GASTROINTESTINAL NEMATODIASIS IN PIGS
AND THE INFLUENCE ON THE PHYSIOLOGY
OF DIGESTION

SUMMARY OF PHD THESIS

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Introduction

Production of pig meat is the main branch in Romania, this sector accounting for them for over 50% of total meat production (in weight), about the same proportion being found in the total consumption of the population. Even if the number of intensive pig units is constantly expanding and antiparasitic medication applied to is more modern and effective, can not be said that the firm is free of infestation. An increase in the prevalence nematodoses is found in other European countries but also in other regions of the globe.

Economic problem lies in the fact that often evolves nematodoses subacute and chronic forms or subclinical develops when the common examinations (inspection, palpation, auscultation) reveals no visible pathological manifestations. Although measures are programmed to prevent parasitic infestations they are still insufficient and it adds to the total costs of maintaining animals. Use of phytotherapy in the treatment of parasitic diseases has been applied since ancient times and the use of plants, alone or in combination, is increasingly studied because of the need for finding alternative ways to deal effectively.

Pigs were used as a model for numerous studies of nutrition due, not least to the physiological peculiarities of the digestive tract that are similar to human. Study of the parasite development in pigs is an experimental model to understand the mode of action of the parasites from the same Genus (*Ascaris lubricoides*) on humans.

The thesis was structured in two parts. The bibliography includes a chapter with 8 sections and four chapters from own research.

In the research part we investigated the epidemiology of nematodes infestation in some counties of Transylvania, we studied the antiparasitic effect of plant extracts, have been evaluated biochemical, haematological, immunological and histopathological aspects from *Ascaris suum* experimental infestations in pigs and have analyzed the effects of *Ascaris suum* in natural infested pigs on digestion and food processing.
I. MONITORING OF EPIDEMIOLOGICAL INDICATORS OF DIGESTIVE NEMATODOSES IN PIGS FROM THE INTENSIVE, SEMI INTENSIVE AND EXTENSIVE SYSTEMS IN SOME COUNTIES IN NORTHWESTERN TRANSYLVANIA

Material and methods.

Were studied a total of 1862 faecal samples from 5727 pigs raised in different growth. The purpose of this investigation were studied digestive nematodoses pigs from a total of 3 units with semi-intensive growing system, a farm with intensive system and 7 villages in north-western Romania.

Parasitological examination was conducted by Willis and McMaster method. Data were centralized and processed in tables indexed by extensivity (E-%), intensivitate (I-OPG) and the instant prevalence (Pi-%). Statistical calculation was performed with Graph Pad program in the state using the unpaired t-test in ANOVA categorical analysis system.

Results

The diagnosed parasites were *Ascaris suum*, *Oesophagostomum sp.* And *Trichocephalus suis* to all categories of pigs investigated.

At 0-3 months category averaged throughout the extensivity was: the infestation with *A. suum* for 41 ± 8.4% for the extensive system, 11.10 ± 13.87% for semi-intensive system (p <0.05), the infestation with *Oesophagostomum sp.* 18.5 ± 5.25% for the extensive system, 5.18 ± 4.62% for semi-intensive system, and the infestation of *Trichocephalus suis* system extensively 8.5 ± 3.41%, 8.14 ± 7.14% for semi-intensive system. Maximum intensity was determined in the *Oesophagostomum sp.* infestation of 2300 EPG.

The youth category averaged over the whole period extensivity was: in the infestation with *A. suum* of 47.46 ± 7.34% for the extensive system, 11.10 ± 13.87% for semi-intensive system, 10.83 ± 5.2% in the intensive system is (p <0.02) in the infestation with *Oesophagostomum sp.* of 48.41 ± 7.69% for the extensive system, 6.1 ± 5.35% for semi-intensive system (p <0.02), and the infestation with *Trichocephalus suis* 10.12 ± 5.16 % for extensive system, 8.61 ± 10.63% for semi-intensive system (p <0.01). Maximum of determined intensity was for the infestation with *Oesophagostomum sp.* 7600 EPG in the
extensive system. Extensivity for sows category averaged throughout the period was: the infestation with *A. suum* of 54.5 ± 7.54% for the extensive system, 15 ± 13.22% for semi-intensive system, the infestation with *Oesophagostomum sp.* of 60.29 ± 5.62% for the extensive system, 8.88 ± 7.69% for semi-intensive system (p <0.05), and the infestation of *Trichocephalus suis* 2.94 ± 3.39% for extensive system, 4.44 ± 3.84% for semi-intensive system. Maximum intensity of 4400 EPG was measured in the infestation with *Oesophagostomum sp.* with significant differences between farming systems in the infestation with *Trichcephalus suis* (p <0.05).

Extensivity of the infestations for breeding boars category averaged over the period was: the infestation with *A. suum* 23.75 ± 11.08% for the extensive system, 33.33 ± 38.18% for semi-intensive system, the infestation with *Oesophagostomum sp.* 30 ± 7.07% for the extensive system, 25 ± 25% for semi-intensive system, and in *Trichocephalus suis* infestation of 1.25 ± 2.5% for the extensive system, 8.33 ± 14.43% for semi-intensive system. Maximum intensity in sows was measured in *A.suum* infestation of 6100 EPG in the semi intensive system and there were recorded significant differences between farming systems evaluated (p <0.02).

II. BENCHMARKING THE EFFECT OF EXTRACTS FROM PLANTS AS ANTIPARASITIC MEDICINE IN PIGS BRED IN EXTENSIVE AND SEMI INTENSIVE GROWING SYSTEMS

Material and methods.
The study was conducted from 01.02.2008-01.04.2008 in a total of 60 pigs aged up to 5 months male and female. Regarding the phytotherapy with four types of plant extracts: oil pumpkin (*Cucurbita pepo*) (group I) oil, marigold (*Calendula officinalis*) (group II), hydroalcoholic extract of wormwood (*Artemisia absinthium*) (group III), the French marigold hydroalcoholic extract (*Tagetes patula*) (group IV) and a medicinal product based on benzimidazoles derivatives (15%) (group V) we investigated the evolution of the infestations with nematods. Group VI was considered as control. Samples were collected both before treatment and at 7, 14 and 28 days.
Results

Fig. 1 The variation of extensivity of *Oesophagostomum sp.* infestation

Fig. 2 The variation of extensivity of *Trichcephalus suis.* infestation

Fig. 3 The variation of extensivity of *Ascaris suum* infestation in the 6 studied groups
Pumpkin oil (*Cucurbita pepo*), had reduced efficacy in the treatment of *Oesophagostomum* infestation and in *Ascaris suum* and *Trichocephalus suis* was insignificant, which can be seen only in reducing the number of eggs.

Calendula oil (*Calendula officinalis*) has efficacy for esophagostomosis and was moderately effective in ascariasis.

Hydroalcoholic extract of wormwood (*Artemisia absinthium*) had an average efficiency of *Ascaris suum* on but not on *Oesophago stomum sp*.

Whole plant extract French marigold (*Tagetes patula*), had an outcome better than the other extracts as antiparasitic both against esophagostomosis and against ascariasis. Upon *Trichocephalus suis* had diminished effect. If the group treated with modern anti-parasite product (oxibendazole, 15%), the results were superior to all plant extracts.

**CHAPTER 3. EVALUATION OF BIOCHEMICAL, HEMATOLOGICAL, IMMUNOLOGICAL AND HISTOPATHOLOGICAL PRODUCED BY ASCARIS SUUM IN AN EXPERIMENTAL INFESTATIONS IN PIGS**

The objectives of this chapter were: achievement of experimental infestations with 1000 eggs of *Ascaris suum* in pigs; investigation of biochemical and hematological blood parameters in *Ascaris suum* artificial infestation, the assessment of *Ascaris suum* impact on the immune system of pigs and analysis of histopathological aspect involving the experimental ascariasis in pigs.

**Material and methods.**

The study was conducted between December 2008 - July 2009 on a number of 15 pigs (3 groups, n = 5), mixed race with similar age (5 months). Lots 2 and 3 were infected artificially in the feed, with 1000 eggs each individual and group 1 was control. Animals in group 3 was dewormed (Stankiewich et al., 1992) with mebendazole while artificial infestation. Eggs were collected from *Ascaris suum* female uterus, obtained after slaughter animals (Eriksen, 1990), then were cultured in plain water at temperature of 25⁰C.
Biochemical determinations were aimed at evaluating: AST, ALT, PAL, GGT, total plasma protein, albumin, δ globulins and was performed using UV-VIS Spectrophotometer MASTER +. Haematological examination was performed using Abacus Junior hematology analyzer. In terms of assessing immunity tests were performed on non-specific cellular immune effectors highlighting: total leukocyte count, total eosinophils, white cell count, phagocytic activity, immunological tests on some restructuring (determination of circulating immune complexes), and total immunoglobulin dosing was mediated immune reactivity specific measured by assessing the ability of sensitized leukocytes for blastisation. In the histopathological examination swine were slaughtered on day 56 pi, were harvested organ portions were then stained using Goldner’s trichrom.

Results

Fig. 4 Ascaris suum females used for egg collection

Fig. 5 Harvesting eggs A. suum

Fig. 6 Growing eggs of A. suum
In infected and untreated group in a number of three pigs (60%) were diagnosed with Ascaris suum infestation. Infestation intensity in this group averaged 150 ± 141.42 EPG. Biochemical investigations revealed that serum levels of ALT, GGT, total protein, δglobulins, ranged from physiological limits of species and at the infected and untreated group was found 14 days pi an increase of plasma AST (p <0.005) to 59.8U/L and PAL up to 480 U/I, (max. 395 U/I) (p> 0.05).

**Fig. 7** Variation of AST means

**Fig. 8** Variation of PAL means
Values of erythrocytes parameter were employed between the reference limits, with low differences between the three groups. In terms of immunological *A suum* infestation has no significant effect influenced the specific or nonspecific mediated immunity. But there were differences in the formation of circulating immune complexes (p > 0.05) were insignificant but an increase in total immunoglobulin (p < 0.05).

**Fig. 9** Variata mediei imunoglobulinelor totale

Histopathological investigations performed in *Ascaris suum* experimental infestation have highlighted the following aspects: in the gut: edema, denudation and villous tip necrosis in the area of contact with the parasite and formation of detritus as a barrier between the mucous and nematode. The *A. suum* larvae migration in the liver causes important changes affecting the structure and functions, it tracks presenting traumatic polymorphic and disorganized proliferating lobules of different intensities conjunctiva, marked eosinophilic infiltration, in the lung lesions consist of infiltration peribronchial; disappearance of zonal bronchial epithelium, detritus in the lumen bronchiolus moderate, zonal loss of muscle (in the place where the larvae entered through the wall).
**Fig. 10** Experimental group artificially infested with *A. suum* -gut. (Goldner’s Trichrom, ob.4X): a) parasite cuticle; b) the internal organs of the parasite, C) gut villi

**Fig. 11** Experimental group artificially infested with *A. suum* -gut. (Goldner’s Trichrom, ob.40X): a) parasite cuticle b) detritus-looking barrier, c) peak villous denudation.

**Fig. 12** Experimental group artificially infested *A. suum* -liver. (Goldner’s Trichrom, ob.40X): a) traumatical duct after larval migration, b) extravasated erythrocytes, c) hepatocyte apoptosis
**Fig. 13** Experimental group artificially infested with *A. suum* -liver. (Goldner’s Trichrom, ob.20X): a) small groups of hepatocytes, b) eosinophilic infiltration

**Fig. 14**. Experimental group artificially infested with *A. suum* -liver. (Goldner’s Trichrom, ob.40X): a) collagen fibers, b) blood vessel, c) fibroblast

**Fig. 15** Experimental group artificially infested with *A. suum* -lung. (Goldner’s Trichrom, ob.40X): a) detritus in the lumen bronchiolus, b) bronchial epithelium detached as flaps
CHAPTER 4. ASCARIS SUUM NATURAL INFESTATION EFFECTS ON FOOD DIGESTIVE PROCESSING AND DIGESTIBILITY IN PORK

Investigations conducted under this chapter were aimed at evaluating the effects of the natural infestation with *Ascaris suum* in pigs on digestive processing and food recovery.

**Material and methods.**
The study was conducted during July-August 2009 on a total of 10 pigs castrated males divided into two groups (n1 = 5, n2 = 5) of the hybrid race with the age of 5 months. Pigs were housed individually, using silicon dioxide (SiO$_2$) as an indicator to the digestive inert. Gross chemical composition was determined feces and fodder using Weende methodology and silicon dioxide (SiO$_2$) solubilisation method of ashes in hydrochloric acid.

**Results**

![Graph](image)

**Fig. 17.** The percentage of silica determined from faecal samples and feed.
Coefficient values of the digestive use of dry matter, crude protein, crude fat, crude cellulose, nitrogen free extract and crude ash were higher in control group compared with infested lot.
ADC for crude fat had a higher value in the control group compared to infected group, the difference being significant statistic (p <0.01).

Ascaris suum natural infestation has a direct influence on the weight of pigs, the control group with an increase of growth compared with controls, but with uninsured mathematical difference (p>0.05).