osteoid and even fibrous tissue leading to bone deformities and locomotor disorders. Induction of estrogen may increase the calcium level during the laying period, immediately after laying or during ovulation.

Laying hens have after the laying of liver metabolism changes reflected in the increase of TGO. The L1 values were obtained from 159 U / L in group L2 of 134 U / L and in group L3 133U / L, values that exceed physiological limits.

Values TGP submit amendments to increase the muscle and liver illnesses, but their significance in diagnosis is limited.

In the three experimental groups have values of 18 U / L to L1, which forms VALUATION physiological limits, compared with 10 U / L recorded in group L3 and 12 U / L in group L2.

If carbohydrate metabolism, glucose increases pinealectomia and impaired glucose tolerance, while the hypoglycemic effect of pineal peptide extract, which is achieved through the mechanism of potentiation of the hypoglycaemic action of insulin and stimulate glycogen deposition in liver and muscle.

Anabolic properties have been highlighted and protein metabolism, resulting in decreased body weight pinealectomia and accelerating uptake of amino acids in the blood, chronic administration of pineal peptide extracts from these animals was followed by a significant increase in muscle protein nitrogen concentration, coupled with weight restoration injury.

On lipid metabolism, increasing lipid carried pinealectomia total cholesterol and fatty acids in the blood and pineal peptide extract reduced their blood concentration in normal food, the animals undergoing treatment supplemented with cholesterol to produce atherosclerosis lesions Experimental pineal peptide extract caused the atherosclerotic vascular disorders.

Structural changes in the three control groups were observed with optical microscopy.

Following investigations have evaluated the effects caused by photic pinealectomia, results have been interpreted in terms of variable values of hematological and biochemical indicators tracked.

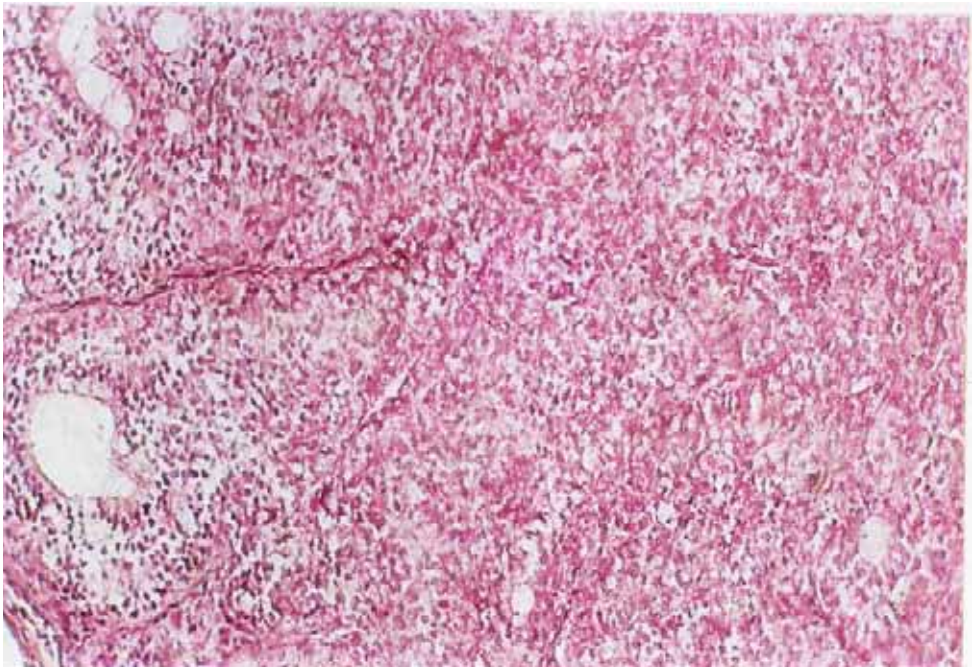

Fig. 1-Pineal gland well developed, containing normal secretory cells

Microscopic aspects revealed in the pineal gland of hens from L1 many secretory cells with small round nuclei and areas with dark cells arranged radial around the glandular ducts, reflecting secretory gland normality.

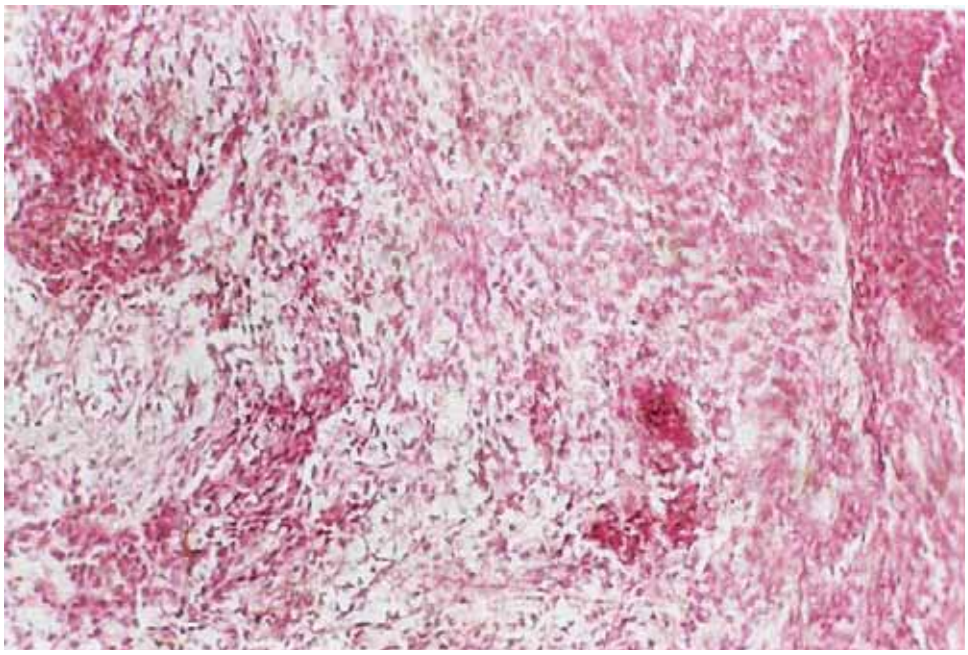

Fig.2-Section through the pineal gland – normal secretory activity with balanced proportion of both cell types

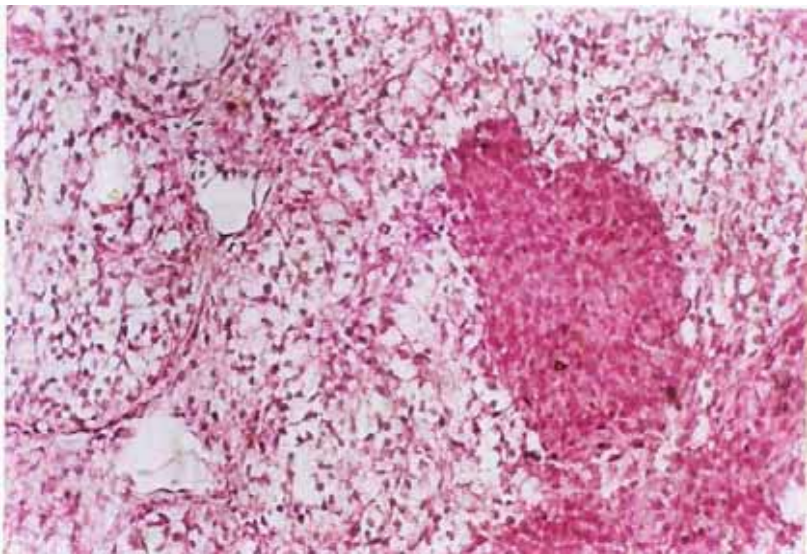

Fig.3- Pineal gland well developed, with compact groups of dark cells, alternating with clear cells and poor cystic formations

Thus, through optical microscopy, in the epiphysis of hens in the control group was observed in the presence of both normal cell types (clear and dark), with few cystic cavities, which shows a normal secretory activity.

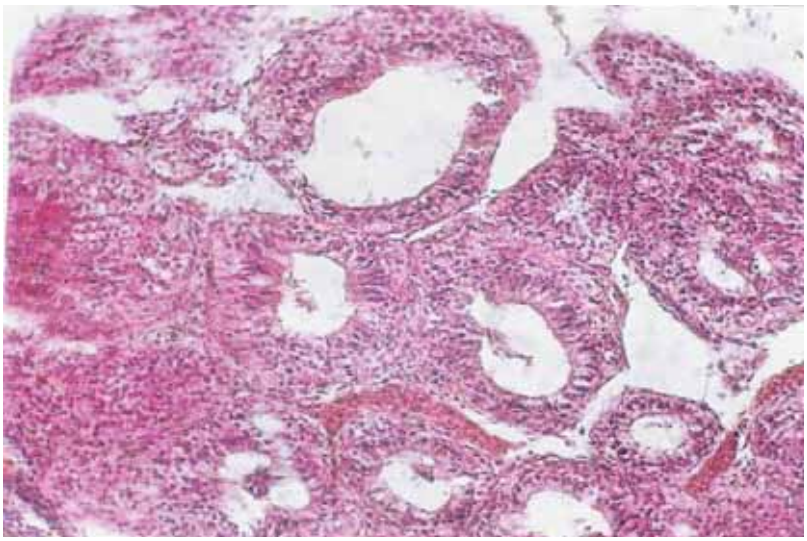

Fig.4-Atrophic epiphysis, with poor glandular parenchyma and many cystic cavities, delimited by ciliated ependimary cells

Photic pinealectomy induced atrophic changes in the pineal gland, represented by numerous and frequent cystic cavities and glial cells were due to inhibition of secretion of pineal gland tissue by continuous light stimulation. Pineal hormones involved in reproduction modulation in those animals dependent of physical factors in the external environment participate in photoperiodism regulation, fluid- electrolyte balance and intermediary metabolism regulation and in the modulation of central nervous system excitability and sleep-awake cycle.

In birds, the pineal involved in the regulation of reproductive status, which is characteristic to several hours during the ovulatory cycle.

Ovulatory cycle starts at 17 weeks a productive expression under the control of hypothalamic-pituitary-gonadal complex, complex and affected by neurohormonal factors epiphyseal ambient vibration mode (photic, thermal, seasonal).

Photic ablation of the pineal reveals the pineal hormones intervention as antigonadotropic effect causing gonadostimulant effect change as a result of the phase relationship between biological rhythms and circadian light-dark cycle with the changing relationship between circadian rhythm of photosensitivity and the cycle of artificial lighting.