CHAPTER 4:
CLASSIFICATION OF THE POLYSTOME DIGESTIVE TRACTS

4.1. Introduction

Monogenean flatworms exhibit two distinct diets, resulting in their subdivision into two subclasses, namely Polyopisthocotylea and Monopisthocotylea, which differ in their biology. The structure of the haptor also plays a role in this subdivision (Arafa *et al*., 2013; Bentz *et al*., 2003; Watson & Whittington, 1996).

Species from Monopisthocotylea are mobile over the surface of their hosts and mainly feed on the mucus as well as on the host epithelial tissues (Watson & Whittington, 1996). The ultrastructure of the gastrodermis of members of the Monopisthocotylea is simpler (continuous), exhibiting a single cell type. Haematin is absent in the gut of members of the Monopisthocotylea (Rubio-Godoy, 2007).

In contrast species from Polyopisthocotylea are less mobile and primarily feed on blood (sanguinivorous diet) (Watson & Whittington, 1996). The ultrastructure of the gastrodermis of members of the Polyopisthocotylea is more complex (discontinuous), exhibiting two cell types, namely the connecting syncytium and the digestive cells (Rubio-Godoy, 2007).

Both Monopisthocotylea and Polyopisthocotylea monogeneans demonstrate a penchant for attaching to specific sites on the host species. This reveals recognition by the parasites for variation in microhabitat conditions. In particular Polyopisthocotylea monogeneans have been shown to have species-specific differences in the distribution on the host (Perkins *et al*., 2010; Rubio-Godoy, 2007).

Monogenean parasites do not directly result in host mortality, instead host mortality is indirectly caused by secondary infections that are associated or caused by monogeneans. Thus Monopisthocotylea and Polyopisthocotylea also differ in the nature of the pathological damage they wreak on their hosts (Arafa *et al*., 2013; Bentz *et al*., 2003; Perkins *et al*., 2010; Rubio-Godoy, 2007; Watson & Whittington, 1996).

The feeding behaviour and activity of monopisthocotyleans can result in the epithelium being eaten away to the bone. Additional damage caused by the feeding habits of monopisthocotyleans is the creation of small puncture wounds, caused by the marginal hooklets being inserted into the epithelium that may aid bacterial and other opportunistic infections. These infections can lead to more serious injury such as kidney damage and/or systemic osmoregulatory problems (Rubio-Godoy, 2007).

Polyopisthocotyleans are not visibly pathogenic to their hosts, partly because the attachment method (structure of the haptor) of these monogeneans differs from that used by monopisthocotyleans. Polyopisthocotyleans use clamps that grasp the tissues of the host, instead of piercing the host by means of anchors or marginal hooks (Rubio-Godoy, 2007).

However as they are blood feeders, they are more likely to cause internal damage to their host including anaemia and potentially host death. They may also cause damage to the gills of their host,

including aneurysms, haemorrhage, loss of lamellar structure, epithelial hyperplasia as well as secondary infections by fungi and/or bacteria (Arafa *et al*., 2013; Rubio-Godoy, 2007).

Polystomatidae parasites are all similar in appearance however, there are various characteristics with which the genera and subsequently the species can be distinguished from one another, including the size, shape, position and/or number of hamuli, ovary, uterus, testes, pharynx, suckers and hooks among others. There is also variation in the form and shape of the digestive tract, particularly in the shape of the caeca. Variation in the intestinal tract can also be found in the presence/absence and number of medial and lateral diverticula. Another important characteristic to consider is the presence/absence and number of anastomosis.

Thus the type of digestive tract, particularly if the shape of the digestive tract is specific to certain polystomes, can be used as an important taxonomic characteristic, as the intestinal tract can aid in the identification and classification of polystome species into specific genera. Thus the type of intestinal tract present in a species can be used as a classification tool by classifying intestinal tracts based on shape.

4.2. Methodology

The different forms of the polystome digestive tracts were studied from all available literature. Particular focus was placed on the shape of the caeca and the number, length and presence or absence of the medial and lateral diverticula as well as anastomoses. A series of intestine types were tabulated from the most simple to the most complex. Included in the table is a short description of each digestive tract type as well as a list of the appropriate genera.

Aspects such as the feeding habits of the parasites as well as the degree of branching (number of anastomes), were also researched to determine if they play a role in determining which polystome species contain which form of the different digestive tracts that occur in the Polystomatidae family. Within a species there is variation in the number of anastomes and this was taken into account.

4.3. Results and Discussion

The results of this study have been tabulated, from the most simple intestinal tract form to the most complex, with a description of each as well as the various polystome genera in which the specific intestinal tract occurs. Thus, the following table can be used as a classification tool by classifying the different intestinal tracts of the 24 Polystomatidae genera based on shape.

Oculotrema Stunkard, 1924 is the single mammalian Polystomatidae genus that has a **Type A** digestive tract, containing unequal length intestinal caeca. The caeca are not diverticulated with no intestinal anastomoses (Stunkard, 1924).

Neopolystoma Price, 1939, *Parapolystoma* Ozaki, 1935, *Polystomoidella* Price, 1939, *Polystomoides* Ward, 1917, *Nanopolystoma* Du Preez, Wilkinson & Huyse, 2008, *Pseudopolystoma* Yamaguti, 1963,

all have a **Type B** digestive tract containing caeca of equal length that are not confluent at the posterior end of the body. The caeca are unbranched (lack diverticula) and no anastomosis were observed (Du Preez *et al*., 2007; Du Preez *et al*., 2008; Uchida & Itagaki, 1979; Price, 1939; Vande Vusse , 1976; Lim & Du Preez, 2001). However the caeca of *Parapolystoma* may or may not contain diverticula (Price, 1939; Vande Vusse, 1976).

Concinnocotyla Pichelin Whittington & Pearson, 1990 and *Sphyranura* Wright, 1879 both have a **Type C** digestive tract that is an elongate loop extending from the pharynx nearly to the haptor, with the caeca posteriorly rounded and lacking diverticula as well as anastomosis. There are two Type C digestive tracts that are identical, the only difference is the number of suckers present, *Concinnocotyla* contains four suckers, while *Sphyranura* only has two (Alvey, 1936; Hughes & Moore, 1943).

Diplorchis Ozaki, 1931, *Sundapolystoma* Lim & Du Preez, 2001, *Eupolystoma* Kaw, 1950, *Kanaka* Raharivolololniaina, Verneau, Berthier, Vences & Du Preez, 2011 and *Neodiplorchis* Yamaguti, 1963 all have a **Type D** digestive tract that is an elongate loop extending from the pharynx into the haptor. The caeca are rounded posteriorly and may or may not contain small diverticula (Du Preez *et al*., 2003; Raharivolololniaina *et al*., 2011; Kaw, 1950; Tinsley, 1978a).

Madapolystoma Du Preez and *Metapolystoma* Combes, 1976 have a **Type E** digestive tract which bifurcates with the caeca converging posteriorly. Medial diverticula are present, while anastomoses are absent (Berthier *et al*., 2014).

Neoriojatrema Imkongwapang & Tandon, 2010 contains a **Type F** digestive tract which bifurcates with intestinal caeca diverticulating in both lateral and marginal aspects and lacking prehaptoral anastomoses (Imkongwapang & Tandon, 2010).

Mesopolystoma Vaucher, 1981, *Parapseudopolystoma* Nasir & Zambrano, 1983*, Pseudodiplorchi* Yamaguti, 1963 and *Riojatrema* Lamothe-Argumedo, 1964 all contain a **Type G** digestive tract is confluent posteriorly, with the caeca diverticulating to form prehaptorial as well as haptorial anastomosis (<5). Short lateral diverticula and a network of medial diverticula are present (Biserkov & Hadjinikolova, 1993; Nasir & Zambrano, 1983; Imkongwapang & Tandon, 2010; Lamothe-Argumedo, 1964).

Protopolystoma Bychowsky, 1957 contains a **Type H** digestive tract which lacks intestinal anastomoses between the caecal terminations but contains prehaptoral and terminal anastomoses. This digestive type is similar to that of Type G but contains longer lateral diverticula, a larger number of medial diverticula and may not always merge in the haptor (Tinsley & Jackson, 1998).

Wetapolystoma Gray, 1983 has a **Type I** digestive tract that bifurcates and extends from the pharynx into the haptor. The caeca are rounded posteriorly, forming a number of lateral diverticula as well as a network of medial diverticula that merge to form several (>5) anastomosis (Gray, 1993; Lamothe-Argumedo, 1986).

The digestive tract of *Polystoma* Zeder, 1800 is with or without prehaptorial anastomoses (Price, 1939; Biserkov & Hadjinikolova, 1993; Diengdoh & Tandon, 1991). Thus this genus contains species with varying digestive tract types, typically Types E-I, with Type G occurring more frequently. The digestive tract is more complex in some *Polystoma* species such as in *Polystoma makereri* which appears to be Type I, while in others it is simpler such as in *Polystoma gallieni* which appears to be Type E.

Table 4.1: The different Polystomatidae digestive tract types from simple to complex

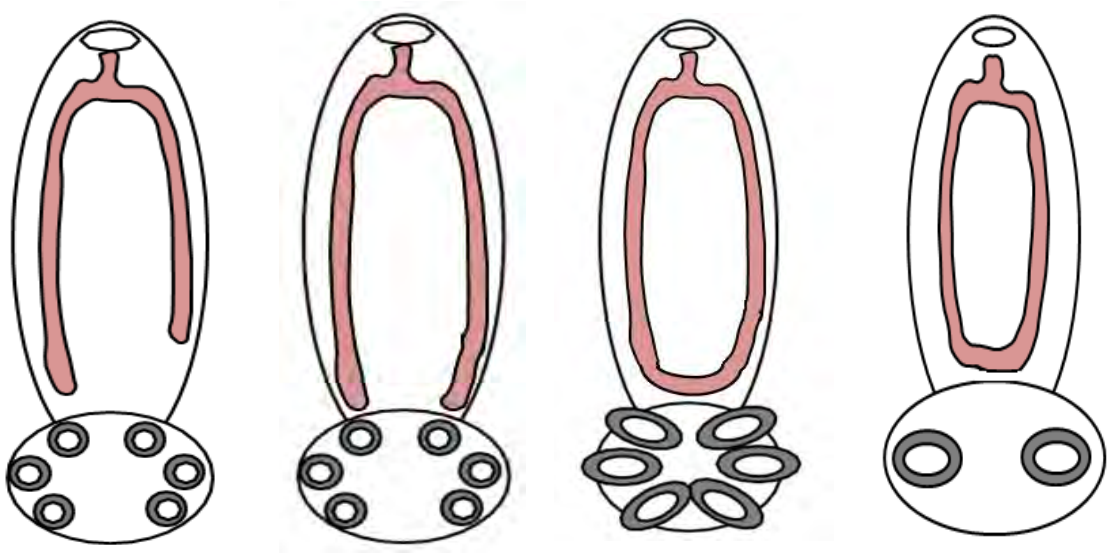
Digestive tract type	Type A	Type B	Type C
Figure			
Description	Digestive tract with unbranched caeca of uneven length.	Digestive tract with unbranched caeca of equal length.	Digestive tract with unbranched caeca that merge posteriorly near the haptor.
Genera	*Oculotrema*	*Nanopolystoma* *Neopolystoma* *Parapolystoma* *Polystomoidella* *Polystomoides* *Pseudopolystoma*	*Concinnocotyla* *Sphyranura*

Table 4.1 continued

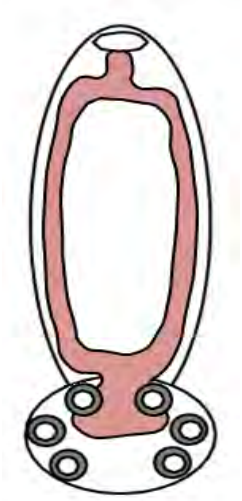
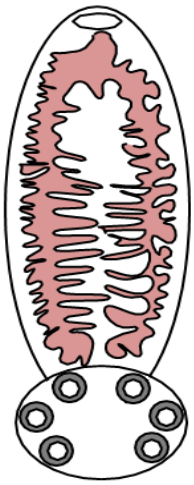
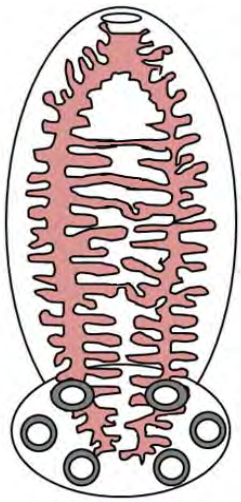
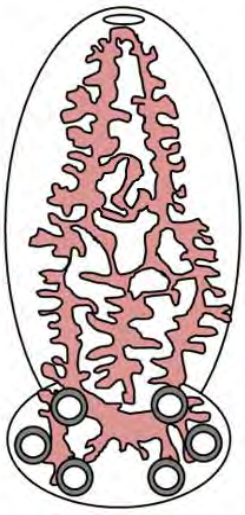
Digestive tract type	Type D	Type E	Type F
Figure			
Description	Digestive tract with unbranched caeca that merge posteriorly and extend into haptoral region.	Digestive tract with caeca, extending into haptor, that branch forming a number of medial diverticula.	Digestive tract with caeca that branch forming short lateral diverticula and a network of medial diverticula.
Genera	*Diplorchis* *Eupolystoma* *Kankana* *Neodiplorchis* *Sundapolystoma*	*Madapolystoma* *Metapolystoma* *Polystoma*	*Neoriojatrema* *Polystoma*

Table 4.1 continued

Digestive tract type	Type G	Type H	Type I
Figure			
Description	Digestive tract with caeca, extending into haptor, that branch forming short lateral diverticula and a network of medial diverticula, resulting in a number (<5) of anastomosis.	Digestive tract with caeca, may extend into haptor, that branch forming a number of lateral diverticula and a network of medial diverticula, resulting in a number (<5) of anastomosis.	Digestive tract with caeca, extending into haptor, that branch forming a number of lateral diverticula and a network of medial diverticula, resulting in a number (>5) of anastomosis.
Genera	*Mesopolystoma* *Parapseudopolystoma* *Polystoma* *Pseudodiplorchis* *Riojatrema*	*Polystoma* *Protopolystoma*	*Polystoma* *Wetapolystoma*

Note that the figures are only interpretations of the various polystome figures. Variations may occur for certain characteristics, such as the size and shape of the intestinum as well as the amount of diverticula and anastomoses present. Mucus feeders have simpler digestive tract forms, while those with a sanguiforous diet have more complex intestines (Aisien & Du Preez, 2009; Du Preez & Moeng, 2004; Du Preez, Wilkinson & Huyse, 2008; Watson & Whittington, 1996).

This statement is supported by various studies previously done on polystomes, for instance Platt (2000b) stated that the digestive tract of the mucus feeding *Neopolystoma fentoni* found in a freshwater turtle from Costa Rica lacks diverticula and anastomoses making the digestive tract more simple while Bourgat & Murith (1980) showed that the digestive tracts of the blood feeding *Polystoma lamottei* and *Polystoma aeschlimanni* contain both diverticula as well as anastomoses, thus their digestive tracts are more complex.

Stunkard (1916) explained that species in the genus *Polystoma* show greater structural variation, particularly in the digestive tract and reproductive system. For example the caeca of *Polystoma integerrimum* is greatly branched (with anastomoses) throughout the body and haptor. However, the caeca of *Polystoma claudecombesi* has less branching and no anastomoses (Du Preez & Kok, 1995).

Thus the diet of a polystome does play a role in the type of digestive tract. If the parasite feeds on epithelial cells and/or mucus, then the digestive tract is most likely Types A-D, while those that feed on blood contain digestive tract Types E-I. It also appears as if the degree of branching (number of anastomes) plays a role in determining the digestive tract type as the greater the degree of branching, the more complex the digestive tract is such as that of Type I.