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PhD THESIS

ABSTRACT

Investigations on the presence and impact of thyroid endocrinosis in the canine population from Muscel area

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CONTENT

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***Key words** : thyroid, dog, iodine, thyroid hormones, thyroxine, triiodothyronine, hypothyroidism, minerals*

ABSTRACT

A number of endogenous and exogenous factors exert direct and indirect effects on neuroendocrine system. The hypothalamus as the main mediator in the relationship between body and environment maintains body homeostasis via endocrine system. As a command center hypothalamus regulates metabolic processes in the body (the anabolic and catabolic ones) through thyroid, whose integration in the neuroendocrine system is achieved by interposing pituitary gland that acts as an interface between the nervous system and endocrine system. In the case of thyroid the "messenger" bearing the information from the pituitary gland, holding a regulatory and integrator role, is represented by TSH. Depending on the intensity of the thyroid stimulation by FSH, it responds by secretion of thyroxin according to the degree of stimulation. They observed that in dogs the description of the thyroid function through TSH does not lead to convincing results. Furthermore, T3 and T4 thyroid hormone levels are strongly influenced by a number of endogenous and exogenous factors that determine its complex dynamics. The most important environmental factor influencing thyroid status is represented by iodine. Iodine is part of thyroxin and triiodothyronine, therefore, either its shortage or its excess influence the thyroid activity a great deal. In humans there were established the optimal levels of iodine which must be provided daily to avoid the occurrence of pathology associated with iodine deficiency. Programs and measures implemented to ensure an optimum level of iodine in human nutrition have reduced the incidence of endemic goiter and cretinism, but on the other hand it is suspected that on the background of certain habits (the addition of a supraoptimal iodized salt to food) it is favored the occurrence of a specific excess iodine thyroid diseases consisting in the occurrence of thyrotoxicosis and autoimmune diseases. Unlike humans, for dogs they have not determined the optimal iodine nutrition level which must be insured by food. The difficulty of determining the level derives from the polymorphism linked to the waist, so that the ingestion of iodine should be properly related to the metabolisable energy of food intake, and not to its mass. Some

data suggest the existence of some adaptation mechanisms of dogs at lower levels of iodine through efficient recycling processes of ingested iodine.

In dogs it was noticed that the thyroid function cannot be measured simply in terms of TSH dosage, as there are many dogs with high levels of it in which, by no other means of investigation (biochemical and histological) to be demonstrated the existence of a thyroid disfunction.

On the other hand, there are cases in which the TSH ranges within normal levels, but which, through other diagnostic tools it is likely the existence of thyroid disturbance. Consequently, at present, there are conducted the total thyroxin dosage and total triiodothyronine, and, according to some works, the determination of unbound thyroxin, too, but often with unsatisfactory results.

Corroborating the clinical data, which are assumed to be the consequence of canine hypothyroidism, with TSH screening and thyroid hormone, does not result in convincing results, especially that the associated clinical picture of hypothyroidism is quite uncharacteristic. The clinical, acknowledged characteristics in literature, with bilateral symmetrical alopecia, dry hair, "rat tail", facial myxedema occur in an extremely small number of dogs affected by hypothyroidism.

In this context, the aim of this study was to supplement the information concerning dogs' thyroid activity also in the context of food diversification, containing industrially prepared food whose iodine content varies greatly. Moreover, the work suggests not only a way of iodine dosage from canine blood serum, but also a way to investigate the relationships between iodine and TSH, iodine, thyroxin and triiodothyronine. The study aimed at highlighting some possible interrelationships among some minerals, iodine, TSH and thyroid hormones, too.

As main objective the study aims to improve the information related to thyroid physiology in dogs, the importance of iodine played in maintaining the state of euthyroidism, but also to determine the amount of iodine in the serum of dogs in an area characterized by iodine deficiency.

In the first part of the thesis - Bibliographic study – there are presented data from literature concerning the anatomy, histology, biochemistry and physiology of the thyroid. We also researched survey data on iodine status in humans and dogs, nationally and internationally, as it is often encountered overlapping of human food consumption with that of dogs

To better understand the effects that thyroid hormone exerts on the body there has been studied the role it has on certain tissues and organs.

The studies conducted in the second part of the thesis –Personal researches - have evaluated the dog thyroid activity in relation to the iodine, TSH and some micronutrients levels. The relationship established between the iodine serum concentration and the type of food given to dogs (cooked, commercial, cooked + commercial) was also considered. By making clusters there were investigated the relationships established between iodine, TSH, thyroxin, triiodothyronine and some micronutrients but also the degree of mutual determination. By making these clusters we were able to determine to what extent the TSH level answers statistically, by feedback, to thyroxin and triiodothyronine levels, thus trying to explain the poor diagnostic value of TSH for canine hypothyroidism.

The relationships established among the parameters under study are complex, the mutual influences are direct, determined by statistical analysis of the main components, not being reduced to a minimum number of variables, meaning that the oscillation of all the studied variables cannot be explained by one or two variables.

In this study a particular issue was to create a protocol allowing blood dosage of iodine by ICP-OES technique. The dosage of serum sodium is quite difficult in terms of physic-chemical properties of that element. Sublimation of this element in the serum samples, when they are heated, means that the serum samples should be prepared in the cold to reduce the loss of iodine. The proposed method in this paper allows the analysis of serum free chemical preparation and therefore the analysis errors induced by the steps of preparation are reduced. In addition, this method is much less expensive (both in terms of the cost of the necessary equipment and of the reagents which are to be used) and faster compared to the preparation methods using the chemical preparation of the serum and equipment such as ICP -MS. Blood iodine dosage was a main issue of this study as we wanted to highlight the role of this trace element for the adjustment of pituitary gland - the thyroid gland axis.

The variations of iodine and TSH divide dogs in the study in three distinct groups which raises suspicions about the presence of internal and external factors that influence the relationship between pituitary and thyroid glands in that the same serum levels of iodine modulate different responses of the pituitary gland. Identifying the three groups of dogs, with a very high probability, and the existence of dogs placed marginally within the group support both the possibility of subgroups and having another factor by which the existence of these groups is explained more accurately in terms of statistics and physiology.

The adjustment of the pituitary gland – thyroid gland axis can be explained by the influence of the concentration of iodine serum on the value of TSH serum in a percentage between 46% and 70%, which demonstrates the existence of another factor or combination of factors, too, which have a major influence on serum TSH concentration. On the other hand, in the three groups which were identified, the variation in serum thyroxin and triiodothyronine can be explained, in a small percentage, by the variation of serum TSH which means that many other factors contribute to maintaining optimal concentrations of the two serum thyroid hormones. Such low interdependence between both TSH and thyroid hormones shows that in case of dogs, the diagnostic value of thyroid endocrines, by means of TSH, is limited.

The types of food (marketed, cooked, marketed+ cooked) influence significantly ($p = 0.0002$) the serum iodine concentration.The lowest blood iodine concentrations are recorded in dogs fed with cooked food, unlike dogs fed with a mixture of commercial food and cooked food, which recorded the highest level. Following investigations it was found that different types of food affect thyroid activity by iodine intake. From a nutritional standpoint thethyroid is able to compensate iodine deficiency, although it is hard to state below which limit it can be said that dietary intake of iodine is insufficient. In the same context it is likely for iodine intake to be in excess with different implications on thyroid physiology and pathology.

It is difficult to quantify iodine intake provided nutritionally as it is known that during storage of food the amount of iodine decreases in time, so the lack of determination thereof, carried out around feeding time or the determination of correction coefficients, make it difficult to quantify the actual intake of iodine.

Minerals serve multiple metabolic functions through the integration of some of them in the enzyme systems or within the insurance of electrochemical gradients which are vital for the ions transport (including iodine) at cellular level.Instead some minerals play a not well-defined part and can be considered contaminants that could favor or intervine in the thyroid function.

The hypothesis stated in this work is that some minerals might exert direct and indirect influence on the dynamics of thyroid so serum concentrations of minerals (aluminum, boron, calcium, chromium, copper, iron, potassium, magnesium, manganese, sodium , strontium, zinc) were measured and by applying appropriate statistical methods we investigated this aspect.

The choice of above mentioned minerals, in the absence of evidence in literature, was imposed by the available material resources (equipment, reagents) and by the hypotheses advanced by some authors. Thus the study shows that boron has the most significant effect on thethyroid physiology, this one acting synergistically with thyroid TSH for all the three groups of dogs identified, but antagonistically with triiodothyronine and thyroxine .The investigation of serum aluminum has allowed to establish the influence of this mineral on TSH and thyroid hormones and also a ranking of the three groups.

There could not be any evidence of a direct correlation between calcium and TSH, thyroxine and triiodothyronine but the study reveals a direct relationship between calcium, strontium and magnesium for all the groups identified. Sodium and potassium do not exert direct influence on TSH and thyroid hormones.Although some studies raise suspicions of involvement copper deficiency in the occurrence of hypothyroidism, this study could not catch a direct correlation between this one and thyroid hormones

The data collected for this study have not revealed the existence of significant statistical relationship between dogs' size, sex, age and the serum TSH and thyroid hormone,but the complex analysis of the many factors that can interfere with thyroid activity remains a major desideratum.