# STUDIES ON THE EFFECT OF INFECTION WITH IRRADIATED NEOASCARIS VITULORUM ON THE WHITE RAT

## THESIS

Presented to the Zoology Department

Faculty of Science

Ain Shams University

# By

MONA MOHAMED AMIN (B.Sc.)
Department of Radiation Research
for Health

National Center for Radiation Research

and Technology

Atomic Energy Authority,

Cairo-Egypt

29 h32

In Partial Fulfilment of the Requirements for the Degree of

MASTER OF SCIENCE

1988

<

#### Board of Scientific Supervision

- Dr. Gamal Mohamed Edris Abu Sinna,
   Professor of Physiology, Department
   of Zoology, Faculty of Science,
   Ain Shams University.
- Dr. Mohamed Abd El-Hamid Mansour,

  Head of the Radiation Research

  Department for Health, National

  Center for Radiation Research and

  Technology.
- Dr. Amin Abd El-Baki Ashour
   Lecturer of Parasitology, Department
   of Zoology. Faculty of Science.
   Ain Shams University.



### CONTENTS

|                 | F  | age |
|-----------------|--|-----|
| ACKNOWLEDGMENT  |  |     |
| CHAPTER I : .:  | INTRODUCTION                                 | 1   |
| 1 -             | - Aim of the present work                    | 2   |
| CHAPTER II : I  | Review of literature                         |     |
| 1-              | - Historical Review                          | 3   |
| 2-              | - Induction of parasitic immunity            |     |
|                 | by irradiation                               | 8   |
| 3-              | - Effect of helminth parasites on            |     |
|                 | host blood                                   | 18  |
| CHAPTER III : N | Material and Methods:                        |     |
| 1-              | - Experimental animals                       | 26  |
| 2-              | Preparation of infective                     |     |
|                 | N. vitulorum stage                           | 26  |
| 3-              | Infection of rats                            | 27  |
| 4-              | Haematological studies                       | 28  |
| 5 -             | Alkaline phosphatase activity                | 30  |
| 6-              | Determination of Transminases                |     |
|                 | activities                                   | 30  |
| 7               | Immunological studies                        | 33  |
| 8-              | Statistical analysis                         | 40  |
| CHAPTER IV : Re | sults:                                       |     |
| 1 –             | The effect of irradiated and non-            |     |
|                 | irradiated <u>N.vitulorum</u> infective eggs | В   |
| Central I       | on the blood picture of the rat              | 42  |

|         |     |   | 2- Serum alkaline phosphatase activity. § | 5.5 |
|---------|-----|---|---|-----|
|         |     |   | 3- Serum Transaminases activities 5       | 56  |
|         |     |   | 4- The effect of irradiated and non-      |     |
|         |     |   | irradiated N. $vitulorum$ infective       |     |
|         |     |   | eggs on the immunological response        |     |
|         |     |   | of the rat: 6                             | 50  |
|         |     |   | - Ciramoval Precipitin Test 6             | 50  |
|         |     |   | - Immunodiffusion Test 6                  | 9   |
|         |     |   | - Ring Precipitin Test 7                  | 7   |
| CHAPTER | v   | : | DISCUSSION                                | 10  |
| CHAPTER | VI  | : | SUMMARY9                                  | 12  |
| CHAPTER | VII | : | REFERENCES 9                              | 5   |
|         |     |   |   |     |

ARABIC SUMMARY

#### Acknowledgment

I am deeply grateful and indebted to Professor Dr. Gamal Abu Sinna, Professor of Physiology, Department of Zoology, Faculty of Science, Ain Shams University, for his kind supervision and for his valuable criticism, planning this study and reading the manuscript. Without his guidance, this work would not have been accomplished. To him I owe more than can be expressed.

I would like to express my sincere gratitude and deeply thanks to Dr. Mohamed Abd El-Hamid Mansour, Head of the Radiation Research Department for Health, National Center for Radiation Research and Technology, for suggesting and planning this study, supervising the work and reading the manuscript, and for providing me with all the facilities for the accomplishment of this work.

My deep gratitude and sincere thanks are due to Dr. Amin Abd El-Baki Ashour, Lecturer of Parasitology, Department of Zoology, Faculty of Science, Ain Shams University for planning of this study, helpful advice, revising the manuscript and for his kind encouragement.

I offer my gratitude and thanks to Professor Dr. Hamid Roushdy El-Kady, Chairman of Atomic Energy Establishment not only for suggesting the subject, but also for the great guidance and help to overcome many of the difficulties that arose in the course of this research. Also my best thanks to Chairman and all the staff of N.C.R.R.T. for providing facilities during this work.

However, I would like to express my deeply thanks for each of the members of Zoology Department, Faculty of Science, Ain Shams University. Also, My best thanks for Radiation Research Department for Health, for their helps.

CHAPTER I

#### Introduction

Buffaloes are known to be of most economic importance in Egypt for production of milk and meat well as for working in farms. Since long time ago, parasitic infestations are considered to be the main of low productivity of animals. cause Neoascaris vitulorum is one of the most dangerous intestinal parasites specially affecting buffalo tropical and subtropical countries. It is considered to be the only ascarid that occur in cattle. The different ascarids like most nematodes possess a developmental migratory route through most of the vital organs and tissues in the body of the host, before reaching the sexually mature stages in the intestine. Infestation with gastrointestinal worms often cause outbreaks of diseases producing a continual source of economic loss.

Recently, there is a great interest in the feasibility of using ionizing radiations as a possible mean of reducing or controlling parasitic diseases of animals. One of the developing and hopeful routes of vaccination is to use feebly irradiated parasite as a vaccinating antigen. However, it is of importance to make full understanding of the antigenic components of the parasite as well as of the antibodies raised against the introduced parasitic antigens.



#### Aim of work

It was aimed in the present study to have a good understanding of the effect of irradiated N. vitulorum infesting eggs on some haematological and immunological aspects of the host. The white rat was used as a small laboratory animal model capable of being infected with the parasite. The infective eggs were irradiated with three doses of gamma irradiations and comparison was carried out using rats infected with non-irradiated eggs as well as rats which were not infected at all. The aspects studied included red blood cells count (RBC), blood haemoglobin level (HB), haematocrit value (PCV), white blood cells count (WBC), differential white blood cells, count of mean corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV), the serum activities of the enzymes glutamate pyruvate transaminase (GPT), glutamate oxaloacetate transaminase and alkaline phosphatase. immunoprecipitation techniques were used in an attempt for testing the pattern of the evoked immunity in the sera of rats infected with the three doses irradiated N. vitulorum eggs.

# CHAPTER II

#### Historical Review

Although Neoascaris vitulorum (Goeze, 1782; Travassos, 1927) is regarded as one of the most important and common helminth parasites of cattle and buffaloes in tropical and subtropical countries, yet the literature dealing with its biology is still lacking. However, the biology of N. vitulorum has been taken into consideration by Tubangu (1947) and de Leon & Juplo (1966) in Philippines, Gadzheiv (1951) and (1957) in U.S.S.R., Sarwar & Nawaze (1951) in Pakistan, Sinniah (1954) in Ceylon, Lancaster (1958) in Malaysia; Serivastava (1963) in India, Bikov (1965) in the Italian region, Selim & Tawfik (1966) & (1974) in Egypt and Chaudhri & Riaz (1984) in Pakistan. Mohan (1968) reviewed most of the reports concerning the biology of N. vitulorum.

This parasite has been previously described under the name <u>Ascaris vitulorum</u> Goeze (1782) by Neuman (1883), Ransom (1911), Baylis & Daubney (1922), Boulenger (1922) and Macfie (1922). Later on, the genus <u>Neoascaris</u> was proposed to be separated from genus <u>Ascaris</u> by Travassos (1927) on account of the presence of a ventricles or granular bulb at the posterior end of the oesophagus in the bovine <u>Ascaris</u>.



Although this species was described once more by Baylis (1936), yet the same author used the nomenclature Ascaris vitulorum again without consideration of the presence of the ventricles as a reasonable cause for establishing the new Skryabin & Karokhin (1945) separated the genera which possess a posterior granular ventricles at the base of the oesophagus from family Ascarida to a new family which was mentioned family Anisakidae to which genus Necesscaris was included. This family had been amended by Hartwich (1954) and restricted to the forms having a uniramous excretory system. The author proposed that ascarids possessing a biramous excretory system, having an oesophagus with a posteriorly globoid or spherical part without appendix and with lips possessing labial wings should be included in a separate family which was named Toxocaridae. Consequently, it was proposed that genus Necascaris should be transferred to the family Toxocaridae, as its numbers possess the above mentioned characters.

Mozgovoi (1952) summarized the various ascarids in relation to the route of migration in the vertebrate hosts as follows:

1- Ascaridioid, represented by <u>Ascaridia galli</u> in which the larvae do not migrate via the blood system of the host.

- 2- Ascarioid, represented <u>Ascaris lumbricoides</u> in which the larvae have a hepato-pulmonary migration.
- 3- Toxcocaroid, represented by <u>Toxocara</u> and <u>Neoascaris</u> in which the larvae migrate from the pregnant animal to the foetus via the placenta and become adult shortly after its birth.
- 4- Anisakoid, represented by <u>contracaecum</u> and <u>Pseudoanisakis</u> in which larval development occurs in an intermediate host.

Sprent (1952) classified these ascarids into 2 types. The author's idea was based on the migratory paths following the infection of white mice with the larvae:-

The first group includes those parasites which adopt the tracheal pattern (Ascaris lumbricoides and Parascaris equorum) where the larvae penetrate the intestinal wall passing via the liver to the lungs. In the lung they break out into the bronchial tree and return to the intestine where they grow to maturity. Consequently, infection with those worms is invariably acquired by postnatal infection of the host. The second group follows a somatic course. This route is known for Toxocara canis and Ascaris columnaris.

Sprent (1954) reported a 3<u>rd</u> type in which the larvae develop in the intestinal wall without any further need to migrate through the tissues. This type

is represented by <u>Toxascaris</u> <u>leonina</u> and <u>Toxocara cati</u>.

Furthermore, Lee (1958) and Soulsby (1965 & 1968) criticized the fore-mentioned groupings. They indicated that the developmental cycle of <u>N</u>. <u>vitulorum</u> follows the same route as <u>Toxocara canis</u> and its larvae undergo the somatic type of migration.

Enyenihi (1971) and Chauhan & Pande (1972) studied the migration of N. vitulorum larvae in Guinea pigs and calves. The 2nd stage larvae were found in the liver, heart, lungs and kidney one day after Guinea pigs infection. The lungs contained the 2nd stage larvae of increasing size up to the 6th day of infection where they multimoult developing the 3rd-stage larvae. A peak larval count was recorded on the 10th day infection them the count decreases until there was non on the 30th day. No 3rd stage larvae were found in the liver, heart, kidneys. On the 6th day, the 3rd stage larvae developed in the lungs start to break into the alveoli and migrate through the bronchioles, up to the trachea and into the oesophagus. The 3rd stage larvae were found in the intestinal contents from the 6th day. Those larvae which reached the intestine of both Guinea pigs and calves are expelled in the faeces. Nο patent period was observed for these larvae.