Metazoan parasite survey of selected macro-inshore fish of southeastern Australia, including species of commercial importance

FINAL REPORT
FRDC Project No. 2007/225
KS Hutson, SR Catalano & ID Whittington
28 February 2011

Australian Government
Fisheries Research and Development Corporation

JAMES COOK UNIVERSITY AUSTRALIA

South Australian Museum

THE UNIVERSITY OF ADELAIDE AUSTRALIA

Australian Government
Department of the Environment, Water, Heritage and the Arts

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Cover image: A parasitic isopod, *Ceratothoa* sp., from the mouth of silver trevally *Pseudocaranx georgianus* captured in Coffin Bay, South Australia. Image provided by Brian Saunders.

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Non-technical Summary

Parasites have the potential to limit the growth of Australian fishing industries, especially aquaculture, through mortality, morbidity and reduced marketability. A majority of the parasites of recreational, commercial and farmed Australian finfish has not been collected, studied or described. We surveyed 12 important finfish species and documented their parasite assemblages, placing emphasis on parasitic crustaceans (e.g. sea-lice) and helminths (e.g. flukes). Morphological methods and, in some cases, molecular tools were used to facilitate parasite identification. More than 120 parasite species were identified. Parasites were used as biological tags to identify geographic population structure in one commercial fish species, the southern garfish. We assessed parasite risks to sea-cage aquaculture for two species of finfish in Australia, mulloway and barramundi, and indicate appropriate methods to adopt in animal husbandry in the event of parasite outbreaks in mulloway and barramundi culture which will help improve the viability of the industries. A comprehensive, user-friendly, richly illustrated website (MarineParasites.com) has been created that details parasite biology, pathology and host-specificity, enabling lay people to identify different types of parasites in common fish species encountered in Australia.

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1 GENERAL INTRODUCTION

1.1 Background

The oceans of Australia have the greatest diversity of marine fishes in the world. The parasite communities that infect individual fish species are also incredibly diverse, but are extremely poorly studied in Australia. Identification of parasites infecting recreational, commercial and cultured finfish is important to determine fish stock structure, provide import and export risk assessments and to deliver appropriate parasite management in aquaculture. However, taxonomic research on parasites of finfish has been largely limited to certain centres on the east coast, often related to where parasitologists have been employed. The Marine Parasitology Laboratory at The University of Adelaide enabled direct access to the southeast region of the continent, providing an unparalleled opportunity to study metazoan parasites of fishes where the vast majority of species are unknown. Relocation of the PI to James Cook University, Queensland, in the final year of the project enabled further examination of important tropical species.

We chose 12 key native fish species to undertake a parasite survey in southern region of Australia, including barramundi in the northeast. The survey included recreational and commercial species, candidate species for aquaculture and currently farmed species. All metazoan parasites encountered were collected and identified to genus or species where possible, to compile taxon lists with biogeographical and biological relevance. Parasites and some representative fish specimens and also tissue samples were lodged and are now curated in Museum Victoria, the Natural History Museum, London and the South Australian Museum for future study. Although we recovered a broad range of parasite species, we concentrated our taxonomic work on copepod, monogenean and aporocotylid trematode species (for which the research team has particular expertise). These parasite groups are also frequently associated with pathology, morbidity and/or mortality in finfish aquaculture and can cause mortalities in natural fish populations.

1.2 Need

Metazoan parasites threaten the sustainability and profitability of the Australian finfish aquaculture industry. It is critical, therefore, to collect and identify local parasite species and determine which are potentially harmful. In the northern hemisphere, where intensive finfish aquaculture began in the 1960s, there have been many studies assessing metazoan parasite fauna of wild fish and fish farmed in sea-cages. In Australia such research is rare. The potential for parasitic disease problems in sea-cage aquaculture has not yet been fully realised in Australia because current stocking densities are low, farm locations are dispersed and the industry cannot be considered to be as intensive.

In the sea-cage environment, farmed fish may acquire parasite infections found in local populations of wild fish. The occurrence of wild fish near sea-cage farms provides an opportunity for transfer of parasites between wild and farmed populations. The parasite assemblages of wild fish species and the potential risks of these parasites for sea-cage aquaculture are largely unknown. By gathering fundamental biological data about parasites, we will gain a better understanding of how to manage and control them on fish in captivity.

Knowledge of parasite biology, how parasites impact on their hosts and development of diagnostic tools are critical for effective parasite management. This project used a powerful combination of morphological and, in some instances, molecular techniques to provide a comprehensive understanding of copepod, monogenean and aporocotylid trematode parasites infecting recreational, commercial and cultured finfish species in southeastern Australia, plus barramundi (*Lates calcarifer*) in the northeast. A combined morphological and molecular approach will resolve questions that are unable to be addressed using only one approach.

Accurate species identification is essential to understand biodiversity, biogeography, biosecurity, biology, ecology, conservation and animal health. Parasite species and assemblages are a valuable tool to determine geographic population structure in wild fish and to identify appropriate fishery management units (MacKenzie 1999; MacKenzie 2002; Braicovich and Timi 2008). The capacity to correctly identify parasitic agents of fishes is also essential for fisheries

management (Barber *et al.* 2000) and maintenance of biosecurity. If a parasite species causing disease is not identified accurately, it may be difficult to develop control and treatment methods and assess risk factors (e.g. Young *et al.* 2007). Several parasite species mistakenly combined as one species can result in incorrect estimates of diversity (Poulin and Morand 2000). We highlight the relevance of thorough morphological taxonomy to applied science and emphasise its use as a fundamental scientific tool in all aspects of our research methodology.

1.3 Objectives

For clarity, the original 10 objectives (presented in brackets) are reported under seven main chapters as follows:

Chapter 2: Metazoan parasites of important fishes in Australia

To complete a parasite survey of selected finfish species in Australia (1)

Chapter 3: Biogeographical relevance of parasite-host checklists: a case study

To provide a host-parasite checklist with biogeographical relevance (3)

Chapter 4: Parasite redecriptions

To provide taxonomic revisions of copepods, monogeneans and trematodes including redescriptions of poorly described taxa & description of new species (2)

Chapter 5: Morphological and molecular tools to distinguish blood fluke parasites

To provide taxonomic revisions of copepods, monogeneans and trematodes including redescriptions of poorly described taxa & description of new species (2)

To use DNA sequences to distinguish blood fluke species and enable diagnostic tools for industry (4)

Enable appropriate site selection for expansion of the industry away from infection sources (9)

Chapter 6: Potential parasitic threats to southern Australian finfish aquaculture

Identify parasites of potential threat to the sustainability of the sea-cage aquaculture industry (6)

Develop case studies of parasites of potential threat to aquaculture (7)

Chapter 7: Assessing parasite risk and identifying appropriate husbandry

Provide risk analyses for the parasites identified to understand a) the likelihood of parasite establishment and proliferation and b) the consequence of establishment and proliferation for sustainable aquaculture (5)

Identify parasites of potential threat to the sustainability of the sea-cage aquaculture industry (6)

Identify appropriate husbandry practices to manage and control parasite infections and thereby reduce morbidity and mortality in fish stocks (8)

Chapter 8: MarineParasites.com

Develop an interactive product (i.e. website) that enables rapid identification of marine parasite species for the public, recreational anglers and sea-cage aquaculture industry (10)

2 METAZOAN PARASITES OF IMPORTANT FISHES IN AUSTRALIA

2.1 Abstract

Accurate identification of fishes and their parasites is fundamental to the development, management and sustainability of fisheries and aquaculture worldwide. We examined a total of 29 fish species including currently farmed fish species, candidate aquaculture species and commercial and recreational species to determine their metazoan parasite assemblages and infection parameters. We identified more than 120 parasite species. Host tissue samples for fish studied and at least one voucher specimen of each parasite species were deposited in recognised curated museum collections. Although a range of parasite fauna was encountered, we placed taxonomic emphasis on copepod, monogenean and aporocotylid (blood fluke) species, because these parasite groups are frequently associated with pathology, morbidity and/or mortality in finfish aquaculture. During the course of this study, redescriptions have been provided for the monogeneans *Microcotyle arripis* Sandars, 1945 and *Kahawaia truttae* Dillon and Hargis, 1965 from *Arripis georgianus* and *A. truttaceus*, respectively and the copepod *Kabataia ostorhinchi* Kazatchenko, Korotaeva & Kurochkin, 1972 from knifejaw *Oplegnathus woodwardi* in Chapter 4. A new blood fluke (*Paradeontacylix* n. sp.) from *Seriola hippos* in described in Chapter 5.

2.2 Introduction

Accurate species identification is essential to understand biodiversity, biogeography, biosecurity, biology, ecology, conservation and animal health. For example, the European medicinal leech, used worldwide for surgery and gene regulation research, was recently identified as three species, not one (Siddall *et al.* 2007). There is now a need to reconsider decades of biomedical research on this widely used model organism. Furthermore the controversy about the identity of leeches potentially impacts regulatory laws and the conservation status of medicinal leeches (Kutschera 2006). Similarly, inadvertent grouping of separate fish species into a single species can have disastrous repercussions for commercially harvested fisheries. Apparent sustainability for a single, overexploited commercially fished species may be an illusion produced by

continued catches of a second morphologically similar but more fecund species (Dulvy and Reynolds 2009a).

The capacity to correctly identify parasitic agents of fishes is also essential for finfish aquaculture and maintenance of biosecurity. If a parasite species causing disease is not identified accurately, it may be difficult to develop control and treatment methods and assess risk factors (e.g. Young *et al.* 2007). Taxonomic rigour in the identification of parasites is also essential for fisheries management (Barber *et al.* 2000) because parasite species and assemblages are a valuable tool to determine geographic population structure in wild fish and to identify appropriate fishery management units (e.g. Chapter 3; MacKenzie 1999; 2002; Braicovich and Timi 2008). Several parasite species mistakenly combined as one species can result in incorrect estimates of biodiversity (Poulin and Morand 2000). Clearly, morphological taxonomy remains a fundamental scientific tool and is of immediate relevance to applied marine science. There is an increasing need to survey fishes and their parasite fauna across broad geographic ranges and identify them accurately to species.

Fundamental knowledge of parasite assemblages is missing for southern Australian fishes, including cultured and commercially and recreationally important species. The aim of this study was to discover and report metazoan parasite fauna of important fishes in southern Australian waters and barramundi in tropical waters, to provide comprehensive host and parasite identifications and deposit host and parasite voucher material in recognised curated museum collections.

2.3 Methods

2.3.1 Fish collections and deposition of fish tissue

Eleven fish species were targeted throughout southern Australia and barramundi (*Lates calcarifer*) was sampled in northeastern Australia (Table 1) with the aid of numerous stakeholders in the Australian fishing industry. Species were targeted because of their relative importance to Australian aquaculture, commercial and recreational fisheries (Table 1). Additional fish species were sampled opportunistically. Specimens were sought from collection

trips aided by the aquaculture industry (Bluewater Barramundi, Hinchinbrook, Queensland; Clean Seas Tuna, Port Augusta, South Australia; Clear Water Aquaculture, Boston Bay, New South Wales), commercial fishers (Innes Brothers Inc., Greenwell Point, New South Wales), fish markets (Safcol fish market, Mile End, South Australia; Sydney Fish Market, Pyrmont, New South Wales), fishing tournaments (Port Stephens Interclub Gamefishing Tournament, New South Wales; Port MacDonnell Tuna and Sportsfish Tournament, South Australia) and fishing charters (Alan Bevan, Shikari Charters, Fremantle, Western Australia). Tissue samples (fin clips) were collected from representative fish species, stored in 95% undenatured ethanol and lodged with the Australian Biological Tissue Collection at the South Australian Museum (SAMA), North Terrace, Adelaide, 5000, South Australia. Fish were identified using morphological descriptions and keys in Gomon *et al.* (2008).

Table 1. Fish species sampled for metazoan parasites in this study. *Indicates twelve original target fish species.

Fish Family Fish species	Common name	Importance
Arripidae		
*Arripis trutta***	eastern Australian salmon	Commercial/Recreational
*Arripis truttaceus***	western Australian salmon	Commercial/Recreational
*Arripis georgianus***	Australian herring	Commercial/Recreational
Carangidae		
Elagatis bipinnulata	rainbow runner	Recreational
*Seriola lalandi***	yellowtail kingfish	Aquaculture/Commercial/Recreational
*Seriola hippos***	Samson fish	Recreational
Seriola dumerili	greater amberjack	Candidate/Recreational
Centrolophidae		
Hyperoglyphe antarctica	deep sea trevalla	Commercial/Recreational
Cheilodactylidae		
Nemadactylus valenciennesi	queen snapper/blue morwong	Commercial/Recreational
Nemadactylus macropterus	jackass morwong/terakihi	Commercial/Recreational
Coryphaenidae		
*Coryphaena hippurus**	dolphin fish/mahi mahi	Candidate/Recreational
Hemiramphidae		
*Hyporhamphus melanochir***	southern garfish	Commercial/Recreational
Kyphosidae		
Girella tricuspidata	luderick	Commercial/Recreational
Latidae		
*Lates calcarifer**	barramundi	Aquaculture
Latridae		
Latris lineata	striped trumpeter	Candidate aquaculture
Mugilidae		
Aldrichetta forsteri	yelloweye mullet	Commercial/Recreational
Oplegnathidae		
Oplegnathus woodwardi	knifejaw	Recreational
Percichthyidae		
Polyprion oxygeneios	hapuku	Commercial/Recreational/Candidate
Polyprion americanus	wreckfish	Commercial/Recreational/Candidate
Platycephalidae		
Platycephalus aurimaculatus	flathead	Commercial/Recreational
Rachycentridae		
*Rachycentron canadum***	cobia	Aquaculture/Recreational
Sciaenidae		
*Argyrosomus japonicus**	mulloway	Aquaculture/Recreational
Scorpaenidae		
Helicolenus percoides	red gurnard perch	Recreational
Sillaginidae		
*Sillaginodes punctatus**	King George whiting	Commercial/Recreational
Sparidae		
Acanthopagrus butcheri	black bream	Commercial/Recreational
*Chrysophrys auratus**	snapper	Commercial/Recreational
Triakidae		
Galeorhinus galeus	tope/school shark	Commercial/Recreational
Mustelus antarcticus	gummy shark	Commercial/Recreational
Urolophidae		
Urolophus paucimaculatus	sparsely spotted stingaree	-

2.3.2 Fish necropsies

Fish were examined for ectoparasites by studying the exterior surfaces, including fins, inside the mouth, fin sulcus and branchiostegal membranes with the naked eye and microscope. The gills were removed, placed in seawater and examined under a dissecting microscope for ectoparasites. An incision was made along the belly of the fish and the pyloric caeca, intestine and stomach were removed and opened separately in saline solution. The solution was vigorously shaken to dislodge endoparasites, allowed to settle, then the supernatant was poured off and discarded and the remaining solution was examined under the dissecting microscope for endoparasites. The gall bladder was removed and opened in a cavity block before transfer of a small amount of the gall fluid onto a glass slide for examination under a compound microscope. After organ removal, the viscera, swim bladder, muscle and body cavity was examined for endoparasites. All cestode cysts detected were opened in freshwater or saline.

2.3.3 Parasite preservation, staining and mounting

Live digeneans were killed in boiling saline and both dead and alive digeneans and monogeneans were preserved in either 10% formalin or 95% AR grade undenatured ethanol (depending on the freshness of fish sourced, the state and type of parasites and whether molecular analyses were to be performed). Preserved parasites were washed twice in MiliQ water before being stained using Mayer's haematoxylin and destained in 3% HCl. Parasites were dehydrated in an ethanol series (70%, 90%, 95% and 100%) before being cleared in cedar wood oil and mounted ventral side up on a slide beneath a coverslip in Canada balsam. Mounted parasites were examined using a compound microscope to enable identification. Prior to morphological examination, caligids and nematodes were preserved in 70% ethanol then cleared in lactophenol. Cestodes were removed from their cysts before preservation and cleared in glycerol. Myxozoans were observed by examining the gall fluid under a compound microscope.

2.3.4 Parasite identification

Parasites were identified by morphological examination of whole mounted or cleared preserved material. Published records, keys in scientific papers and parasite diversity books assisted in identification, with distinctive characters being used to classify parasites to genus and species. In addition, voucher and type specimens were requested from national and/or international museums when required for comparison with the material collected in this study. Additional

parasite specimens from the target host species were examined from four museum collections including: The Natural History Museum, London; Australian Museum, Sydney; Museum Victoria, Melbourne; and the South Australian Museum, Adelaide (SAMA). Parasite material collected in this study is deposited in the Australian Helminthological Collection (AHC) and the Crustacean collection (C) at SAMA, the Parasitology Collection at the Queensland Museum (G), the Parasitic Worms Collection at the Natural History Museum (BMNH) and the United States National Parasite Collection (USNPC).

2.4 Results and Discussion

2.4.1 Fishes sampled

We examined a total of 1641 individuals from 30 fish species and 20 families for metazoan parasites (Table 2). Extreme care should be taken in the interpretation of these results where few fish were sampled. Low sample sizes can severely underestimate parasite diversity, especially for parasites that exhibit low prevalence.

Table 2. Fish species and number examined for metazoan parasites in this study. Accession numbers are provided for fish tissue supplied to the Australian Biological Tissue Collection at the South Australian Museum, Adelaide. *Indicates twelve original target fish species.

Fish family and species	Common name	No. examined	ABTC
Arripidae			
*Arripis trutta***	eastern Australian salmon	23	108509–108777
*Arripis truttaceus***	western Australian salmon	67	
*Arripis georgianus***	Australian herring	183	
Berycidae			
Centroberyx gerrardi	Bight redfish/red snapper	1	83993, 97459, 102996,
Carangidae			
Elagatis bipinnulata	rainbow runner	1	Not accessioned
*Seriola lalandi**	yellowtail kingfish	136	81991–82004, 82932–45, 84961-65, 81214, 81149, 108505
*Seriola hippos**	Samson fish	12	84002, 102982–102984
Seriola dumerili	greater amberjack	2	92925
Centrolophidae			
Hyperoglyphe antarctica	deep sea trevalla	3	97434–36
Cheilodactylidae			
Nemadactylus valenciennesi	queen snapper/blue morwong	16	83999, 97440–2, 97464, 103004
Nemadactylus macropterus	jackass morwong/terakihi	57	97450–53, 102998–9
Coryphaenidae			
*Coryphaena hippurus**	dolphin fish/mahi mahi	35	97431–2, 102985–7
Hemiramphidae			
*Hyporhamphus melanochir**	southern garfish	274	20763
Kyphosidae			
Girella tricuspidata	luderick	30	Not accessioned
Latidae			
*Lates calcarifer**	barramundi	26	Not accessioned
Latris lineata	striped trumpeter	2	102988–91
Mugilidae			
Aldrichetta forsteri	yelloweye mullet	120	Not accessioned
Oplegnathidae			
Oplegnathus woodwardi	knifejaw	3	97462, 103003
Percichthyidae			
Polyprion oxygeneios	hapuku	3	97427–9, 102992
Polyprion americanus	wreckfish	3	97430
Platycephalidae			
Platycephalus aurimaculatus	flathead	2	Not accessioned
Rachycentridae			
*Rachycentron canadum**	cobia	30	Not accessioned
Sciaenidae			
*Argyrosomus japonicus**	mulloway	20	Not accessioned
Scorpaenidae			
Helicolenus percoides	red gurnard perch	15	84001, 97456, 97465–6
Sillaginidae			
*Sillaginodes punctatus**	King George whiting	416	83884, 97448
Sparidae			
Acanthopagrus butcheri	black bream	7	Not accessioned
*Chrysophrys auratus**	snapper	141	82401
Triakidae			
Galeorhinus galeus	tope shark	2	Not accessioned
Mustelus antarcticus	gummy shark	10	97461, 103001
Urolophidae			
Urolophus paucimaculatus	sparsely spotted stingaree	1	Not accessioned

**Accession numbers requested from ABTC

2.4.2 Target fish species sampled

Arripidae

Arripis georgianus

Arripis trutta

Arripis truttaceus

The Arripidae is a commercially and recreationally important fish family endemic to southern Australian and New Zealand (NZ) waters. The family comprises a single genus *Arripis* which includes four species (Paulin 1993). Three species are important commercial and recreational fisheries in Australian waters: Australian herring or tommy rough (*Arripis georgianus*); eastern Australian salmon (*A. trutta*); and western Australian salmon (*A. truttaceus*). Approximately 3,000 tons of 'Australian salmon' (*A. trutta* and *A. truttaceus* combined) is taken commercially in Australian waters per annum, while ~100 tons of *A. georgianus* is captured commercially in South Australia (ABARE 2009).

Arripis georgianus and *A. truttaceus* each comprise a single stock and share the same single spawning ground off the south-west coast of WA (Fairclough *et al.* 2000; Jones 2008). *Arripis trutta* spawns in the Lakes Entrance area of Victoria and off Eden and Bermagui in southern NSW (Stanley and Malcolm 1977). The stock structure of *A. trutta* is currently unknown (Smith *et al.* 2008). Arripids also have a tendency to aggregate near structure (Dempster and Kingsford 2004b; Neira 2005a) and have been observed in, and around, sea-cages in South Australia (Hutson pers. obs.). Despite the economic and ecological importance of these three arripid species, few studies have examined the parasite species associated with each host.

We identified seventeen parasite species from *A. georgianus*, 20 from *A. truttaceus* and 12 from *A. trutta* (Tables 3–5) and recorded 35 new host-parasite records (Tables 3–5). Redscriptions are provided for two species of microcotylid monogeneans from the three arripid hosts examined in Chapter 4. The majority of crustacean parasite species found infecting the three *Arripis* spp. have been associated with mass mortalities in aquaculture overseas. The importance of this finding is discussed in Chapter 6.

Table 3. Metazoan parasite fauna from *Arripis georgianus* collected in this study and presented in previous literature. [A] = new host record; [B] = new Australian record; [C] = from current study. Abbreviations: SA, South Australia; WA, Western Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	Viscera	[A]	SA[C]	AHC 45416
Order: Tetraphyllidea Carus, 1863				
Type 1	Intestine	[A]	SA[C]	AHC 29765
Type 2	Intestine	[A]	SA[C]	AHC 29766
Tentaculariidae				
Nybelinia thyrsites Korotaeva, 1971	Intestine	[A]	SA[C]	AHC 29788
Digenea				
Acanthocolpidae				
Monostephanostomum georgianum Kruse, 1979	Digestive tract	Bray and Cribb (2002)	SA, SA[C]	AHC 29759–60
Monostephanostomum manteri Kruse, 1979	Digestive tract	Braicovich and Timi (2008)	SA, SA[C]	AHC 29761
Bucephalidae				
Telorhynchus arripidis Crowcroft, 1947	Digestive tract	[A]	SA[C]	AHC 29764
Hemiuridae				
Elytrophalloides humerus Bray, 1990	Stomach, intestine	[A]	SA[C]	AHC 29755–58
Erilepturus tiegsi Woolcock, 1935	Digestive tract	Bray (1990)	WA, SA[C]	AHC 29754 AHC 29789
Opecoelidae				
Pseudopecoeloides arripi Aken'Ova *et al*., 2009	Intestine, caeca	Aken'Ova *et al.* (2009)	WA, SA, SA[C]	AHC 29762–63
Monogenea				
Microcotylidae				
Microcotyle arripis Sandars, 1945	Gills	Sandars (1945)	WA, SA, SA[C]	AHC 29751–53 AHC 29883–84

				BMNH 2009.12.28.1–2 USNPC 102673.00– 102675.00
Nematoda				
Anisakidae *Hysterothylacium* sp.	Digestive tract	[A]	SA[C]	AHC 45417 – 18
Camallanidae Railliet and Henry, 1915 *Procamallanus* sp.	Digestive tract	[A]	SA[C]	AHC 45421
Philometridae *Philometra* sp.	Gonad	[A]	SA[C]	AHC 45422 – 23
Trichinellidae *Capillaria* sp.	Digestive tract	[A]	SA[C]	AHC 45419 – 20
Branchiura				
Argulidae *Argulus diversicolor* Byrnes, 1985	Skin	[A]	SA[C]	BMNH 2009.261
Copepoda				
Caligidae *Caligus punctatus* Shiino, 1955	Gills	Catalano and Hutson (2010)	SA[B, C]	C6814

Table 4. Metazoan parasite fauna from *Arripis truttaceus* collected in this study and presented in previous literature. [A] = new host record; [B] = new Australian record; [C] = from current study. Abbreviations: ND, not determined; SA, South Australia; TAS, Tasmania; VIC, Victoria.

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Ceratomyxidae				
Ceratomyxa sp.	Gall bladder	[A]	SA[C]	G465430–31
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	Viscera	Beveridge and Campbell (1996)	VIC, SA[C]	AHC 45424
Order: Tetraphyllidea Carus, 1863				
Type 1	Intestine, caeca	[A]	SA[C]	AHC 29777–78
Type 3	Caeca	[A]	SA[C]	AHC 29779
Tentacullariidae				
Nybelinia thyrsites Korotaeva, 1971	Stomach	Beveridge and Campbell (1996)	SA, SA[C]	AHC 29780 AHC 45425
Digenea				
Acanthocolpidae				
Monostephanostomum georgianum Kruse, 1979	Digestive tract	[A]	SA[C]	AHC 29773–74
Monostephanostomum manteri Kruse, 1979	Digestive tract	[A]	SA[C]	AHC 29912–14
Bucephalidae				
Telorhynchus arripidis Crowcroft, 1947	Digestive tract	[A]	SA[C]	AHC 29775 AHC 29791
Hemiuridae				
Erilepturus tiegsi Woolcock, 1935	Digestive tract	[A]	SA[C]	AHC 29770–72 AHC 29790
Elytrophalloides humerus Bray, 1990	Stomach	[A]	SA[C]	AHC 29792
Elytrophallus sp. Manter, 1940	Gills—artefact	[A]	SA[C]	AHC 29793

Lepocreadiidae				
Opechona kahawai Bray and Cribb, 2003	Intestine, caeca	Bray and Cribb (2003a)	TAS, SA[C]	AHC 29776
Monogenea				
Microcotylidae				
Kahawaia truttae Lebedev, 1969	Gills	[A]	SA[C]	AHC 29767–69, 29882, 29885–86 BMNH 2009.12.28.3–4 USNPC 102676.00–102677.00
Nematoda				
Anisakidae				
Contracaecum sp.	Stomach, caeca	[A]	SA[C]	AHC 45428–29
Hysterothylacium sp.	Digestive tract	[A]	SA[C]	AHC 45426–27
Philometridae				
Philometra sp.	Stomach	[A]	SA[C]	AHC 45430
Branchiura				
Argulidae				
Argulus diversicolor Byrnes, 1985	Skin	Catalano and Hutson (2010)	SA[C]	C6823
Copepoda				
Caligidae				
Caligus bonito Wilson, 1905	Gills	Catalano and Hutson (2010)	SA[C]	C6819
Caligus longipedis Bassett-Smith, 1898	Gills	Catalano and Hutson (2010)	SA[C]	C6815–16
Caligus punctatus Shiino, 1955	Skin	Catalano and Hutson (2010)	SA[B, C]	C6817–18

Table 5. Metazoan parasite fauna from *Arripis trutta* collected in this study and presented in previous literature**.** [A] = new host record; [B] = new Australian record; [C] = ambiguous parasite-host record; [D] = from current study. Abbreviations: GAB, Great Australian Bight; NA, not applicable; NSW, New South Wales; NZ, New Zealand; PR, previously recorded from the literature but not found in current study; SA, South Australia; TAS, Tasmania; TS, Tasman Sea; VIC, Victoria.

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Ceratomyxidae				
Ceratomyxa arripica	Gall bladder	Su and White (1994)	TAS	PR
Leptotheca annulata Meglitsch, 1960	Gall bladder	Meglitsch (1960)	NZ	PR
Leptotheca minima Meglitsch, 1960	Gall bladder	Meglitsch (1960)	NZ	PR
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	NR	Beveridge and Campbell (1996)	VIC	PR
Order: Tetraphyllidea Carus, 1863				
Type 3	Intestine	[A]	NSW[D]	AHC 29803
Tentaculariidae				
Nybelinia sp. larva Baker, 1971	Stomach	Baker (1971)	NZ	PR
Digenea				
Acanthocolpidae				
Monostephanostomum georgianum Kruse, 1979	Digestive tract	[A]	SA[D]	AHC 29796–97
Monostephanostomum manteri Kruse, 1979	Intestine, caeca	Kruse (1979)[C]; Bray and Cribb (2002)	SA[C], TAS, SA[D]	AHC 29798
Bucephalidae				
Prosorhynchus sp. Manter, 1954	NR	Manter (1954)	NZ	PR
Telorhynchoides longicollis Lebedev, 1968	NR	Lebedev (1968) [C]	GAB[C]	PR

Telorhynchus arripidis Crowcroft, 1947	Digestive tract	Crowcroft (1948); Manter (1954); Lebedev (1968)[C]	TAS, NZ, GAB[C], TS, NSW[D]	AHC 29800–01
Telorhynchus kahawai Lebedev, 1968	NR	Lebedev (1968)[C]	GAB[C]	PR
Telorhynchus peacheyi Lebedev, 1968	NR	Lebedev (1968)[C]	GAB[C], TS	PR
Hemiuridae				
Erilepturus tiegsi Woolcock, 1935	Stomach, caeca	Yamaguti (1958)	VIC, SA[D]	AHC 29794–95
Hemiurus (Anahemiurus) sp. Manter, 1947	Digestive tract	Baker (1971)	NZ	PR
Parahemiurus arripidis Lebedev, 1971	NR	Bray and Cribb (2002)	NZ	PR
Parahemiurus sp. Lebedev, 1971	NR	Lebedev (1971)	NZ	PR
Lepocreadiidae				
Opechona kahawai Bray & Cribb, 2003	Caeca	Bray and Cribb (2003a)	TAS, NSW[D]	AHC 29802
Opecoelidae				
Pseudopecoeloides arripi Aken'Ova *et al*., 2009	Stomach	[A]	NSW[D]	AHC 29799
Syncoliidae				
Syncoelium filiferum Sars, 1885	Gills	Rohde *et al.* (1980)	NZ	PR
Monogenea				
Microcotylidae				
Kahawaia truttae Lebedev, 1969	Gills	Dillon and Hargis (1965); Lebedev (1969)[C]	NZ, GAB[C], SA[D], NSW[D]	AHC 29781–82
Nematoda				
Anisakidae				
Anisakis sp. larva	Viscera	Hewitt and Hine (1972)	NZ	PR
Contracaecum aduncum Rudolphi, 1802	Digestive tract	Baker (1971)	NZ	PR

Contracaecum sp.	Stomach, intestine, body cavity	Hewitt and Hine (1972)	NZ	PR
Hysterothylacium sp.	Digestive tract	[A]	NSW[D]	AHC 45431
Annelida				
Pisciolidae				
Austrobdella translucens Badham, 1916	Tail fin	Bolton *et al.* (2005) [C]	SA[C]	PR
Copepoda				
Caligidae				
Caligus bonito Wilson, 1905	Gills	Catalano and Hutson (2010)	NSW[D]	C6821
Caligus kahawai Jones, 1988	Body surface	Jones (1988)	NZ	PR
Caligus pelamydis Hewitt, 1963	Gills	Jones (1988)	NZ, NSW[B, D]	C6822
Caligus punctatus Shiino, 1955	Gills	Catalano and Hutson (2010)	NSW[B, D]	C6820
Chondracanthidae				
Chondracanthus australis Ho, 1991	Gills	Ho (1991)	NZ	PR
Ergasilidae				
Abergasilus amplexus Hewitt, 1978	Gills	Hewitt (1978)	NZ	PR
Isopoda				
Cymothoidae				PR
Codonophilus imbricatus Fabricius, 1787	Tongue	Baker (1971)	NZ	
Nerocila orbignyi Guerin,	Body	Hewitt and Hine (1972)	NZ	PR

Carangidae

Seriola lalandi

Seriola hippos

Several *Seriola* spp. have been cultured either commercially or experimentally in the Mediterranean, Korea, Australia, United States and Japan. They are ideally suited to aquaculture as they display high growth performance, are hardy, and have a highly regarded flesh, which in Japan is generally consumed as sushi or sashimi (Rowland 2009). Nonetheless, disease management is an important component of *Seriola* spp. aquaculture as cultured *Seriola* spp. are afflicted by a range of viral, bacterial, fungal and parasitic disease agents, the majority of which have caused significant mortalities and require substantial monetary input to control (Sharp *et al.* 2003).

Nine species of *Seriola* occur worldwide, with four species, namely *S. lalandi, S. hippos, S. dumerili* and *S. rivoliana,* occurring in Australian waters. The former two are discussed below, whereas the latter two *Seriola* spp., along with *S. quinqueradiata* (economically important in Japan and Korea), are discussed briefly later in this section.

Yellowtail kingfish, *S. lalandi*, are distributed in waters of the Pacific and Indian Oceans off South Africa, Japan, southern Australia, New Zealand, Canada and the United States of America (Froese and Pauly 2005). In Australia, *S. lalandi* is a popular recreational species which inhabits southern coastal waters from Queensland to Western Australia and northern Tasmania (Hutchins and Swainston 1986). The primary wild-caught commercial fishery is in New South Wales (NSW) and produces about 200 t per year (Anon. 2005). This species is also farmed in sea-cages in Spencer Gulf, South Australia and currently produces about 1,200 t per annum with a value of around $13 million. Production is expected to increase to 10,000 t by 2012 (Rowland 2009).

Samson fish, *S. hippos,* are distributed in the temperate coastal waters of southern Australia and New Zealand. They are not considered a major commercial or recreational target species due to their poor reputation as a food fish. However, *S. hippos* are recognised for their game fish attributes and targeted by many sportfishers (Rowland 2009).

The natural occurrence of *S. lalandi* and *S. hippos* near locations where *S. lalandi* are farmed in South Australia provides an opportunity for transfer of parasites from wild to farmed populations. Presently, only two parasite species require management in the South Australian *S. lalandi* industry, the monogeneans *Benedenia seriolae* and *Zeuxapta seriolae*. A parasite risk assessment has been provided previously for *S. lalandi* sea-cage aquaculture in South Australia (Hutson *et al.* 2007b).

Up to 40 other metazoan parasite species can infect wild *S. lalandi* in southern and eastern Australian waters (Table 6) and *S. hippos* share at least seven of these species (Table 7). A new aporocotylid blood fluke was found to infect *S. lalandi* and *S. hippos* and is described in Chapter 5.

Table 6. Metazoan parasite fauna of wild and farmed *Seriola lalandi* (yellowtail kingfish) collected in this study and presented in previous records from Australasia. Abbreviations: EC, East Coast; NZ, New Zealand; PR, previously recorded from the literature but not found in current study; SA, South Australia; SC, South Coast; SG, Spencer Gulf; VIC, Victoria; *Farmed fish (-, not our study).

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Ceratomyxidae				
Ceratomyxa buri Yokoyama, 2001	Gall-bladder	Hutson *et al.* (2007b)*	Vic, SG	AHC 34260
Ceratomyxa seriolae Yokoyama, 2001	Gall-bladder	Hutson *et al.* (2007b)*	Vic, SG	AHC 34259
Kudoidae				
Kudoa sp.	Muscle	Hutson *et al.* (2007b)	EC	PR
Sphaerosporidae				
Unicapsula seriolae Lester, 1982	Muscle	Hutson *et al.* (2007b)	EC	PR
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	Body cavity	Hutson *et al.* (2007b)*	SG	AHC 29179
Order: Tetraphyllidea Carus, 1863				
Type 1	Stomach	Hutson *et al.* (2007b)	EC, SG	AHC 29161
Type 4	Digestive tract	Hutson *et al.* (2007b)*	EC, Vic, SG	AHC 29162–65
Tentaculariidae				
Nybelinia thyrsites Korotaeva, 1971	Intestine	Hutson *et al.* (2007a)	EC	AHC 29134
Digenea				
Acanthocolpidae				
Stephanostomum petimba Yamaguti, 1970	Digestive tract	Hutson *et al.* (2007b)	EC, SC, SG	AHC 29146
Tormopsolus orientalis Yamaguti, 1934	Stomach, intestine	Hutson *et al.* (2007b)*	EC, Vic, SG	AHC 29143–44
Bucephalidae				

Bucephalus gorgon (Linton, 1905) Eckmann, 1932	Digestive tract	Hutson *et al.* (2007b)*	EC, SC, SG	AHC 29148
Rhipidocotyle longicirrus Diesing, 1858	Digestive tract	Hutson *et al.* (2007b)*	EC, SG	AHC 29150–51
Telorhynchus sp.	Digestive tract	Hutson *et al.* (2007a)	Vic	AHC 29121– 22
Didymozoidae				
Undetermined species	Viscera	Hutson *et al.* (2007a)	EC, Vic	AHC 29123 & 34174
Hemiuridae				
Dinurus longisinus Looss, 1907	Stomach	Bray *et al.* (1993b)	EC	PR
Ectenurus trachuri Yamaguti, 1934	Stomach	Bray *et al.* (1993b)	EC	PR
Elytrophalloides humerus Bray, 1990	Stomach	Hutson *et al.* (2007b)*	SG	AHC 29155
Elytrophalloides oatesi (Leiper & Atkinson, 1914) Szidat & Graefe, 1967	Stomach	Hutson *et al.* (2007a)	Vic	AHC 29125
Elytrophallus sp.	Stomach	Hutson *et al.* (2007b)*	EC, Vic, SG	AHC 29153
Hirudinella sp.	Stomach	Hutson *et al.* (2007a)	Vic	AHC34176
Lecithasteridae				
Aponurus laguncula Looss, 1907	Stomach	Hutson *et al.* (2007a)	Vic	AHC 29130–31
Lecithaster stellatus Looss, 1907	Stomach	Hutson *et al.* (2007b)	EC	Bray *et al.* (1993a)
Lecithocladium sp.	Stomach	Hutson *et al.* (2007a)	EC	AHC 29127
Parahemiurus merus Linton, 1910	Stomach	Hutson *et al.* (2007b)*	Vic, SG	AHC 29156–59
Lepocreadidae				
Opechona kahawai Bray & Cribb, 2003	Stomach	Hutson *et al.* (2007a)	Vic	AHC 29132–33

Pleurus digitatus Looss, 1899	Stomach	Hutson *et al.* (2007a)	EC	AHC29129
Aporocotylidae				
Paradeontacylix godfreyi Hutson & Whittington, 2006	Heart	Hutson *et al.* (2007b)	SG, Vic	AHC 28904–08
	Gills	Bullard and Overstreet (2004)	Miami, Florida	PR
Paradeontacylix sanguinicoloides McIntosh, 1934	Heart	Hutson *et al.* (2007b)	EC	AHC28909
	Heart	Hutson *et al.* (2007b)	Vic	AHC 28911
Paradeontacylix n. sp.				
Monogenea				
Capsalidae				
Benedenia seriolae Yamaguti, 1934	Skin	Sharp *et al.* (2003)	NZ	AHC 29180
	Skin	Hutson *et al.* (2007b)*	EC, Vic, SG	
Heteraxinidae				
Paramicrocotyloides reticularis Rohde, 1978	Gills	Hutson *et al.* (2007b)	EC	AHC 29105–06
Zeuxapta seriolae (Meserve, 1938)	Gills	Sharp *et al.* (2003)	NZ	AHC 29182
Price, 1962	Gills	Hutson *et al.* (2007b)*	EC, Vic, SG	
Nematoda				
Anisakidae				
Anisakis sp.	Stomach, caeca	Hutson *et al.* (2007b)	SG	AHC 34261
Contracaecum sp.	Caeca	Hutson *et al.* (2007a)	EC	AHC 34184
Hysterothylacium sp.	Stomach, caeca	Hutson *et al.* (2007b)	EC, SG	AHC 34264 & 34266
Pseudoterranova sp.	Intestine	Hutson *et al.* (2007a)	EC	AHC 34178
Spiruridae				
Rhabdochona sp.	Stomach	Hutson *et al.* (2007a)	Vic	AHC 34191
Acanthocephala				
Rhadinorhynchidae				

Australorhynchus tetramorphacanthus Lebedev, 1967	Intestine	Lebedev (1967)	EC, SA	PR
Rhadinorhynchus sp. 1	Intestine	Hutson *et al.* (2007a)	EC	AHC 29141–42
Rhadinorhynchus sp. 2	Intestine	Hutson *et al.* (2007a)	Vic	AHC 34177
Copepoda				
Bomolochidae				
Naricolax chrysophryenus Roubal, Armitage & Rohde, 1983	Nasal cavity	Hutson *et al.* (2007b)*	EC, SG	C6284–98
Caligidae				
Caligus aesopus Wilson, 1920	Gills	Sharp *et al.* (2003)	NZ	PR
Caligus amblygenitalis Pillai, 1961	Cavities	Hutson *et al.* (2007a)	EC	C6311
Caligus chiastos Lin & Ho, 2003	Gills	Hayward *et al.* (2009a)*	SA	PR
Caligus epidemicus Hewitt, 1971	Body surface	Hutson *et al.* (2007b)	EC, SG	C6313
Caligus lalandei Barnard, 1948	Skin Body surface	Sharp *et al.* (2003) Hutson *et al.* (2007a)	NZ EC, Vic	PR C6228–29
Caligus spinosus Yamaguti, 1939	Gill arch	Hutson *et al.* (2007a)	EC, Vic	C6230–31
Caligus sp. 1	Nd	Hutson *et al.* (2007b)*	EC, Vic, SA, SG	C6232 & C6312
Dissonidae				
Dissonus hoi Tang & Kalman, 2005	Nasal cavity	Hutson *et al.* (2007b)	EC, SG	C6317
Lernanthropidae				
Lernanthropus paenulatus Wilson, 1922	Gills	Hutson *et al.* (2007b)	EC, Vic, SG	C6239 & C6309
Lernanthropus sp.	Gills	Sharp *et al.* (2003)	NZ	PR
Lernaeopodidae				
Neobrachiella sp.	Gills	Sharp *et al.* (2003)	NZ	PR
Parabrachiella seriolae Yamaguti	Buccal folds	Hutson *et al.* (2007b)	EC, Vic, SG	C6321

& Yamasu, 1960				
Parabrachiella sp.	Gills	Hutson *et al.* (2007a)	EC	C6238
Pennellidae				
Peniculus sp.	Body surface	Hutson *et al.* (2007a)	EC, Vic	C6233–34

Table 7. Metazoan parasite fauna of *Seriola hippos* (Samson fish) collected in this study and presented in previous records from Australasia. Abbreviations: EC, East Coast; PR, previously recorded from the literature but not found in current study; SA, South Australia; SC, South Coast; SG, Spencer Gulf; WC, West Coast; (-, not our study).

Parasite	Microhabitat	Reference	Location	Accession number
Digenea				
Acanthocolpidae				
Stephanostomum petimba Yamaguti, 1970	Caeca	Hutson *et al.* (2007b)	EC, SC, SG	AHC 29147
Tormopsolus attenuatus Bray & Cribb, 2001	Caeca	Hutson *et al.* (2007b)	SA	AHC 29145
Bucephalidae				
Bucephalus gorgon (Linton, 1905) Eckmann, 1932	Caeca	Hutson *et al.* (2007b)	SA	AHC 29149
Hemiuridae				
Erilepturus hamati (Yamaguti, 1934) Manter, 1947	Stomach	Hutson *et al.* (2007b)	SA	AHC 29152
Elytrophallus sp.	Stomach	Hutson *et al.* (2007b)	SA	AHC 29154
Lecithasteridae				
Parahemiurus merus Linton, 1910	Stomach	Hutson *et al.* (2007b)	SA	AHC 29160
Aporocotylidae				
Paradeontacylix sanguinicoloides McIntosh, 1934	Heart	Hutson *et al.* (2007b)	SA	AHC 28910
Paradeontacylix sp.	Heart	Chapter 5	SA	AHC 28912
Monogenea				
Capsalidae				
Benedenia seriolae Yamaguti, 1934	Skin	Hutson *et al.* (2007b)	SA	AHC 29181
Heteraxinidae				

Zeuxapta seriolae (Meserve, 1938) Price, 1962	Gills	Hutson *et al.* (2007b)	EC	-
Nematoda				
Anisakidae				
Anisakis sp.	Stomach	Hutson *et al.* (2007b)	SA	AHC 34262
Contracaecum sp.	Stomach	Hutson *et al.* (2007b)	SA	AHC 34263
Hysterothylacium sp.	Stomach	Hutson *et al.* (2007b)	SA	AHC 34265
Copepoda				
Caligidae				
Caligus lalandei Barnard, 1948	Body surface	Hutson *et al.* (2007b)	SA	C6314
Caligus sp. 1	Nd	Hutson *et al.* (2007b)	SA	C6315
Caligus sp. 2	Gill arches	Hutson *et al.* (2007b)	SA	C6316
Lepeophtheirus sp.	Nd	Hutson *et al.* (2007b)	SA	C6318
Parapetalus spinosus Byrnes, 1986	Gills	Hutson *et al.* (2007b)	SA, WC	C6320
Dissonidae				
Dissonus hoi Tang & Kalman, 2005	Nasal cavity	Hutson *et al.* (2007b)	WC	-
Lernanthropidae				
Lernanthropus paenulatus Wilson, 1922	Gills	Hutson *et al.* (2007b)	SA	C6319
Laernaeopodidae				
Parabrachiella seriolae Yamaguti & Yamasu, 1960	Buccal, fin sulcus	Hutson *et al.* (2007b)	SA	C6322

Coryphaenidae

Coryphaena hippurus

Coryphaena hippurus is a large pelagic species that has a worldwide distribution in tropical and subtropical waters (Adams 2009). It is commonly referred to as dolphinfish or mahi mahi. In Australian waters, dolphinfish are found in Western Australia, Northern Territory and from Queensland to Montague Island in New South Wales (NSWDPI 2007). This species is limited in habitat by sea surface temperatures of 19–20ºC and extensions of distribution occur with seasonal variations in water temperature (NSWDPI 2007).

Dolphinfish are captured by commercial and recreational fishers in waters around Australia and New Zealand (Kingsford and Defries 1999). Dolphinfish have several features which make them a suitable candidate for farming, including high fecundity and continuous spawning, exceptional growth rate, efficient food conversion and a high market price (Langdon 1991a; Kingsford and Defries 1999). This species has been trialled for culture in Western Australia (Kingsford and Defries 1999). However, before this candidate aquaculture species is farmed successfully, it will be crucial to learn to diagnose and control parasitic infections that can lead to disease and mortality, given the wide suite of parasite species that infect this host species.

Numerous studies have investigated the parasite assemblage of this top-level predator (e.g. Langdon 1991a; Dyer *et al.* 1997; Carbonell *et al.* 1999; Williams and Bunkley-Williams 2010), and subsequently, a diversity of parasite species are known from this host species (Table 8). We examined 35 *C. hippurus* and identified seven parasite species (Table 8). Of note was a new host record for *Capsala laevis* (Verrill, 1875) which is well known to infect marlin (*Tetrapterus audax*). Parasite specimens found were juveniles and identification was not possible based on morphology but identification was achieved using molecular techniques and sequence data were compared with data generated for adult *C. laevis* from *T. audax* off Port Stephens, NSW (Perkins *et al.* 2009).

Table 8. Metazoan parasite fauna of wild and farmed dolphin fish *Coryphaena hippurus* collected in this study and presented in previous records from Australasia. Abbreviations: Aus, Australia; CI, Canary Islands; EA, Eastern Australia; EP, Eastern Pacific; GM, Gulf of Mexico; IO, Indian Ocean; KC, Kerala Coast; Med, Mediterranean; N/g, Not given; NSW, New South Wales; PNG, Papua New Guinea; PO, Pacific Ocean, PR, Puerto Rico; SA, South Australia; USA, United States of America; WA, Western Australia; *Farmed fish (-, not our study); ? = questionable record.

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Kudoidae				
Kudoa thyrsites Gilchrist, 1924	Muscle	Langdon (1991b)*	WA	
	Muscle	Williams and Bunkley-Williams (2010)	EA	
Kudoa sp.	N/g	Williams and Bunkley-Williams (2010)	N/g	
Myxidiidae				
Myxidium sp.	Kidney	Langdon (1991a)	Exmouth & Esperance regions	
Cestoda				
Bothriocephalidae				
Bothriocephalus janickii Markowski, 1971	N/g	Dyer *et al.* (1997)	Southern Sea, Bay of Bengal	
Echinophallidae				
Echinophallus lonchinobothrium Monticelli, 1890	N/g	Williams and Bunkley-Williams (2010)	Med	
Eucestoda incertae sedis				
Plerocercoides lonchophorus Guiart, 1935	Uncertain record	Unc record (W&BW 1996)	Uncertain record	
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	Walls of stomach and intestine, viscera	Williams and Bunkley-Williams (1996)	PR, USA	
Tetrarhynchus sp.	Uncertain record	Unc record (W&BW 1996)	Uncertain record	

Lacystorhynchidae				
Floriceps saccatus Cuvier, 1817	Abdominal cavity	Carbonell *et al.* (1998)	Western Med, CI, Eastern Atlantic	
Order Tetraphyllidea				
Scolex pleuronectis Müller, 1787	Intestine	Williams and Bunkley-Williams (1996)	PR	
Order Trypanorhyncha				
Trypanorhyncha sp.	N/g	Williams and Bunkley-Williams (2010)	EP	
Otobothriidae				
Otobothrium cysticum Mayer, 1842	Stomach wall, viscera	Williams & Bunkley-Williams (1996) [as O. crenacolle]	Bermuda, USA	
Otobothrium dipsacum Linton, 1897	Body cavity, intestine	Williams & Bunkley-Williams (1996)	PR	
Phyllobothriidae				
Pelichnibothrium speciosum Monticelli, 1889	Stomach, outer layer of intestine and viscera	Williams & Bunkley-Williams (1996)	PR	
Rhinebothrium flexile Linton, 1890	Intestine, liver, viscera	Williams & Bunkley-Williams (1996)	PR	
Pterobothriidae				
Pterobothrium acanthotruncatum Escalante & Carvajal, 1984	Viscera	Williams & Bunkley-Williams (1996)	Peru	
Pterobothrium heteracanthum Diesing, 1850	N/g	Williams (2010)	N/g	
Sphyriocephalidae				
Dibothriorhynchus claviger	Uncertain records	Uncertain records	Eastern Atlantic	
Hepatoxylon megacephalum	viscera	present study	SA	AHC 45794

Rudolphi 1819			
Hepatoxylon stenocephala Bosc, 1811	N/g	N/g	Eastern Atlantic
Hepatoxylon trichiura Holten, 1802) Dollfus, 1942	Stomach, viscera, body cavity, intestinal tract	Williams and Bunkley-Williams (1996)	PR, USA, Brazil
Tentaculariidae			
Nybelinia bisulcata (Linton, 1889) Dollfus, 1929	Stomach wall, body cavity	Williams and Bunkley-Williams (1996)	PR, USA
Nybelinia lamonteae Nigrelli 1938	Mesenteries	Williams and Bunkley-Williams (1996)	PR
Nybelinia scoliodoni Vijayalakshmi, Vijayalakshmi & Gangadharam, 1996	Stomach wall	Palm and Overstreet (2000)	Coastal waters off Ocean Springs, Mississippi, GM
Nybelinia sp.	N/g	Williams and Bunkley-Williams (2010)	EP, IO
Tentacularia coryphaenae Bosc, 1797	Body cavity, stomach, intestine Viscera	Williams and Bunkley-Williams (1996) Dyer (1997)	USA, La Parguera PR
Trypanorhyncha incertae sedis			
Tetrarhynchus discophorus Rudolphi, 1819	N/g	Williams and Bunkley-Williams (2010)	Med
Tetrarhynchus macrobothrius Rudolphi, 1819	N/g	Williams and Bunkley-Williams (2010)	Med
Tetrarhynchus papillosus Rudolphi, 1809	Uncertain record	Unc record (W&BW 96/2010)	Uncertain record

Tetrarhynchus sp.	Uncertain record	Unc record (W&BW 96/2010)	Uncertain record
Tetrarhynchid	N/g	Williams and Bunkley-Williams (2010)	PR
Families to be identified			
Plicocestus janickii	Uncertain record	Unc record (W&BW 96/2010)	Uncertain record
Plerocercoids of a trypanorhynch tapeworm	Stomach, intestine	Langdon (1991a)	Florida, USA
Digenea			
Acanthocolpidae			
Stephanostomum coryphaenae Manter, 1947	Stomach, intestine, rectum	Williams and Bunkley-Williams (1996)	Brazil, USA, Bimini, Curaçao
	Stomach, intestine	Dyer *et al.* (1997)	PR
Accacoeliidae			
Accacladium serpentulum Odhner, 1928	N/g	Williams and Bunkley-Williams (2010)	EP
Tetrochetus coryphaenae Yamaguti, 1934	Intestine	Williams and Bunkley-Williams (1996)	La Parguera, USA, Curaçao, Netherlands, Bahamas, Jamacia, Mexico, Panama, Japan
	Stomach, intestine	Dyer *et al.* (1997)	PR
Bathycotylidae			
Bathycotyle branchialis Darr, 1902	Gills	Burnett-Herkes (1974)	Straits of Florida, USA
	Gills	Carbonell *et al*. (1999)	Med
Bathycotyle coryphaenae Yamaguti 1938	Gill filaments	Williams and Bunkley-Williams (1996)	USA, Japan, PR
Didymozoidea			
Monilicaecum sp.	Stomach contents	Williams and Bunkley-Williams (2010)	N/g
Torticaecum sp.	N/g	Williams and Bunkley-	PR

		Williams (2010)	
Hemiuridae			
Dinurus barbatus Cohn, 1903	Stomach, intestine	Dyer *et al.* (1997)	PR
	Stomach	Carbonell *et al.* (1999)	Med, CI
	Stomach	Rekha and John (2004)	KC, India
Dinurus breviductus Looss, 1907	Stomach, intestine	Dyer *et al.* (1997)	PR
	Stomach	Carbonell *et al.* (1999)	Med, CI
Dinurus ivanosi Rekha & John 2004	Stomach	Rekha & John (2004)	KC, India
Dinurus longisinus Looss, 1907	Stomach	Bray (1990)	Cape Province, WA
	Stomach	Bray *et al.* (1993a)	PNG
	Stomach	Carbonell *et al.* (1999)	Med, CI
	Stomach	Rekha & John (2004)	KC, India
Dinurus scombri Yamaguti, 1934	Stomach	Rekha & John (unpl)	IO
Dinurus tornatus Rudolphi, 1819	Stomach, intestine	Rapaptopoulou and Lambertsen (1987)	Florida, USA
	Stomach	Carbonell *et al.* (1999)	Med, CI
	Stomach	Rekha & John (2004)	KC, India
	Stomach	Williams and Bunkley-Williams (1996)	PR, USA, Bahamas, Brazil, Pacific coast of Panama
Dinurus sp.	N/g	Dyer *et al.* (1992)	La Parguera
Lecithochirium branchialis Stunkard & Nigrelli, 1934	N/g	Williams and Bunkley-Williams (2010)	Med
Lecithocladium excisum (Rudolphi, 1819) Liihe, 1901	Stomach	Carbonell *et al.* (1999)	Med
Hirudinellidae			
Hirudinella sp.	Stomach	Carbonell *et al.* (1999)	Med

Hirudinella ventricosa (Pallas, 1774) Baird, 1853	Stomach	Williams and Bunkley-Williams (1996)	USA	
	Stomach	Dyer *et al.* (1997)	PR	
Lepocreadiidae				
Opechona bacillaris (Molin, 1859) Looss, 1907	N/g	Bray and Gibson (1990)	North-east Atlantic	
Opecoelidae				
Helicometrina nimia Linton, 1910	N/g	Williams and Bunkley-Williams (2010)	EP	
Opecoeloides furcatus Bremser in Rudolphi, 1819	N/g	Williams and Bunkley-Williams (2010)	Med	
Monogenea				
Capsalidae				
Benedenia hendorffii (von Linstow, 1889) Odhner, 1905	Skin	Whittington *et al.* (2001b)	Chile	
Benedenia seriolae (Yamaguti, 1934) Meserve, 1938	N/g	Williams and Bunkley-Williams (2010)*	Aus	
Benedenia sp.	Cornea, skin, gills	Langdon (1991) Whittington *et al.* (2001b)*	WA	
Capsala laevis (Verrill, 1875) Johnston, 1929	N/g	Williams and Bunkley-Williams (2010)	N/g	
	Skin	present study	NSW	Not accessioned
Neobenedenia melleni (MacCallum, 1927) Yamaguti, 1963	N/g	Colorni (1994)*	Red Sea	
Neothorcocotylidae				
Neothroacocotyle acanthocybii (Meserve, 1938) Hargis, 1956	N/g	Williams and Bunkley-Williams (2010)	Pacific	

Udonellidae			
Udonella caligorum Johnston, 1835	N/g	Williams and Bunkley-Williams (2010)	PR
Nematoda			
Anisakidae			
Anisakis physeteris Baylis, 1923	N/g	Williams and Bunkley-Williams (2010)	EP
Anisakis typica Diesing, 1860	N/g	Mattiucci *et al.* (2002)	IO
	N/g	Palm *et al.* (2008)	Indonesia
Anisakis type I	N/g	Mattiucci *et al.* (2002)	IO
Anisakis type II	N/g	Williams and Bunkley-Williams (2010)	PO
Anisakis sp.	N/g	Dyer *et al.* (1997)	Nagasaki
Contracaecum sp.	N/g	Williams and Bunkley-Williams (2010)	Brazil
Hysterothylacium marinum Linnaeus, 1767	N/g	Dyer *et al.* (1997)	Aus
Hysterothylacium pelagicum Deardorff & Overstreet, 1982	Stomach, pyloric caeca and intestine	Williams and Bunkley-Williams (1996)	PR, USA, Hawaii
	Intestine	Dyer *et al.* (1997)	PR
Hysterothylacium sinense Li, 2007	Intestine	Li *et al.* (2007)	Yellow Sea, China
Hysterothylacium type HA	N/g	Deardorff *et al.* (1982)	Hawaii, Pacific
Hysterothylacium type HB	N/g	Deardorff *et al.* (1982)	Hawaii, Pacific
Hysterothylacium type HC	N/g	Deardorff *et al.* (1982)	Hawaii, Pacific

Hysterothylacium sp.	N/g	Williams and Bunkley-Williams (2010)	Peru
Terranova type HA	Viscera	Deardorff *et al.* (1982)	Hawaii, Pacific
Ascarididae			
Ascaris sp.	N/g	Williams and Bunkley-Williams (1996)	Pacific
Porrocaecum sp.	N/g	Williams and Bunkley-Williams (2010)	Panama
Cystidicolidae			
Cystidicola lepturum Rudolphi, 1819	?	Williams and Bunkley-Williams (2010)	Med
Metabronema magna Taylor, 1925	Pyloric caeca, pancreatic tissue	Carbonell *et al.* (1999)	Med
Dracunculidae			
Philometroides sp.	Abdominal cavity	Carbonell *et al.* (1999)	Med
Philometridae			
Philometra sp.	?	Williams and Bunkley-Williams (2010)	Florida, USA
Trichosomoididae			
Huffmanela schouteni Campbell, 1991	Stomach contents	Moravec and Garibaldi (2003)	Western Ligurian Sea
Acanthocephala			
Gen. et. sp. indet.	KSH	present study	SA
Polymorphidae			
Corynosoma obtuscens Lincicome, 1943	N/g	Williams and Bunkley-Williams (2010)	EP
Corynosoma sp.	N/g	Williams and Bunkley-Williams (2010)	EP

Rhadinorhynchidae				
Nipporhynchus katsuwonis (Harada, 1928) Chandler 1934	N/g	Dyer *et al.* 1997	Izu, Kanagawa Prefecture	
Rhadinorhynchus dujardini Golvan, 1969	N/g	Williams and Bunkley-Williams (2010)	Pacific	
Rhadinorhynchus pristis Rudolphi, 1802	Stomach, intestine	Williams and Bunkley-Williams (1996)	La Parguera, Curaçao	
	Stomach, pyloric caecal wall, pancreatic tissue	Carbonell *et al.* (1999)	Med, Atlantic Ocean	
			N/g	
Serrasentis sagittifer (Linton, 1889) Van Cleave, 1923	Intestine and pyloric caeca; body cavity	Williams and Bunkley-Williams (1996)		
Copepoda				
Caligidae				
Caligus balistae Steenstrup and Liitken, 1861	Fins, body	Williams and Bunkley-Williams (1996)	PR, USA	
Caligus belones Krøyer, 1863	Gills	Burnett-Herkes (1974)	USA	
Caligus bonito Wilson, 1905	Roof of mouth, gill filaments	Williams and Bunkley-Williams (1996)	Atlantic	
	Gills mucus mass, inner surface of operculum	Carbonell *et al.* (1999)	Med	
	Gills, operculum	present study	NSW	SAM C6915–20
Caligus coryphaenae Steenstrup & Lutken, 1861	Gills, body surface	Williams and Bunkley-Williams (1996)	PR	
	Gills mucus mass, inner surface of operculum	Carbonell *et al.* (1999)	Med	
	Dorsal fin	present study	NSW	SAM C6908
Caligus curtus Müller, 1785	N/g ?	Williams and Bunkley-Williams (1996)	Brazil	

Caligus patulus Wilson, 1937	Gills	Burnett-Herkes (1974)	USA	
Caligus productus Müller, 1785	Gills, mouth	Williams and Bunkley-Williams (1996)	PR, USA	
	Gills mucus mass, inner surface of operculum	Carbonell *et al.* (1999)	Med	
	Gills	present study	NSW	SAM C6906–7
Caligus quadratus Shiino, 1957	Operculum, mouth, body, gills	Williams and Bunkley-Williams (1996)	USA	
	Gills mucus mass, inner surface of operculum	Carbonell *et al.* (1999)	Med	
	Gills, gill washing, operculum	present study	NSW	SAM C6909–14
Caligus wilsoni Müller, 1785	Body	Williams and Bunkley-Williams (1996)	USA	
Dysgamus sp.	N/g	Williams and Bunkley-Williams (2010)	PR	
Euryphoridae				
Euryphorus brachypterus Gerstaecker, 1853	N/g	Williams and Bunkley-Williams (2010)	Pacific	
Euryphorus nordmanni Milne-Edwards, 1840	Inner surface of operculum, gill chamber, mouth	Williams and Bunkley-Williams (1996)	PR	
Euryphorus nymphae Steenstrup & Lutken, 1861	Gills mucus mass, inner surface of operculum	Carbonell *et al.* (1999)	Atlantic	
Lernaeopodidae				
Brachiella thynni Cuvier, 1830	Pectoral fin	Williams and Bunkley-Williams (1996)	PR	

Charopinopsis quaternia Wilson, 1935	Gill filaments	Williams and Bunkley-Williams (1996)	PR, USA	
	Gills	present study	NSW	SAM C6921–22
Lernaeenicus hemiramphi Kirtisinghe 2009	N/g	Williams and Bunkley-Williams (2010)	Indo Pacific	
Lernaeenicus longiventris Wilson, 1917	Head embedded in hosts body	Williams and Bunkley-Williams (1996)	USA, northwest Atlantic, west coast of Africa	
Lernaeenicus sp.	N/g	Williams and Bunkley-Williams (2010)	Pacific	
Neobrachiella coryphaenidae Pearse, 1952	Gill filaments	Carbonell *et al.* (1999)	Med	
Neobrachiella sp.	N/g	Williams and Bunkley-Williams (2010)	Pacific	
Pennellidae				
Peniculus fistula von Nordmann, 1832	N/g	Williams and Bunkley-Williams (2010)	Med	
Pennella filosa Linnaeus, 1758	Dorsal & anal fins, dorsolateral muscular tissue, abdominal cavity	Carbonell *et al.* (1999)	Med	
Pennella varians Steenstrup & Lutken, 1861	N/g	Williams and Bunkley-Williams (2010)	Med	
Pennella sp. 1	Skin, fins	Williams and Bunkley-Williams (1996)	PR	
Pennella sp. 2	N/g	Williams and Bunkley-Williams (2010)	Pacific	

Sarcotretes nordicornis Steenstrup & Lutken, 1861	N/g	Williams and Bunkley-Williams (2010)	N/g
Pseudocycnidae			
Pseudocycnus appendiculatus Heller, 1865	Gill filaments	Williams and Bunkley-Williams (1996)	PR, Atlantic
Amphipoda			
Family to be identified			
Dithyrus faba Murphy, 1914	Stomach contents	Williams and Bunkley-Williams (2010)	N/g
Isopoda			
Cymothoidae			
Anilocra physodes Linnaeus, 1758	N/g	Williams and Bunkley-Williams (2010)	Med
Cymothoa eremita Brünnich, 1783	N/g ?	Williams and Bunkley-Williams (2010)	IO
Glossobius auritus Bovallius, 1885	Stomach contents	Williams and Bunkley-Williams (2010)	Western Atlantic
Glossobius impressa Say, 1818	Stomach contents	Williams and Bunkley-Williams (2010)	Western Atlantic
Livoneca ovalis Say, 1818	Stomach contents	Williams and Bunkley-Williams (1996)	USA
Nerocila excisa Richardson, 1914	Stomach contents	Williams and Bunkley-Williams (2010)	Western Atlantic
Noriletica indica Milne-Edwards, 1840	Stomach contents	Yamauchi *et al.* (2005)	Philippines
Idoteidae			

Idotea metallica Murphy, 1914	Stomach contents	Williams and Bunkley-Williams (2010)	N/g
Other			
Gnathid isopod	Skin	Langdon (1991a)*	Albany

Hemiramphidae

Hyporhamphus melanochir

The southern garfish *Hyporhamphus melanochir*, a schooling fish that generally concentrate in sheltered bays, shallow inshore waters and estuaries, is endemic to temperate southern Australian waters (Gomon *et al.* 2008). The species is commercially harvested throughout its distribution, with the national commercial catch dominated by those taken in the coastal waters of SA (Steer *et al.* 2010). Catches in SA typically exceed 300 t per annum and is worth an estimated AUD$3 million per year (Knight *et al.* 2007). Recent assessment of the South Australian garfish fishery suggests that it is currently overexploited, with considerable age truncation and declining catches (Fowler *et al.* 2008; Steer *et al.* 2009).

Considering the economic importance of this species, it is surprising that the parasite assemblage has not been previously documented. We record 14 parasite species from this host species in SA. Fish were examined from nine sites in three regions (Spencer Gulf, Gulf St Vincent and northern Kangaroo Island) in South Australia to investigate spatial variation in parasite abundance (Chapter 3).

Table 9. Metazoan parasite fauna of southern Australian garfish *Hyporhamphus melanochir* examined in this study and presented in previous literature. Under infection parameters prevalence is expressed as a percentage (%) followed in parentheses by the number of fish infected and the total number of fish examined; mean intensity is follows in parentheses by maximum intensity. [A] = new host record; [B] = from current study. Abbreviations: NZ, New Zealand, SA, South Australia, WA, Western Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Order: Tetraphyllidea Carus, 1863				
Type 1	Intestinal tract	present study [A]	SA	AHC 29819
Type 2	Intestinal tract	present study [A]	SA	AHC 29820
Type 3	Intestinal tract	present study [A]	SA	AHC 29821
Digenea				
Acanthocolpidae				
Acanthocolpid metacercaria	Gills	present study [A]	SA	AHC 29815–16
Bivesiculidae				
Bivesiculid type	Intestinal tract	present study [A]	SA	AHC 29814
Conohelmins sp.	Intestinal tract	present study [A]	SA	AHC 29812–13
Opecoelidae				
Opecoelid type	Intestinal tract	present study [A]	SA	AHC 29817–18
Monogenea				
Axinidae				
Axine sp.	Gills	present study [A]	SA	AHC 29810–11

Nematoda				
Anisakidae				
Hysterothylacium sp.	Intestinal tract	present study [A]	SA	AHC 45439
Philometridae				
Philometra sp.	Body cavity	present study [A]	SA	AHC 45440
Acanthocephala				
Micracanthorhynchinidae				
Micracanthorhynchina hemirhamphi	Stomach	Baylis (1944)	NZ	PR
(Baylis 1944) Ward 1951	Intestinal tract	present study	SA	AHC 45438
Copepoda				
Bomolochidae				
Bomolochus bellones Burmeister, 1833	Operculum	Collette (1974)		PR
	Gills	present study	SA	C6843
Penneildae				
Lernaeenicus hemiramphi Kirtisinghe, 1932	Eye	Heegaard, (1962)		PR
Lernaeenicus sayori Yamaguti	Flesh	Collette (1974)	WA, SA	PR
	Flesh	present study	SA	C6844
Isopoda				
Cymothoidae				
Mothocya halei Bruce 1986	Branchial cavity	Bruce (1986)	SA	PR
	Branchial cavity, mouth	present study	SA	C6842

Latidae

Lates calcarifer

Barramundi, *Lates calcarifer*, is a large carnivorous fish found in tropical regions of Australia and throughout the Indo-West Pacific (Russell and Garrett 1985). In Australia, its natural distribution extends from the Ashburton River in Western Australia, throughout the Northern Territory, to the Maryborough River in Queensland. It is a highly opportunistic, fecund species that dominates many tropical rivers throughout its range because of a dynamic and flexible biology. The species supports substantial commercial and recreational fisheries in Australia and Asia and also a growing aquaculture industry (Keenan 1994).

Lates calcarifer was first bred in Australia in 1984 using intensive techniques developed in Asia. More recently, extensive larval rearing techniques in marine or brackish water ponds and greenwater culture have been used (Barlow *et al.* 1996). Problems have been encountered with disease, cannibalism and declining market prices. Highly intensive shorebased growout systems (freshwater or saltwater) combined with a year round supply of hatchery produced fish is currently practiced in South Australia and New South Wales. Total production was estimated at 450 t in 1995, valued at AUD$ 4.5 million (Battaglene and Fielder 1997). Other barramundi aquaculture strategies include cage culture in open sea and estuaries, marine raceways and intensive production in closed recirculation systems.

We examined 26 farmed *L. calcarifer* collected from a farm in Hinchinbrook, Queensland (Table 2). A risk assessment for the parasites that infect this species is provided in Chapter 7.

Table 10. Metazoan parasite fauna of wild and farmed barramundi *Lates calcarifer* collected in this study and presented in previous records from Australasia. Abbreviations: N/g = Not given; NT, Northern Territory; Qld, Queensland; *Farmed fish.

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Myxozoa gen et. sp. indet	Gall bladder	Rückert *et al.* (2008)*	Sumatra	PR
Cestoda				
Order Tetraphyllidea				
Scolex pleuronectis Müller, 1787	Intestine, stomach, pyloric caeca	Rückert *et al.* (2008)*	Sumatra	PR
Tentaculariidae				
Nybelinia indica Chandra 1986	Stomach, stomach wall	Rückert *et al.* (2008)*	Sumatra	PR
Digenea				
Bucephalidae				
Prosorhynchus sp.	Stomach	Rückert *et al.* (2008)*	Sumatra	PR
Hemiuridae				
Erilepturus hamati (Yamaguti, 1934) Manter, 1947	Stomach	Bray *et al.* (1993)	NT	PR
Sanguinicolidae				
Cruoricola lates Herbert *et al.*, 1994	Blood vessels	Herbert *et al.* (1994)	Qld, Malaysia	PR
Parasanguinicola vastispina Herbert & Shaharom 1995	Branchial arteries, dorsal aorta, mesenteric venules and renal artery	Herbert and Shaharom-Harrison (1995)*	Malaysia	PR
Cryptogonimidae				
Pseudometadena celebesensis^ Yamaguti, 1952	Intestine & pyloric caecae	Rückert *et al.* (2008)*	Sumatra	PR

Monogenea				
Capsalidae				
Benedenia epinepheli (Yamaguti, 1937) Meserve, 1938	Gills, body surface	Rückert *et al.* (2008)*	Sumatra	PR
Capsalid gen. et sp. Indet	Gills, body surface	Rückert *et al.* (2008)*	Sumatra	PR
Neobenedenia sp.1**	fins	present study*	Qld	AHC 35006–11
Neobenedenia melleni (MacCallum, 1927) Yamaguti, 1963**	Gills, body surface	Deveney *et al.* (2001)* Rückert *et al.* (2008)*	Qld Sumatra	PR PR
Diplectanidae				
Diplectanum sp.	Gills	Fletcher and Whittington (1998) present study	NT Qld	PR Not accessioned
Diplectanum latesi Tripathi 1957	Gills	Tingbao *et al.* (2006)	N/g	PR
Diplectanum narimeen Unnithan, 1964	Gills	Tingbao *et al.* (2006)	N/g	PR
Diplectanum penangi Laing & Leong, 1991	Gills	Tingbao *et al.* (2006)*	China*, Malaysia, Thailand	PR
Diplectanum setosus Nagibina, 1976	Gills	Tingbao *et al.* (2006)	N/g	PR
Laticola lingaoensis Tingbao *et al.*, 2006	Gills	Tingbao *et al.* (2006)*	China	PR
Laticola latesi Tripathi, 1957	Gills	Tingbao *et al.* (2006)*	China*, India, Malaysia, Thailand	PR
Laticola paralatesi Nagibina, 1976	Gills	Tingbao *et al.* (2006)*	China*, NT	PR
Pseudorhabdosynochus epinepheli Yamaguti 1938	Gills	Rückert *et al.* (2008)*	Sumatra	PR
Pseudorhabdosynochus lantauensis Beverley-Burton & Suriano, 1981	Gills	Rückert *et al.* (2008)*	Sumatra	PR

Pseudorhabdosynochus monosquamodiscusi Balasuriya & Leong, 1995	N/g	In: Tingbao *et al.* (2006)	N/g	PR
Nematoda				
Anisakidae				
Hysterothylacium sp.	Intestine, liver, mesenteries, pyloric caeca	Rückert *et al.* (2008)*	Sumatra	PR
Raphidascaris sp.	Stomach wall	Rückert *et al.* (2008)*	Sumatra	PR
Raphidascaris sp. 2	Intestine	Rückert *et al.* (2008)*	Sumatra	PR
Terranova sp.	Intestine, liver, mesenteries, pyloric caeca	Rückert *et al.* (2008)*	Sumatra	PR
Acanthocephala				
Rhadinorhynchidae				
Serrasentis sagittifer (Linton, 1889)Van Cleave, 1923	Mesenteries	Rückert *et al.* (2008)*	Sumatra	PR
Copepoda				
Caligidae				
Caligus epidemicus Hewitt, 1971	N/g	Johnson *et al.* (2004b)*	Thailand	PR
Lernanthropidae				
Lernanthropus latis Yamaguti, 1954	Gill filaments	Ho and Kim (2004a)	Gulf of Thailand, Celebes, India, Sri Lanka	PR
		present study	Qld	Not accessioned
Isopoda				
Cymothoidae				
Cymothoa indica Schioedte & Meinert, 1884	Body surface	Rajkumar (2005) *	India	PR

^Note incorrect spelling in Rückert *et al.* 2008 (*celebensis* instead of *celebesensis*); **Likely to be the same taxon, requires further study.

Rachycentridae

Rachycentron canadum

Rachycentron canadum, the only member of the Rachycentridae, is a commercially and recreationally important species. It is commonly known as cobia, and is found in the warm-temperate to tropical waters of the West and East Atlantic, throughout the Caribbean and in the Indo-Pacific off India, Australia and Japan (Benetti *et al.* 2010). Cobia are generally solitary fish that occasionally occur in small schools that are often associated with floating objects such as fish-attracting devices, buoys and logs, and fixed structures such as oil rigs, piers and jetties (Velde *et al.* 2010). In Australia, cobia are not commonly encountered in large numbers, and therefore not currently a target species in any state or Commonwealth fishery (Fry and Griffiths 2010). However, the large size, fighting ability and superb eating qualities have made this species an important sportfish. Because of its fast growth rates, reaching 6–10 kg in 12–14 months, superb flesh quality, adaptability to cage-culture and strong resistance to diseases, cobia is also an excellent candidate for aquaculture (Sun *et al.* 2006; Velde *et al.* 2010).

Early attempts to culture cobia in the United States (US) began in the 1970s using eggs collected from the wild. Research accelerated in the late 1990s and today there now exists a large body of knowledge on cobia culture in the US (Benetti *et al.* 2010). In Taiwan, cobia cage aquaculture was initiated in the early 1990s (Liao *et al.* 2004). By 1999, four commercial hatcheries were operating, producing around 3 million cobia juveniles annually (Benetti *et al.* 2010). Since then, cobia aquaculture production has been steadily expanding in Asia, primarily in Taiwan, Vietnam and China, but also in other Southeast and Indo-Pacific Asian countries including the Philippines, Indonesia, Iran and Reunion Island. Floating cages continue to be the preferred method of growout worldwide. Most recently, Australia and Marshall Island have begun developing hatchery and cage culture operations for cobia (Benetti *et al.* 2010).

Several parasite species are problematic in cobia culture (see Liao *et al.* 2004 for review). Only captive fishes were able to be sampled for this research (CSIRO Cleveland and James Cook University, Queensland; Table 2). *Neobenedenia* sp. 2 was detected in fish sampled from James Cook University (Table 11). Cobia were originally

sourced from Good Fortune Bay Ltd, Bowen, and it is not clear whether fish were infected prior or post transported to the aquarium facility at James Cook University.

Table 11. Metazoan parasite fauna of wild and farmed *Rachycentron canadum* (cobia) collected in this study and presented in previous records from Australasia. Abbreviations: Aus, Australia; GM, Gulf of Mexico; IO, Indian Ocean; Med, Mediterranean; N/g, Not given; PO, Pacific Ocean; PR, Puerto Rico; RS, Red Sea; SCS, South China Sea; Qld, Queensland; USA, United States of America; *Farmed fish.

Parasite	Microhabitat	Reference	Location	Accession number
Myxozoa				
Sphaerospora-like myxosporean	Blood, glomerulus, renal tubules and renal interstitium	Chen *et al.* (2001)*	Taiwan	PR
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	N/g	Williams and Bunkley-Williams (1996)	Med	PR
Rhynchobothrium longispine Linton, 1890	N/g	Williams and Bunkley-Williams (1996)	USA	PR
Phyllobothriidae				
Rhinebothrium flexile Linton, 1890	Encapsulated in intestine, liver and viscera	Williams and Bunkley-Williams (1996)	USA	PR
Tentaculariidae				
Nybelinia bisulcata (Linton, 1889) Dollfus, 1929	Stomach, body cavity, intestine	Williams and Bunkley-Williams (1996)	USA	PR
Unknown family				
Tetraphyllid	Intestine, stomach, pyloric ceca, bile duct and gall bladder	Williams and Bunkley-Williams (1996)	N/g	PR
Cestoda gen. sp.	N/g	Arthur and Te (2006)	Mekong River Delta, Gulf of Tonkin	PR

Digenea				
Acanthocolpidae				
Stephanostomum cloacum (Srivastava, 1938) Manter & Van Cleave, 1951	N/g	Williams and Bunkley-Williams (1996)	IO	PR
Stephanostomum dentatum (Linton, 1900) Manter, 1931	Intestine	Williams and Bunkley-Williams (1996)	USA	PR
Stephanostomum imparispine (Linton, 1905) Manter, 1940	Intestine	Williams and Bunkley-Williams (1996)	Atlantic coast of USA	PR
	Body cavity, gills, inner organs, intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	PR
Stephanostomum microsomum Madhavi, 1976	N/g	Williams and Bunkley-Williams (1996)	IO	PR
Stephanostomum pseudoditrematis Madhavi, 1976	N/g	Williams and Bunkley-Williams (1996)	IO	PR
Stephanostomum rachycentri	N/g	Williams and Bunkley-	IO	PR

		Williams (1996)		
Tormopsolus filiformis So-Bernal & Hutton, 1958	Rectum	Williams and Bunkley-Williams (1996)	Gulf coast of Florida	PR
	Intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	PR
Tormopsolus spatulum Hafeezullah, 1978	N/g	Williams and Bunkley-Williams (1996)	IO	PR
Bucephalidae				
Bucephalus varicus Manter, 1940	Stomach, intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	PR
Cryptogonimidae				
Paracryptogonimus morosovi (Parukhin, 1965) Yamaguti, 1971	N/g	Williams and Bunkley-Williams (1996)	PO	PR
	Intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	
Derogenidae				
Derogenes varicus O.F. Müller, 1784	Stomach, intestine	Arthur and Te (2006)	Gulf of Tonkin	
Didymozoidae				
Neometanematobothrioides rachycentri (Parukhin, 1969) Yamaguti, 1971	N/g	Williams and Bunkley-Williams (1996)	PO	PR
	Body cavity, gills	Arthur and Te (2006)	Gulf of Tonkin, RS	
Gorgoderidae				
Phyllodistomum parukhini Yamaguti, 1971	N/g	Williams and Bunkley-Williams (1996)	RS	PR
	Kidney, urinary bladder	Arthur and Te (2006)	Gulf of Tonkin, RS	
Hemiuridae				
Dinurus selari Parukhin, 1966	Stomach, intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	

Lecithochirium canadus Bilqees, 1972	N/g	Williams and Bunkley-Williams (1996)	IO
Lecithochirium monticellii (Linton, 1898) Skrjabin & Guschanskaja, 1955	Intestine	Williams and Bunkley-Williams (1996)	USA
Lecithocladium jagannathi Ahmad, 1981	N/g	Williams and Bunkley-Williams (1996)	IO
Plerurus digitatus Looss, 1899	N/g	Williams and Bunkley-Williams (1996)	IO
Tubulovesicula angusticauda (Nicoll, 1915) Yamaguti, 1934	Stomach, intestine	Arthur and Te (2006)	Gulf of Tonkin, RS
Lecithasteridae			
Aponurus carangis Yamaguti, 1952	Stomach, intestine	Arthur and Te (2006)	Gulf of Tonkin, RS
Lepocreadiidae			
Lepidapedon megalaspi Paruchin, 1966	N/g	Williams and Bunkley-Williams (1996)	PO
	Intestine	Arthur and Te (2006)	Gulf of Tonkin, RS
Pseudolepidapedon pudens (Linton, 1900) Yamaguti, 1971	Intestine	Williams and Bunkley-Williams (1996)	USA
Mabiaramidae			
Mabiarama prevesiculata Freitas & Kohn, 1966	Stomach	Williams and Bunkley-Williams (1996)	Brazil
Aporocotylidae			
Psettaroides sp.	Heart	Bunkley-Williams and Williams (1996)	Northern GM

Sclerodistomidae				
Hemiuroidea gen. sp.	Intestine	Arthur and Te (2006)	Gulf of Tonkin, RS	
Sclerodistomum cobia Linton, 1905	N/g	McLean *et al.* (2008)	N/g	
Sclerodistomum rachycentri Parukhin, 1978	N/g	Williams and Bunkley-Williams (1996)	IO	
Unknown family				
Laruea straightum Jehan 1973	N/g	Williams and Bunkley-Williams (1996)	IO	
Monogenea				
Capsalidae				
Dioncus agassizi Goto, 1899	Gills	Williams and Bunkley-Williams (1996)	USA	
Dioncus rachycentris Hargis, 1955	Gills	Ogawa *et al.* (2006)	USA	
Neobenedenia girellae (Hargis, 1955) Yamaguti, 1963	Body surface	Ogawa *et al.* (2006)*	Taiwan	
Neobenedenia sp. 2	Cornea, body surface around head	present study*	Qld	AHC 45819–20; 35001–-02
Nematoda				
Anisakidae				
Anisakis sp. larva	Body cavity, gonads, kidneys, liver, stomach, intestine	Arthur and Te (2006)	Gulf of Thailand, Gulf of Tonkin, RS	
Goezia pelagia Deardorff & Overstreet, 1980	Stomach	Williams and Bunkley-Williams (1996)	USA	
	N/g	Akther *et al.* (2004)	Mexico	
Iheringascaris inquies (Linton,	Stomach	Williams and Bunkley-	USA	

1901) Deardorff and Overstreet, 1981	Intestine	Williams (1996) Arthur and Te (2006)	Gulf of Tonkin, RS
	Stomach	McLean *et al.* (2008)	Aus
Philometridae			
Philometroides sp.	Body cavity	Arthur and Te (2006)	Gulf of Tonkin
Unknown			
Nematoda gen. sp.	N/g	Arthur and Te (2006)	Mekong River Delta, Gulf of Tonkin, RS
Acanthocephala			
Rhadinorhynchidae			
Serrasentis nadakali George and Nadakal, 1978	Intestine	George and Nadakal (1981)	India
Serrasentis sagittifer (Linton, 1889) Van Cleave, 1923	Intestine, pyloric caeca, body cavity, mesenteries, external surfaces of internal organs	Williams and Bunkley-Williams (1996)	USA, Europe, West Africa
	Pyloric caeca, intestine	Arthur and Te (2006)	Gulf of Thailand, Gulf of Tonkin, RS
Copepoda			
Caligidae			
Caligus coryphaenae Steenstrup & Lutken, 1861	N/g	Williams and Bunkley-Williams (1996)	PO
Caligus epidemicus Hewitt, 1971	N/g	Ho *et al.* (2004)*	Philippines
Caligus haemulonis Krøyer, 1863	N/g	Williams and Bunkley-Williams (1996)	USA
Caligus lalandei Barnard, 1948	N/g	McLean *et al.* (2008)*	Taiwan
Lepeophtheirus plectropomi	N/g	Williams and Bunkley-	PO

Nuñes-Ruivo, 1956	N/g	Williams (1996) McLean *et al.* (2008)*	Taiwan
Parapetalus occidentalis Wilson, 1908	Gills	Williams and Bunkley-Williams (1996)	USA
Tuxophorus caligodes Wilson, 1908	Body surface	Williams and Bunkley-Williams (1996)	USA
Euryphoridae			
Euryphorus nordmanni Milne-Edwards, 1840	Inner surface of operculum, gill chamber, mouth	Williams and Bunkley-Williams (1996)	USA
Pennellidae			
Lernaeenicus longiventris Wilson, 1917	Body surface	Williams and Bunkley-Williams (1996)	USA
Lernaeolophus hemirhamphi Krøyer, 1863	N/g	Williams and Bunkley-Williams (1996)	GM
Lernaeolophus sultanus Nordmann, 1864	Body surface	Williams and Bunkley-Williams (1996)	USA

-Williams and Bunkley-Williams (1996) also report barnacles (Cirripedia: *Conchoderma virgatum*) from cobia

Sciaenidae

Argyrosomus japonicus

The mulloway *Argyrosomus japonicus* is a large, estuarine sciaenid distributed around southern Australia, southern Africa, Pakistan, India, China, Korea, and Japan (Silberschneider *et al.* 2009). In South Africa and Australia, mulloway is a highly desired commercial and recreational species, as well as an elusive sportfish (Fielder and Bardsley 1999; Taylor *et al.* 2006). In Australia, over 975 t of mulloway was taken by recreational fishers in 2000 (Taylor *et al.* 2006). This species is now classified as overfished in eastern Australia and there are concerns about sustainability, with the bulk of the fishery targeting immature fish (Silberschneider and Gray 2008; Taylor *et al.* 2009).

Mulloway is also an emerging aquaculture species in Australia, with good food conversion ratios and fast growth rates between 15–30 °C (Hayward *et al.* 2007b; Ballagh *et al.* 2010). This species can be farmed in sea cages, coastal earthen ponds and recirculating aquaculture systems. During the relatively short history of culture of mulloway, there have been few disease or parasite problems. However, juveniles are reportedly susceptible to ectoparasitic protozoans and brood stock in recirculation tanks are susceptible to problems with the monogenean, *Benedenia sciaenae* (see Hayward *et al.* 2007b).

We surveyed sea-caged mulloway from Botany Bay, New South Wales and wild mulloway from Port Adelaide South Australia as part of this project (Chapter 7). Ten parasite species were found and are presented with records published previously (Table 12). A parasite risk assessment is provided for sea-cage aquaculture of this species in Chapter 7.

Table 12. Metazoan parasite fauna of wild and farmed mulloway *Argyrosomus japonicus* collected in this study and presented in previous records from Australasia. Abbreviations: N/g, Not given; NSW, New South Wales; NZ, New Zealand; SA, South Australia; QLD, Queensland; WA, Western Australia; VIC, Victoria; *Farmed fish.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Empty cysts	Body cavity	present study	SA	Not accessioned
Dasyrhynchidae				
Dasyrhynchus pacificus Robinson, 1965	Viscera	Beveridge and Campbell (1993)	NSW	PR
Otobothriidae				
Poecilancistrium calyophyllum Diesing, 1850	Encysted in flesh	Robinson (1965)	NSW	PR
Pterobothriidae				
Pterobothrium sp. Diesing, 1850	viscera	Young (1939)	NSW	PR
Digenea				
Acanthocolpidae				
Stephanostomum bicoronatum (Stossich, 1883) Fuhrmann, 1928	Intestine	Bray and Cribb (2003b)	QLD	PR
Pleorchiidae				
Pleorchis sciaenae Yamaguti, 1938	Intestine	Bray (1986)	South Africa	PR
Monogenea				
Calceostomatidae				
Calceostoma glandulosum Johnson & Tiegs, 1922	Gills	Yamaguti (1963)	QLD	PR
	Gill filaments	Hayward *et al.* (2007b)	SA*	PR
	Gills	present study	NSW*, SA	AHC 35003
Capsalidae				
Benedenia sciaenae van Beneden, 1856	Skin	Whittington (1996)	SA	PR
		present study	NSW*, SA	AHC 45795

Diplectanidae				
Diplectanum sp.	Gills	Williams (1989)	WA	PR
	Gills	present study	NSW*, SA	AHC 35005
Diplectanum glandulosum Williams, 1989	Gill filaments	Williams (1989)	WA	PR
Diplectanum oliveri Williams, 1989	Gill filaments	Williams (1989)	WA	PR
Microcotylidae				
Sciaenacotyle sciaenicola Murray, 1932	Gills	Young (1970)	QLD	PR
	Gills	Hayward *et al.* (2007)	SA*	PR
	Gills	present study	NSW*, SA	AHC 35004
Nematoda				
Philometra sp.	Gonad	present study	SA	Not accessioned
Ascarididae				
Ascaris sp.	N/g	Stead (1914)	VIC	PR
Anisakidae				
Anisakis sp.	Location unspecified	Johnston and Mawson (1951)	VIC	PR
Hysterothylacium marinum Linnaeus, 1767	Location unspecified	Johnston and Mawson (1943)	SA	PR
Terranova sp.	Viscera	Cannon (1977)	SA	PR
Contracaecum (Thynnascaris) *legendrei*	Location unspecified	Johnson & Mawson (1951)	QLD	PR
Dollfus 1933	Location unspecified	Yamaguti (1961)	VIC	PR
Acanthocephala				
Neoechinorhynchidae				
Neoechinorhynchus dorsovaginatus Amin & Christison, 2004	Hind gut (rectum)	Amin and Christison (2005)	South Africa	PR

Copepoda				
Caligidae				
Caligus sp. (male)	Gills	present study	SA	SAMA C6888
Caligus chiastos Lin & Ho, 2003	Gills	Hayward *et al.* (2007)	SA*	PR
Lernanthropidae				
Isobranchia scianophila Heller, 1868	Gill arch	present study	SA	SAMA C6890
Lernanthropus gisleri Van Beneden, 1852	Gills	present study	SA	SAMA C6889
Isopoda				
Ceratothoa sp.	Mouth	present study	NSW	SAMA C6929
Cymothoidae				
Nerocila sigani Bowman & Tareen	N/g	Bruce and Harrison-Nelson (1988)	South Africa	PR

Sillaginidae

Sillaginodes punctatus

The King George whiting, *Sillaginodes punctatus*, which is the largest of the 31 species in the Sillaginidae and also considered the most valuable, is endemic to southern Australia where it occurs throughout the inshore, coastal area (Hyndes *et al.* 1998; Fowler *et al.* 2000). Post-larvae and juveniles recruit to seagrass habitats in sheltered bays and inlets throughout winter and spring, whereas adult fish live in coastal waters, spawning offshore in autumn and winter (Jenkins and May 1994; Jenkins and Welsford 2002).

This species comprises one of the most important inshore finfish fisheries in southern Australia (Jenkins 2005), with each state managing this fishery independently according to different regulations and policies (Fowler *et al.* 2000). The fishery for *S. punctatus* in SA in particular is the most valuable, producing in excess of 75% of the national annual catch (Fowler *et al.* 2002). The harvested biomass in SA is on average about 5 times greater than Victoria and 20 times that of Western Australia (McGarvey *et al.* 2005). This fishery has the potential to be strongly influenced by recruitment variability, because although the species can live up to 20 years, exploitation is primarily concentrated on subadults of 3 to 5 year old (Jenkins 2005). King George whiting also has recreational importance.

Sillaginodes punctatus is the study species of currently enrolled Honours student, Emma Brock at the University of Adelaide. Emma's project received additional support through an ABRS capacity-building scholarship. She is currently writing her thesis and will develop a final report for ABRS (due February 2011). A new species of aporocotylid trematode, *Cardicola* n. sp., has been discovered to infect *S. punctatus* (Table 13) and will be included in a manuscript that examines at least three other *Cardicola* species found in this study (see Tables 14, 19 and 22).

Table 13. Metazoan parasite fauna of King George Whiting *Sillaginodes punctatus* collected in this study and presented in previous records from Australasia. [A] = new host record. Abbreviations: N/g, Not given; NSW, New South Wales; SA, South Australia; WA, Western Australia; VIC, Victoria.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Lacistorhynchidae				
Callitetrarhynchus gracilis Rudolphi, 1819	Intestinal tract, flesh	Beveridge & Campbell (1996)	SA	PR
Order: Tetraphyllidea Carus, 1863	Intestinal tract	present study [A]	SA	AHC 29999
Otobothriidae				
Poecilancistrium sp.	Intestinal tract, flesh		SA	PR
Digenea				
Lepocreadiidae				
Austraholorchis procerus Bray *et al.* 1999	N.g	Bray *et al.* (1999)	SA	
	Intestinal tract	present study	SA	AHC 29998
Lepidapedella sillaginodesi Bray *et al.* 1999	N/g	Bray *et al.* (1999)	SA	
	Intestinal tract	present study	SA	AHC 29996-97
Opecoelidae				
Macvicaria adomeae Aken'Ova *et al.*, 2008	Gut	Aken'Ova *et al* (2008)	SA	
	Intestinal tract	present study	SA	Not accessioned
Macvicaria shotteri Aken'Ova *et al.*, 2008	Intestine, gut	Aken'Ova *et al.* (2008)	WA, SA	
	Intestinal tract	present study	SA	Not accessioned
Macvicaria vitellocopiosa Aken'Ova *et al.*, 2008	Intestine, gut	Aken'Ova *et al.* (2008)	SA	
	Intestinal tract	present study	SA	Not accessioned
Aporocotylidae				
Cardicola n. sp.	Heart	present study [A]	SA	AHC 29994–45

Monogenea				
Microcotylidae				
Polylabris sillaginae Woolcock, 1936	Gill filaments	Hayward (1996)	SA, WA, Vic, NSW	PR
	Gills	present study	SA	AHC 29991–93
Nematoda				
Anisakidae				
Contracaecum sp.	Viscera	Lymbery *et al.* (2002)	WA	PR
Hysterothylacium sp.	Intestinal tract	present study [A]	SA	AHC 45013–14
Cucullanidae				
Cucullanellus sp.	Gut	Johnston and Mawson (1945)	N/g	PR
	Intestinal tract	present study	SA	AHC 45015–16
Cystidicolidae				
Ascarophis cooperi Johnston & Mawson, 1945	Intestinal tract	Johnston and Mawson 1945	SA	PR
Echinocephalus sp.	Intestinal tract	present study [A]	SA	AHC 45809–12
Acanthocephala				
Polymorphidae (larval)	Intestinal tract	present study [A]	SA	AHC 45807–08
Copepoda				
Bomolochidae				
Nothobomolochus sp.	Gills	present study [A]	SA	SAMA C6885
Caligidae				
Caligus longirostris Heegaard, 1962	Skin	Heegaard (1962)	N/g	PR
Lernaeopodidae				
Anaclavella sillaginoides Heegaard, 1962	Gills	Heegaard (1940)	SA	PR
	Gills	present study	SA	SAMA C6886–87
Isopoda				
Gnathiidae (juvenile)	Gills	Present study [A]	SA	C 6936-38

Sparidae

Chrysophrys auratus

The pink snapper *Chrysophrys auratus* (syn. *Labrus auratus, Pagrus auratus*, *P. latus*) lives at depths up to 200 m off the coasts of New Zealand, Australia, Philippines, Indonesia, China, Taiwan and Japan (Sharples and Evans 1995b). It is a demersal predatory fish that is heavily exploited by commercial and recreational fishers (Willis *et al.* 2001). It is also a well known sportfish in southern Australia with flesh of excellent quality (Gomon *et al.* 2008).

Southern and northern populations were considered to be separate species but are now regarded as one species with independent and reproductively isolated populations in Japan and Australasia (Paulin 1990). The northern population has been intensively and successfully farmed for many years in Japan (Foscarini 1988; Hattori *et al.* 2004), with about 40,000 t being produced in 1990 (Battaglene and Talbot 1992). In Japan, the most popular choice for culture is the use of floating net-cages in closed bays protected from sea storms and strong currents. Concrete tanks on land are also suitable, however are more expensive to construct and electrical pumps are needed to supply seawater (Foscarini 1988). The southern population on the other hand is not commercially cultured, although it is considered a prime aquaculture candidate for Australasia and commands a high market profile and price (Battaglene and Talbot 1992). Attempts to culture this species have been unsuccessful and may be attributed to a number of issues, including availability of quality eggs (Cleary and Pankhurst 2000), slow growth of snapper and dark appearance of the skin when held in sea cages (Doolan et al. 2008).

We concentrated our sampling effort on the heart of *C. auratus*, considering that parasite fauna for this species has been well documented in Australia (Table 14). A new species of aporocotylid trematode cf. *Cardicola* was found and will be included in a manuscript that examines at least three other *Cardicola* species from this study (see Table 13, 19 and 22).

Table 14. Metazoan parasite fauna of wild and farmed pink snapper *Chrysophrys auratus* collected in this study and presented in previous records from Australasia. Abbreviations: Aus, Australia; EA, Eastern Australia; NEC, North East Coast; N/g, Not given; NSW, New South Wales; NZ, New Zealand; SA, South Australia; Tas, Tasmania; Vic, Victoria; WA, Western Australia; *Farmed fish.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Dasyrhynchidae				
Plerocercoids	Intestine	Sharples and Evans (1995b)	NZ	PR
Phyllobothriidae				
Plerocercoids	Intestine	Sharples and Evans (1995b)	NZ	PR
Digenea				
Aporocotylidae				
cf. *Cardicola* n. sp.	heart	Present study	SA, NSW	Not accessioned
Didymozoidae				
Gonapodasmius williamsoni Cribb and Williams, 1992	Flesh	Williams *et al.* (1993)	WA	PR
Fellodistomidae				
Proctoeces sp.	Stomach, intestine	Sharples and Evans (1995a)	NZ	PR
Zoogonidae				
Diphtherostomum sp.	Rectum	Sharples and Evans (1995a)	NZ	PR
Monogenea				
Anoplodiscidae				
Anoplodiscus cirrusspiralis Roubal *et al.*, 1983	Pectoral, dorsal, caudal, anal & pelvic fins	Roubal *et al* (1983)	NSW, SA, NZ	PR
	Caudal, pectoral fins, nasal lamella	Roubal *et al* (1992)*	NSW	PR
Capsalidae				
Benedenia sekii (Yamaguti, 1937) Meserve,	Body surface	Roubal *et al.* (1983)	NSW, SA, NZ	PR

1938	Beneath pectoral fin	Sharples and Evans (1995c)	NZ, Sea of Japan	PR
Encotyllabe pagrosomi MacCallum, 1917	Mouth, pharynx	Beumer (1983)	Aus	PR
	Undetermined	Roubal *et al.* (1983)	NSW	PR
Choricotylidae				
Choricotyle australiensis Roubal *et al.*, 1983	Gill arches	Roubal *et al.* (1983)	NSW, Vic, SA, NZ	PR
Diplectanidae				
Lamellodiscus pagrosomi Murray, 1931	Gill filaments	Roubal *et al.* (1983)	NSW, SA, NZ	PR
	Gill filaments	Sharples & Evans (1995c)	Vic, Japan	PR
Microcotylidae				
Aspinatrium spari (Yamaguti, 1937) Yamaguti, 1963	N/g	Sharples & Evans (1995c)	NEC Aus	PR
Bivagina pagrosomi Murray, 1931	Gill filaments	Roubal *et al.* (1983)	NSW, SA, NZ	PR
	Gill filaments	Sharples & Evans (1995c)	Vic	PR
	Gills	Beumer *et al.* (1982)	Bass Strait, Tas	PR
Bivagina tai Ogawa, 2004	N/g	Sharples & Evans (1995c)	Japan	PR
Nematoda				
Anisakidae				
Anisakis sp.	Liver	Sharples & Evans (1995b)	NZ	PR
Hysterothylacium marinum Linnaeus, 1767	N/g	Johnston & Mawson (1945)	SA	PR
Cucullanidae				
Cucullanus sheardi Johnston & Mawson, 1944	N/g	Johnston & Mawson (1945)	SA	PR
Cucullanus sp.	Intestine	Sharples and Evans (1995a)	NZ	PR

Gnathostomatidae				
Echinocephalus uncinatus Molin, 1858	Viscera	Johnston & Mawson (1945)	SA	PR
Philometridae				
Philometra lateolabracis Yamaguti, 1935	Gonad	Sharples and Evans (1995b)	NZ	PR
Copepoda				
Bomolochidae				
Naricolax chrysophryenus Roubal *et al.* 1983	Pectoral fin	Roubal *et al.* (1983)	NSW, SA, NZ	PR
	Nasal cavities	Sharples and Evans (1995a)	NZ	PR
Pseudoeucunthus australiensis, Roubal *et al.* 1983	Unknown	Roubal *et al.* (1983)	NSW	PR
Caligidae				
Caligus chiastos Lin & Ho, 2003	N/g	Ho and Lin (2004)	EA	PR
Caligus laticaudus Shiino, 1960	N/g	Sharples and Evans (1995c)	Japan	PR
Caligus sclerotinosus Roubal *et al.*, 1983	Body surface	Roubal *et al.* (1983)	NSW, SA	PR
Caligus sp. 1	Uncertain	Roubal *et al.* (1983)	NSW	PR
	Body surface	Sharples and Evans (1995c)	NZ	PR
Caligus sp. 2	Uncertain	Roubal *et al.* (1983)	NSW	PR
	Body surface	Sharples and Evans (1995c)	NZ	PR
Caliss willungue Kabata, 1965	Body surface	Roubal *et al.* (1983)	NSW, SA	PR
Lepeophtheirus sekii Yamaguti, 1936	Body surface	Roubal *et al.* (1983)	NSW	PR
	Body surface	Sharples and Evans (1995c)	NZ, Japan	PR
Lepeophtheirus sp. 1	Uncertain	Roubal *et al.* (1983)	NSW	PR
Hatschekiidae				

Hatschekia pagrosomi Yamaguti, 1939	Gill filaments	Roubal *et al.* (1983)	NSW, Vic, SA, NZ	PR
Lernaeopodidae				
Clavellopsis sargi (Kurz, 1877)	Mouth cavity	Roubal *et al.* (1983)	NSW, Vic, SA, NZ	PR
Yamaguti, 1963	N/g	Sharples and Evans (1995c)	Japan	PR
Lernanthropidae				
Lernanthropus atrox Heller, 1865	Gil filaments	Roubal *et al.* (1983)	NSW	PR
	N/g	Sharples and Evans (1995c)	Japan	PR
Isopoda				
Cymothoidae				
Ceratothoa imbricatus Fabricius, 1787	Operculum, mouth	Beumer *et al.* (1982)	Aus	PR
Nerocila monodi Hale, 1940	N/g	Roubal *et al.* (1983)	NSW	PR
Gnathiidae				
Praniza larvae	Gill filament	Roubal *et al.* (1983)	NSW, SA	PR

2.4.3 Non-target fish species sampled

For non-target fishes we focused our efforts on recovering monogeneans, aporocotylid trematodes and copepods.

Carangidae

Elagatis bipinnulata

The rainbow runner, *Elagatis bipinnulata*, is considered an important game fish and occurs circumtropically in marine waters (Berry 1969; Ditty *et al.* 2004). Adults grow to a length of about 4 ft and are fast-swimming fish, usually found near the surface in oceanic waters, sometimes far offshore (Berry 1969). One copepod species and one monogenean species were recorded from *E. bipinnulata* in this study (Table 15).

Table 15. Metazoan parasite fauna of rainbow runner *Elagatis bipinnulata* collected in this study. Abbreviations: NSW, New South Wales.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Caligus confusus Pillai, 1961	Gill arch	present study	NSW	C6902; NHM 2009.275
Monogenea				
Gotocotylidae				
Gotocotyla elagatis Meserve, 1938	Gills	Perkins (2010) present study	NSW	AHC 29732

Seriola dumerili

The greater amberjack, *Seriola dumerili*, is distributed in subtropical coastal waters and the Atlantic, Pacific and Indian Oceans (Rowland 2009) and is currently farmed in Japan and the Spanish Mediterranean. It has many characteristics that satisfy the criteria for the selection of a new species for aquaculture, such as excellent flesh quality and a high market price (Jerez *et al.* 2006). Two aporocotylid trematodes were found (Table 16) which are discussed in detail in Chapter 5.

Table 16. Metazoan parasite fauna of amberjack *Seriola dumerili* collected in this study. Abbreviations: WA, Western Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Aporocotylidae				
Aporocotylidae				
Paradeontacylix n. sp.	Heart	present study	WA	Not accessioned
Paradeontacylix cf. *kampachi*	Heart	present study	WA	Not accessioned

Centrolophidae

Hyperoglyphe antarctica

The deep-sea trevalla (*Hyperoglyphe antarctica*), or bluenose as it is commonly called in NZ, is widespread in southern oceans occuring off the coasts of NZ, southern Australia and Tasmania, South Africa and Tristan da Cunha in the southern Indian Ocean (Horn 1988). Adults of this species are described as "semi-pelagic," living close to the sea floor at depths of 100 and 500 m, but regularly making feeding excursions into the water column (Duffy *et al.* 2000). Juveniles however are surface living, often sheltering under drift algae and flotsam. It is regarded as an important commercial species in Australian waters (Horn 2003). Two parasite species were recorded from *H. antarctica* in this study (Table 17).

Table 17. Metazoan parasite fauna of deep-sea trevalla *Hyperoglyphe antarctica* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Lernanthropidae				
Lernanthropus microlamini Hewitt, 1968	Gills	present study	SA	C6900 NHM 2007.948–49
Monogenea				
Diclidophoridae				
Eurysorchis manteri Mamaev, 1976	Gills	Perkins (2010) present study	SA	AHC 29730 AHC 29973

Cheilodactylidae

Nemadactylus valenciennesi

The blue morwong, also known as queen snapper *Nemadactylus valenciennesi,* lives over rocky reefs in the waters of southern Australia. On the south coast of WA, this species is the greatest contributor to the biomass of the catches taken by the demersal gill-net and longline fishery and one of the species most frequently retained by recreational fishers (Coulson *et al.*

2010). We recorded 8 parasite species from this host species in this study (Table 18) including new monogenean species for which we are currently preparing new species descriptions.

Table 18. Metazoan parasite fauna of queen snapper *Nemadactylus valencienesi* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Lepeophtheirus sp. 1	Body surface, fins	present study	SA	SAMA C6892–94
Lepeophtheirus sp. 2	Body surface	present study	SA	SAMA C6895
Lernanthropidae				
Aethon morelandi Hewitt, 1968	Gills	present study	SA	SAMA C6891
Monogenea				
Capsalidae				
Capsalidae sp. 2 of Perkins *et al.* (2009)	Pectoral fins	Perkins *et al.* (2009)	SA	AHC 29659
	Pectoral fins, gills-artefact, FW bath	present study	SA	AHC 29975–78
Mediavagina sp. of Perkins *et al.* (2009)	Gills	Perkins *et al.* (2009)	SA	AHC 29669
	Gills, rakers, toothpads	present study	SA	AHC 29956-61, AHC 29982
Dactylogyridae				
Dactylogyridae sp. of Perkins *et al.* (2009)	Gills	Perkins *et al.* (2009)	SA	AHC 29662
	Gills	present study	SA	

Nemadactylus macropterus

Nemadactylus macropterus (jackass morwong, tarakihi) is a demersal species commonly found in coastal and continental shelf waters of southern Australia, New Zealand, southern South America, southern Africa and some islands in the Atlantic and Indian oceans (Jordan 2001b; NSWDPI 2008b). Jackass morwong are distributed in Australian waters from Moreton Bay in Queensland to Perth in WA. They occur in depths to 450 m and, in Australian waters, are most abundant between 100 and 200 m (NSWDPI 2008b). Jackass morwong is an important commercial species in the South East Fishery, with the bulk of landings taken off southern New South Wales, eastern Bass Strait and eastern Tasmania (Jordan 2001b). We recorded 8 parasite species from jackass morwong (Table 19), including *Cardicola whitteni* which will be redescribed in a manuscript that examines at least three other *Cardicola* species found in this study (see Table 13, 14 and 22).

Table 19. Metazoan parasite fauna of jackass morwong *Nemadactylus macropterus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Lepeophtheirus sp.	Body surface, fins	present study	SA	SAMA C6898–99
Lernanthropidae				
Aethon garricki Hewitt, 1968	Gills	present study	SA	SAMA C6896; BMNH 2007.951
Clavella group	Gills	present study	SA	SAMA C6897
Monogenea				
Capsalidae				
Encotyllabe chironemi Robinson, 1961	Pharyngeal tooth pads, gills	Perkins (2010) present study	SA	AHC 29722 AHC 29979–81
Mediavagina macropteri Lawler and Hargis, 1968	FW bath	present study	SA	AHC 29983
Microcotylidae				
Microcotyle nemadactylus Dillon & Hargis, 1965	Gills	Perkins (2010) present study	SA	AHC 29729 AHC 29984–88
Trematoda				
Aporocotlyidae				
Cardicola whitteni Manter, 1954	Heart	present study	SA	Not accessioned

Kyphosidae

Girella tricuspidata

Girella tricuspidata, commonly known as luderick, inhabits estuaries and coastal waters along the eastern and southern coast of Australia and around the North Island of New Zealand (Smith and Sinerchia 2004). It is primarily a herbivorous fish, feeding predominately on seagrass and algae, although it also consumes small benthic invertebrates (Gray *et al.* 2010). *Girella tricuspidata* is taken in large quantities in estuarine and coastal commercial and recreational fisheries (Pollock 1981; Gray *et al.* 2010). One parasite species was recorded in this study (Table 20).

Table 20. Metazoan parasite fauna of luderick *Girella tricuspidata* collected in this study. Abbreviations: NSW, New South Wales.

Parasite	Microhabitat	Reference	Location	Accession number

Monogenea				
Monocotylidae				
Polylabris girellae Hayward, 1996	Gills	present study	NSW	AHC 29968

Latridae

Latris lineata

Striped trumpeter *Latris lineata* is a demersal species inhabiting the southern temperate waters of Australia and New Zealand (Morehead *et al.* 2001; Trotter *et al.* 2001). The species was being investigated in the Tasmanian Aquaculture and Fisheries Institue as a candidate species for temperate marine aquaculture in Australia (Cobcroft *et al.* 2001). We recorded four parasite species from wild striped trumpeter in this study (Table 21).

Table 21. Metazoan parasite fauna of striped trumpeter *Latris lineata* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Lernanthropidae				
Aethon n. sp.	Gills	present study	SA	SAMA C6901; NHM 2009.276
Monogenea				
Capsalidae				
Allomegalocotyla johnstoni (Robinson, 1961) Yamaguti, 1963	Gills	Perkins (2010) present study	SA	AHC 29727 AHC 29954
Capsalidae sp. 19 of Perkins (2010)	Pectoral fin, gills	Perkins (2010) present study	SA	AHC 29716 AHC 29962-63
Pseudomegalocotyla latridis Robinson, 1961) Yamaguti, 1963	Gills	present study	SA	AHC 29955

Mugilidae

Aldrichetta forsteri

Yelloweye mullet, *Aldrichetta forsteri*, is a coastal species endemic to southern Australia with a distribution from Shark Bay on the Western Australian coast to Newcastle on the coast of New South Wales. They are common over sandy bottoms in embayments and estuaries (Edmonds *et al.* 1992). This species comprises a substantial proportion of the commercial and amateur estuarine fishery in most of the river systems of south-western Australia (Chubb *et al.* 1981). They are also sought by recreational fishers (Edmonds *et al.* 1992). A new

species of *Cardicola* was found (Table 22) and will be described in a manuscript that examines at least three other *Cardicola* species found in this study (see Tables 13, 14 and 19).

Table 22. Metazoan parasite fauna of yelloweye mullet *Aldrichetta forsteri* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Aporocotylidae				
Aporocotylidae				
Cardicola n. sp.	Heart	present study	SA	Not accessioned

Oplegnathidae

Oplegnathus woodwardi

Oplegnathus woodwardi, commonly known as knifejaw due to its beak-like jaws that are well adapted for crushing hard-shelled invertebrates, is distributed throughout southern Australia, mostly offshore, at depths of 50 to 400 m. It is frequently taken by trawlers but not in commercial quantities and is regarded as an excellent food fish (Gomon *et al.* 2008). One copepod species was recorded from knifejaw in this study (Table 23) and is redescribed (see Chapter 4, section 4.2).

Table 23. Metazoan parasite fauna of knifejaw *Oplegnathus woodwardi* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Trebiidae				
Kabataia ostorhynchi Kazachenko, Korotaeva & Kurochkin, 1972	Gills	present study	SA	NHM 2007. 947, NHM 2009.263–64, NHM 2009.265–74

Percichthyidae

Polyprion oxygeneios

Polyprion oxygeneios, or hapuka as it is commonly called in New Zealand, is highly sought after commercially and recreationally in the southern Indian and Pacific Oceans (Francis *et al.* 1999). Juveniles are thought to live a pelagic existence in surface waters well offshore, whereas adults are demersal in depths of 50-600 m, becoming vulnerable to capture by bottom trawlers (Beentjes and Francis 1999). Three parasite species were recorded from *P. oxygeneios* in this study (Table 24).

Table 24. Metazoan parasite fauna of hapuka *Polyprion oxygeneios* collected in this study. [A] = new Autralian record. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Lepeophtheirus polyprioni Hewitt, 1963	Body surface	present study	SA	SAMA C6905
Monogenea				
Capsalidae				
Calicobenedenia sp.	Caudal fin	present study	SA[A]	AHC 29965
Discocotylidae				
Allocotylophora polyprionum Dillon and Hargis, 1965	Gills	present study	SA	AHC 29964

Polyprion americanus

The wreckfish *Polyprion americanus* is a large demersal fish that inhabits continental and oceanic island slopes of temperate and subtropical waters at both sides of the Atlantic Ocean, at the Mid-Atlantic Ridge, the Mediterranean, southern Indian Ocean and southern Pacific near southern Australia and New Zealand (Peres and Haimovici 2004). Adults live in depths between 100 to 600 m, whereas juveniles occur in surface waters around floating objects or pieces of wreckage, hence giving them their common name (NSWDPI 2008a). This species is characterised by a long-lived pelagic phase and is of high commercial importance (Ball *et al.* 2000; Peres and Klippel 2003). Genetic studies have revealed there are at least three distinct stocks of this species (NSWDPI 2008a). Wreckfish is also a candidate aquaculture species due to the high market price, high growth rate during the pelagic phase and good quality flesh (Machias *et al.* 2003; Papandroulakis *et al.* 2004; Fauvel *et al.* 2008). We recorded three parasite species from this host (Table 25).

Table 25. Metazoan parasite fauna of wreckfish *Polyprion americanus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Lepeophtheirus polyprioni Hewitt, 1963	Caudal fin, gills	present study	SA	SAMA C6903–04
Monogenea				
Capsalidae				
Calicobenedenia polyprioni Kritsky and Fennessy, 1999	Caudal fin	Perkins (2010) present study	SA	AHC 29736

Discocotylidae *Allocotylophora polyprionum* Dillon and Hargis, 1965	Gills	present study	SA	AHC 29966–67

Platycephalidae

Platycephalus aurimaculatus

Platycephalids are flattened dorso-ventrally, with a depressed head, eyes that are directed upwards and the swim bladder is absent from most species, a body form which reflects their benthic habitat (Keenan 1991). A total of 16 species within *Platycephalus* are known in temperate Australian waters (Imamura and Knapp 2009), many of which are of considerable commercial and recreational importance (Jordan 2001a). One cestode was recorded from flathead in this study and was decribed by Kutcha *et al.* (2009) (Table 26; Appendix 3).

Table 26. Metazoan parasite fauna of flathead *Platycephalus aurimaculatus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Cestoda				
Bothriocephalidae *Bothriocephalus australis* Kutcha *et al*., 2009	Intestine	present study	SA	AHC 29643

Scorpaenidae

Helicolenus percoides

Seaperches in *Helicolenus* are widespread, bottom-living teleosts that occur on the continental shelf and slope and offshore seamounts and ridges in temperate waters of the Pacific, Atlantic and Indian oceans. *Helicolenus barathri* was previously recognised in New Zealand waters, however molecular information suggests there is a single species, *H. percoides*. *Helicolenus percoides*, or red gurnard perch, is also found off the eastern coast of Australia in offshore (>300 m) waters of New South Wales (Smith *et al.* 2009). Two monogenean species were recorded from *H. percoides* in this study (Table 27).

Table 27. Metazoan parasite fauna of red gurnard perch *Helicolenus percoides* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Monogenea				
Megalobenedenia helicoleni (Woolcock, 1936) Egorova, 1994	Gills	Perkins *et al.* (2009)	SA	AHC 29670
	Gill arch	present study	SA	AHC 29974
Microcotylidae				
Microcotyle neozealanicus Dillon & Hargis, 1965	Gills	Perkins (2010) present study	SA	AHC 29731 AHC 29971–72

Sparidae

Acanthopagrus butcheri

The black bream, *Acanthopagrus butcheri,* is distributed in temperate waters from Myall Lake, New South Wales to Murchison River, Western Australia, where it is common in estuaries and river mouths (Ferguson and Ye 2008). This species completes their entire life cycle within the natal estuarine environment and is capable of tolerating a wide range of salinities (Partridge and Jenkins 2002). Black bream supports commercial fisheries in Victoria, South Australia and Western Australia with most catches taken from Victoria. This species is also captured by recreational anglers (Morison *et al.* 1998; Ferguson and Ye 2008). One caligid species was recorded from black bream in our study (Table 28).

Table 28. Metazoan parasite fauna of black bream *Acanthopagrus butcheri* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Caligidae				
Caligus epidemicus Hewitt, 1971	Body surface	present study	SA	SAMA C6927–28

Triakidae

Galeorhinus galeus

The tope or school shark, *Galeorhinus galeus*, is a medium-sized shark that occurs in coastal and shelf temperate waters in the northeast and southeast Pacific, northeast and south Atlantic, Mediterranean Sea, southern Australia and New Zealand (Lucifora *et al.* 2006). Its life history is characterised by slow growth, high longevity, late age at maturity, low

fecundity and low mortality (Lucifora *et al.* 2004). In southern Australia, this shark species is an important component of the shark fishery (Punt and Walker 1998). We recorded two copepod species from school shark in this study (Table 29).

Table 29. Metazoan parasite fauna of *Galeorhinus galeus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Kroyeriidae				
Kroyeria sp.	Gills	present study	SA	SAMA C6923–24
Pandaridae				
Nesippus orientalis Heller, 1868	Gills	present study	SA	SAMA C6925

Mustelus antarcticus

The endemic Australian gummy shark, *Mustelus antarcticus*, is a small demersal species that is the main target of the southern shark fishery, which operates primarily in waters off Victoria, Tasmania and South Australia (MacDonald 1988; Gardner and Ward 1998). This species is marketed as 'flake' and is found at least from Geraldton (Western Australia) to Port Stephens (New South Wales), and possibly ventures into southern Queensland (Gardner and Ward 1998; Gomon *et al.* 2008). We recorded 5 parasite species from *M. antarcticus* in this study (Table 30).

Table 30. Metazoan parasite fauna of gummy shark *Mustelus antarcticus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Eudactylinidae				
Eudactylina sp.	Gills	present study	SA	NHM 2007.942–43
Kroyeriidae				
Kroyeria sp.	Gills	present study	SA	SAMA C6923–24
Pandaridae				
Nesippus orientalis Heller, 1868	Gills	present study	SA	SAMA C6925
Monogenea				
Hexabothriidae				
Erpocotyle antarctica Hughes, 1928	Gills	Perkins (2010) present study	SA	AHC 29725 AHC 29969
Monocotylidae				
Triloculotrema japanicae Kearn, 1993		Perkins (2010)		AHC 29733

Urolophidae

Urolophus paucimaculatus

The sparsely spotted stingaree, *Urolophus paucimaculatus,* is a sandy bottom dweller living at depths up to 150 m that feeds on crustaceans and polychaetes. It is endemic to southern Australian waters and reaches a total length of 47 cm. This species is not often retained by fishers (Gomon *et al.* 2008). One copepod was recorded from *U. paucimaculatus* in this study (Table 31).

Table 31. Metazoan parasite fauna of sparsely spotted stingaree *Urolophus paucimaculatus* collected in this study. Abbreviations: SA, South Australia.

Parasite	Microhabitat	Reference	Location	Accession number
Copepoda				
Lernaeopodidae				
Pseudocharopinus sp.	Spiracles	present study	SA	NHM 2007. 950

3 BIOGEOGRAPHICAL RELEVANCE OF PARASITE-HOST CHECKLISTS: A CASE STUDY

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3.1 Abstract

Southern garfish *Hyporhamphus melanochir* were examined for metazoan parasites from nine sites in three regions (Spencer Gulf, Gulf St Vincent and northern Kangaroo Island) in South Australia to document parasite assemblages, identify candidate species suitable for use as biological tags and investigate spatial variation in parasite abundance. Four ectoparasite species and ten endoparasite species were identified representing Cestoda, Trematoda, Monogenea, Nematoda, Acathocephala, Copepoda and Isopoda. *Lernaeenicus hemirhamphi*, *Micracanthorhynchina hemirhamphi*, *Mothocya halei* and *Philometra* sp. were suggested 'permanent' biological markers. Multivariate discriminant function analysis showed that most sites could be distinguished based on differences in parasite abundance. Four endoparasite species (*Conohelmins* sp., *Hysterothylacium* sp., *Micracanthorhynchina hemirhamphi* and *Philometra* sp.) were most important for site characterisation. Limited spatial variation in permanent endoparasite abundance among localities in northern Spencer Gulf provides evidence for a distinct northern Spencer Gulf population with little inter-regional mixing. In contrast, considerable spatial variation in permanent endoparasite abundance between localities sampled off Kangaroo Island implies limited local movement and suggests *Hyporhamphus melanochir* may comprise a metapopulation structure. These results largely align with recent evidence from otolith chemistry that indicates fine-scale geographical population structuring in South Australian waters.

3.2 Introduction

Knowledge of the geographic population structure of fished populations is critical for maintaining overexploited fisheries as it helps determine the spatial scale at which populations should be monitored, assessed and managed. Among the tools for geographic

population determination, parasites have been used on numerous occasions to infer population structure and movement of hosts (e.g. MacKenzie 1987; Boje *et al.* 1997; Timi 2003; Zischke *et al.* 2009). Some parasites may be used as biological tags for populations, where spatial differences in the behaviour and/or feeding habits of host populations, or in the abundance of intermediate hosts, can give rise to significantly different levels of infection.

Parasites should be selected and applied to population delineation cautiously. Variation in parasite fauna and abundance does not necessarily mean that fish with different parasites are from different populations (Lester and MacKenzie 2009). In order to assess the suitability of parasites as biological markers for commercially fished stocks there is a fundamental need to survey large numbers of fish across broad geographic scales to determine patterns of spatial variation and identify candidate parasite species. Rigorous taxonomic identification therefore underpins appropriate management of overexploited stocks (e.g. Dulvy and Reynolds 2009b). Several criteria are proposed for selection of the ideal parasite tag (see MacKenzie and Abaunza, 1998 for review), but it is suggested that the most important criterion for an effective parasite marker is its residence time in the fish (Lester 1990).

There are limitations to most techniques currently used to determine geographic structure of populations. Genetic techniques are not ideal because they will not detect differences of low levels of larval or adult mixing between populations (Hartl and Clark 1989). They also cannot directly measure rates of individual exchange between sites or enable specification of the origin of individuals. While otolith chemistry proposes an alternative technique for evaluating population structure, it is reliant on differences in water chemistry, so it can be problematic to differentiate populations within a homogeneous body of water. Patterns of geographic structure inferred from genetics and otolith chemistry should be validated by independent replication and comparisons against other sources of information such as biological and physical tagging (Thresher 1999).

Parasites offer an alternative tool to artificial tags that may not be used easily for delicate fishes. Hemiramphidae (garfishes) fulfil this criterion, being small, delicate fishes that comprise important commercial and recreational fisheries throughout the world. In Australia, there are 17 species represented in the family, which support fisheries in all Australian states (Collette 1974). The southern garfish *Hyporhamphus melanochir*, endemic to temperate southern Australian waters (Gomon *et al.* 2008) are a schooling fish that generally

concentrate in sheltered bays, shallow inshore waters and estuaries. The species is a priority scalefish species taken in the coastal waters of South Australia worth an estimated AUD$3 million per year (Knight *et al.* 2007). Recent assessment of the South Australian garfish fishery suggests that it is currently overexploited, with considerable age truncation and declining catches (Fowler *et al.* 2008; Steer *et al.* 2009).

A computer-based fishery model is used to assess the commercial catch of *H. melanochir* in South Australia (McGarvey *et al.* 2007). This model integrates demographic information with catch and effort data and is structured to recognise two separate, independent and self-sustaining stocks, one in Spencer Gulf and the other in Gulf St Vincent (Figure 1). Contrary to the current jurisdictional approach, a genetic investigation indicates population homogeneity between *H. melanochir* sampled in Gulf St. Vincent and Spencer Gulf (Donnellan *et al.* 2001). More recently, otolith chemistry has provided evidence for age-related movements and fine-scale population structuring for the species (Steer *et al.* 2009) suggestive of a metapopulation structure.

The aims of this case study were to discover and document the metazoan parasite fauna of *H. melanochir*, identify appropriate candidate species markers, describe spatial variation in parasite abundance and make inferences about the level of population structuring that has been observed through otolith chemistry techniques.

3.3 Materials and Methods

3.3.1 Fish collection

Fresh specimens of *H. melanochir* were collected from Safcol commercial fish market, Mile End, South Australia, from late November 2008 to April 2009. All *H. melanochir* were captured from commercially important areas and are representative of the geographical extremes of the commercial fishery in South Australian waters. Consequently, spatial resolution was largely restricted to the northern reaches of Gulf St Vincent and Spencer Gulf and on the northern shore of Kangaroo Island (Figure 1). Fishery catches, packed in boxes by the fishers, were selected from the market floor, based on their region of origin. For each catch a subsample was chosen haphazardly and without reference to the size of the fish they contained. All fish were harvested in a similar manner by commercial fishers using haul nets, which are limited by legislation to a depth of 5 m or less. Fish from Kangaroo Island were the

exception, as they were collected by commercial fishers using dip nets at night. *Hyporhamphus melanochir* were sampled from nine sites within three regions of South Australia (Figure 1): Gulf of St Vincent (Port Wakefield; Port Parham), Kangaroo Island (American River; Bay of Shoals) and Spencer Gulf (Whyalla; Port Pirie; Port Broughton; Cowell; and Corny Point). A minimum of 28 fish were examined from each site, which reveals parasites at a prevalence ≥ 10% with 95% confidence (Post and Millest 1991).

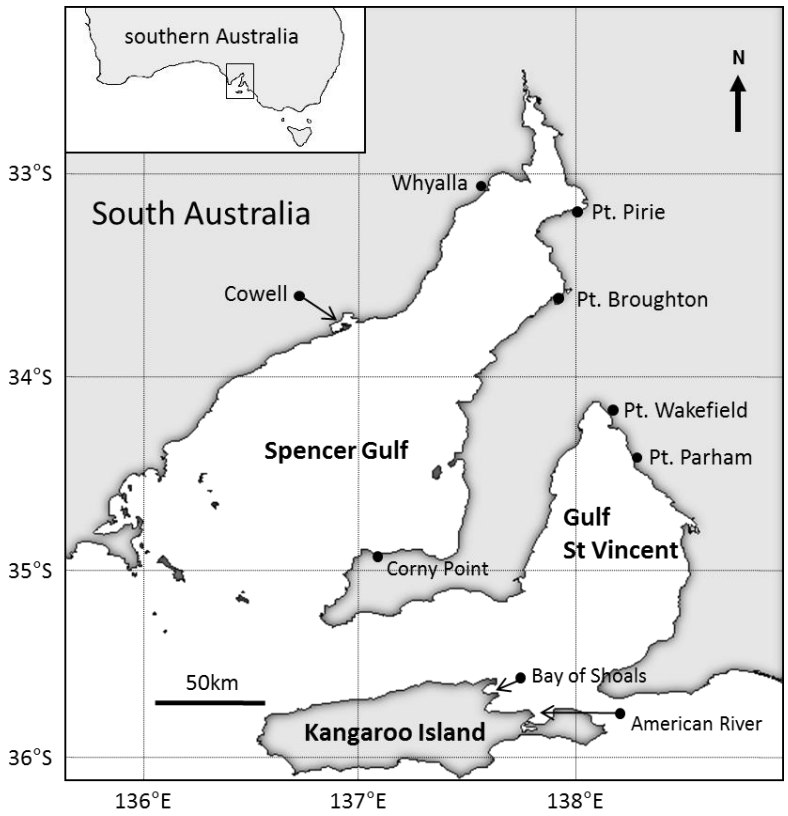

Figure 1. Map indicating *Hyporhamphus melanochir* sampling localities in South Australia including Spencer Gulf (Whyalla, Port Pirie, Port Broughton, Cowell, Corny Point), Gulf St Vincent (Port Wakefield, Port Parham) and Kangaroo Island (American River, Bay of Shoals).

3.3.2 Dissection and parasite preservation

Fish were examined fresh for live parasite recovery or were frozen and examined later. Parasites were collected using standard parasitological techniques. Standard length L_S (upper jaw to the posterior end of the last vertebra) was measured for each fish. Fish sex was recorded and sagittal otoliths were removed for age determination. Parasites were identified to lowest possible taxon following published parasite descriptions, taxonomic keys, voucher material and additional parasitological expertise (see Acknowledgements). Representative parasite specimens, mounted on slides and/or in alcohol for future DNA analysis, have been

deposited in national and State museums. Parasite prevalence and intensity are given in whole numbers and definitions used throughout follow Bush *et al.* (1997).

The age of each fish was determined from sagittal otolith examination. One otolith from each fish was embedded in resin, transversely sectioned using a diamond saw and mounted on a glass slide with superglue. The number of opaque zones, the characteristics of the edge of the otolith, the sample date and the universal birth date of 01 January which falls in the middle of the spawning season (Ye *et al.* 2002) were recorded and used to estimate fish age. Otoliths were examined and interpreted against the *H. melanochir* otolith reference collection held at South Australian Research and Development Institute, Aquatic Sciences.

3.3.3 Statistical Analyses

Summary statistics, including parasite prevalence, intensity and abundance were compiled for each species using the raw parasite data. Parasites were identified as potential biological markers if they exhibited high prevalence (> 10% in any one site), were relatively easy to find, identify and count. They were categorised as 'temporary' or 'permanent' depending on their probable life span and mechanism of attachment on or in *H. melanochir.* To reduce the variance and aid with normalising the distribution, the natural logarithm of the parasite abundance (x) plus one [ln (x + 1)] was applied to the data for all higher statistical analyses. Parasites with a prevalence ≤ 10% in any one site were not considered a component species (see Bush *et al.* 1990) and were excluded.

Spearman rank correlations were used to explore potential relationships between the size and age of *H. melanochir* with parasite abundance and diversity. Furthermore, a Mann–Whitney *U* Test was used to determine if parasite abundance and diversity varied as a function of sex. Single factor analyses of variance were performed separately for each parasite species to examine differences in abundance between sites. *Post-hoc* Hochberg GT2 tests were also carried out to identify significant differences among means. Stepwise discriminant function analysis (DFAs) was used to assess whether the site of capture of each *H. melanochir* could be reliably determined from entire parasite assemblage abundance. A second DFA was carried out to explore whether the reclassification rates improved if only the 'permanent' parasite species were included in the analysis. The entry of predictors to the analysis was determined by the statistical entry of Wilk's lambda (λ) with a *P* (entry) = 0.05 and *P* (exit) = 0.25.

3.4 Results

Two hundred and seventy-four *H. melanochir* (mean ± S.D. = 233 ± 28mm L_S) were examined for metazoan parasites from South Australian waters (Table 32). Females dominated the sex ratio ~6:1. *Hyporhamphus melanochir* size significantly differed between sites (Kruskal–Wallis, $\chi^2 = 143.90$, df = 8, $P < 0.001$), with the largest fish (>280 mm L_S) collected from Cowell, Port Wakefield and the Bay of Shoals (Figure 2). A greater proportion of *H. melanochir* collected from these sites were from the 2+ year age cohort ($\chi^2 = 62.86$, df = 8, $P < 0.001$), whereas 1+year *H. melanochir* tended to dominate the remaining samples (Figure 3).

Table 32. Number of *Hyporhamphus melanochir* (*n*) collected from each site in South Australia with corresponding mean standard length (L_S) and mean age, with respective ranges in parentheses. Predominant habitats including reef and seagrass (RS), seagrass meadow (SM), tidal flat (TL) and unvegetated soft bottom (USB) are indicated, following Bryars (2003).

Region	Site	Habitat	*n*	Mean L_S (mm)	Mean age (yrs)
Spencer Gulf	Corny Point	SM, USB	32	229 (195–277)	1.17 (0–3)
	Cowell	SM	30	261 (199–357)	1.93 (0–3)
	Port Broughton	SM, TF, USB	32	225 (185–287)	1.31 (1–3)
	Port Pirie	SM, TF USB	28	232 (220–257)	1.36 (1–2)
	Whyalla	SM, USB	31	229 (194–262)	1.10 (0–2)
Gulf St Vincent	Port Parham	SM, TF, USB	31	211 (182–251)	0.87 (0–2)
	Port Wakefield	SM, TF, USB	30	218 (188–305)	1.28 (0–3)
Kangaroo Island	American River	RS, USB	32	262 (242–281)	0.97 (0–2)
	Bay of Shoals	SM, USB	28	265 (231–314)	1.75 (1–4)

Fourteen types of parasites were distinguished from *H. melanochir* and four identified to species (Table 33). *Bomolochus bellones* Burmeister 1833 differs to redescriptions of *B. bellones* provided by Kabata (1979) and Ho *et al.* (1983) from: 1) an additional small spine on leg one, 2) a patch of spinules on either side of the ventral midline of the anal somite and 3) a patch of spinules on the ventral surface of the caudal ramus. *Micracanthorhynchina hemirhamphi* (Baylis 1944) Ward 1951 exhibited minor differences in measurements and spine counts from the original description, but largely agreed with *M. hemirhamphi*.

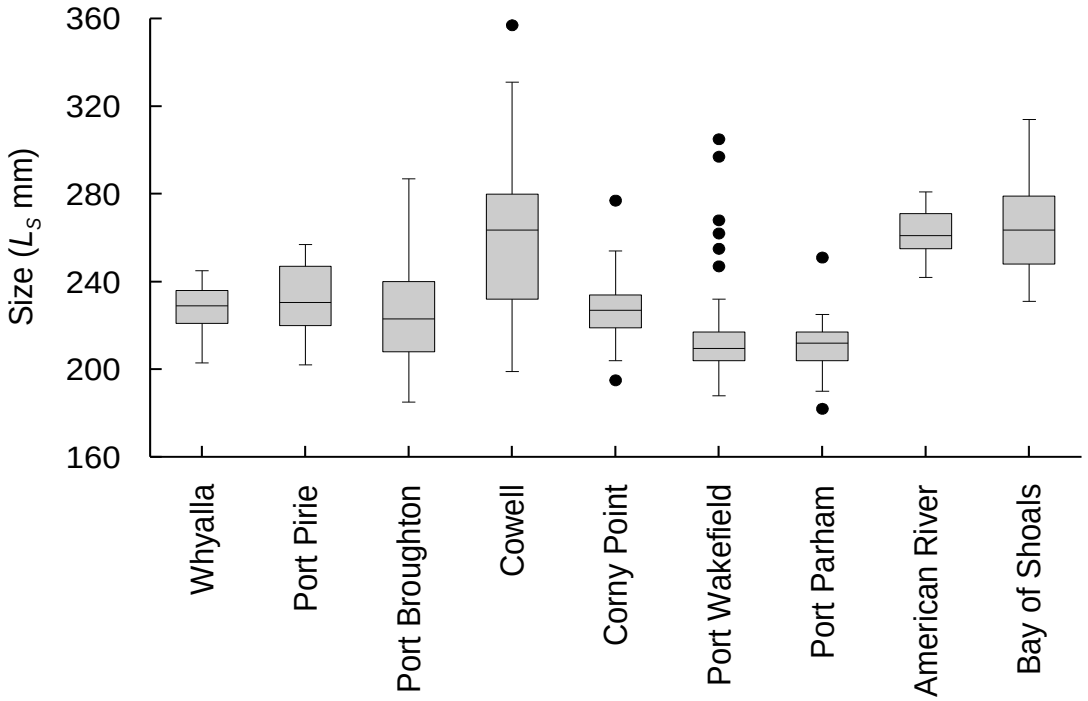

Figure 2. Box and whisker plot for standard length L_S frequencies of *Hyporhamphus melanochir* sampled from nine sites within three regions: Spencer Gulf (Whyalla, Port Pirie, Port Broughton, Cowell, Corny Point), Gulf St Vincent (Port Wakefield, Port Parham) and Kangaroo Island (American River, Bay of Shoals). Whiskers (or caps) indicate extreme values (minimum and maximum) while outliers are shown as a filled circle, placed at the outlying points (an outlier is any point that falls below $Q_L - 1.5*I_{RQ}$ or above $Q_U + 1.5*I_{RQ}$; where I_{RQ} is the difference between the quartiles, Q_L is the value of the lower quartile (bottom of the box) and Q_U is the value of the upper quartile (top of the box)).

Seven parasite species were identified as possible biological markers (Tables 33 and 34). Four species, *Lernaeenicus hemirhamphi, Micracanthorhynchina hemirhamphi, Mothocya halei* and *Philometra* sp. are proposed to exhibit longer residence time in or on the host, based on information available in published literature and their mechanisms of attachment (see Discussion).

3.4.1 Variation in parasite abundance

Parasite prevalence and intensity differed between the nine sites sampled and not all parasite species were found at all sites (Table 34). In some cases parasites were observed only in fish collected from a few sites (e.g. bivesiculid type, opecoelid type, tetraphyllidean metacestodes), whereas *B. bellones* and *Conohelmins* sp. were found at all sites (Table 34).

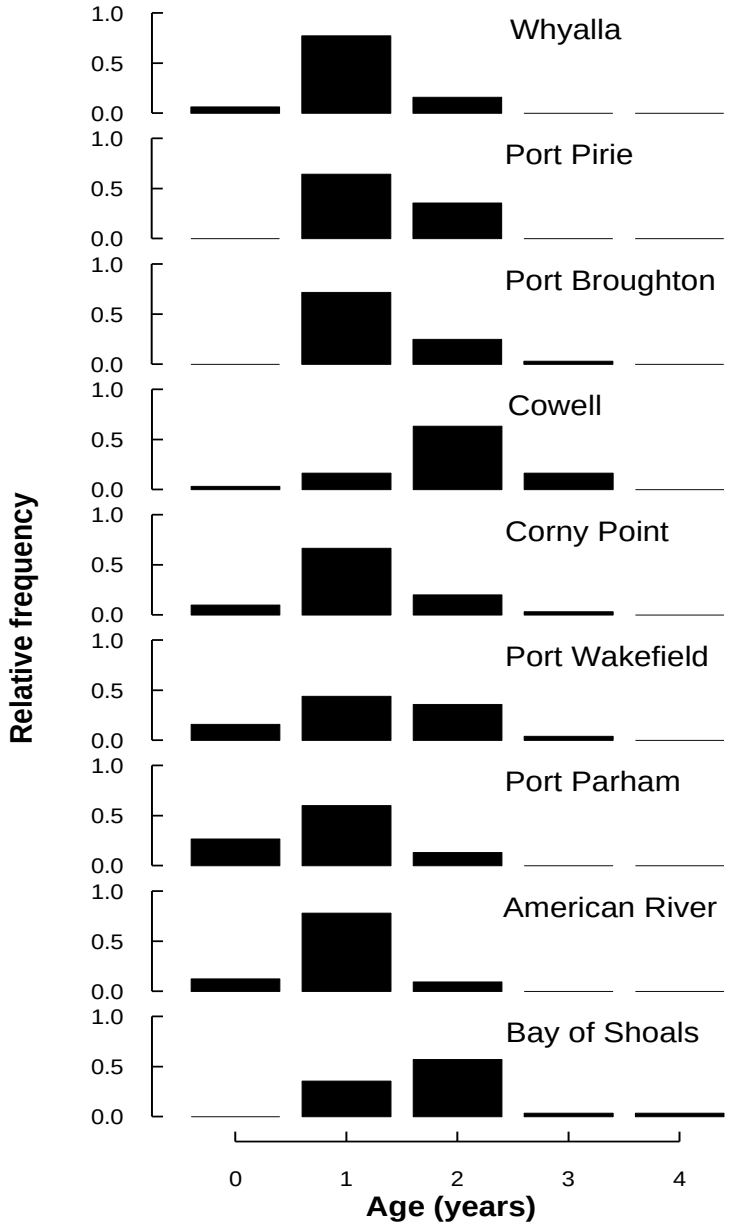

Figure 3. Age-frequency distribution for *Hyporhamphus melanochir* sampled from nine sites within three regions: Spencer Gulf (Whyalla, Port Pirie, Port Broughton, Cowell, Corny Point), Gulf St Vincent (Port Wakefield, Port Parham) and Kangaroo Island (American River, Bay of Shoals).

Abundance of parasites was significantly different between sites sampled (Table 35). Seven parasite species displayed significant spatial variability (Table 35).

There was a positive correlation between *H. melanochir* size and parasite abundance (correlation coefficient = 0.171; $P < 0.01$) and individual parasite species abundance varied with *H. melanochir* size (Figure 4). Permanent parasites were evenly distributed across all sized fish (e.g. *M. halei* and *L. hemirhamphi*), more abundant on fish > 240 mm (e.g. *Micracanthorhynchina hemirhamphi*) or not detected on fish > 250 mm (e.g. *Philometra* sp.)

Table 33. Parasite species from *Hyporhamphus melanochir* indicating microhabitat, museum accession number and use as a biological marker

Higher taxon	Taxon	Microhabitat	Accession no.	Biological marker
Cestoda	Tetraphyllidean type 1	Intestinal tract	AHC 29819	No
	Tetraphyllidean type 2	Intestinal tract	AHC 29820	No
	Tetraphyllidean type 3	Intestinal tract	AHC 29821	No
Trematoda	Acanthocolpid metacercaria*	Gills	AHC 29815-16	No
	Bivesiculid type*	Intestinal tract	AHC 29814	No
	Conohelmins sp.*	Intestinal tract	AHC 29812-13	Temporary
	Opecoelid type*	Intestinal tract	AHC 29817-18	No
Monogenea	*Axine* sp.*	Gills	AHC 29810-11	No
Nematoda	*Hysterothylacium* sp.*	Intestinal tract	AHC 45439	Temporary
	Philometra sp.*	Body cavity	AHC 45440	Permanent
Acanthocephala	*Micracanthorhynchina hemirhamphi*	Intestinal tract	AHC 45438	Permanent
Copepoda	*Bomolochus bellones*	Gills	C6843	Temporary
	Lernaeenicus hemiramphi	Flesh	C6844	Permanent
Isopoda	*Mothocya halei*	Branchial cavity, mouth	C6842	Permanent

*Indicates new host record. Museum collection acronyms and address: Australian Helminth Collection (AHC) and Marine Invertebrate Collection (C), South Australian Museum, North Terrace, Adelaide, South Australia 5000, Australia.

(Figure 6). There was no detectable correlation between *H. melanochir* sex and parasite abundance (Mann–Whitney U = 3,942; $P > 0.05$) or parasite diversity (Mann–Whitney U = 2,083; P = > 0.05). Discriminant function analysis of the entire parasite assemblage data detected significant variations in infection between sites (Wilk's λ =0.09, χ^2 = 644.5, df. = 64, $P < 0.001$). Two discriminant functions explained 84.5% of the between-group variability, with 49.6% of the parasite infection successfully being classified to the site of origin. Separation along the first discriminant function was mainly driven by differences in the infection of the acanthocephalan *M. hemirhamphi*. The second discriminant function was mainly driven by the infection of the trematode *Conohelmins* sp. and nematodes *Hysterothylacium* sp. and *Philometra* sp. [Figure 5(A)]. The analysis gave a classification success range from 20% for Port Wakefield to 97% for American River (Table 36).

The level of classification of individuals to the site from which they originated was 97%, 82% and 54% for American River, Port Broughton and Port Pirie, respectively (Table 36). In some cases *H. melanochir* were incorrectly classified as originating from a site in close proximity (e.g. 25% of *H. melanochir* collected from Port Pirie were statistically assigned to Port Broughton, which is < 40 km south) (Figure 1; Table 36). Some individuals were classified as being broadly distributed amongst both gulfs. For example, 57% of the fish collected from Port Wakefield in northern Gulf St Vincent were incorrectly assigned to sites within northern Spencer Gulf (Table 36).

Table 34. Prevalence and mean intensity of metazoan parasite infections of *Hyporhamphus melanochir* from nine sites within three regions of South Australia. Prevalence (%), expressed in whole numbers, followed by mean parasite intensity and range in parentheses.

Region	Spencer Gulf					Gulf St Vincent		Kangaroo Island	
Site	Corny Pt	Cowell	Pt Broughton	Pt Pirie	Whyalla	Pt Parham	Pt Wakefield	American River	Bay Shoals
No. hosts examined	n=32	n=30	n=32	n=28	n=31	n=31	n=30	n=32	n=28
Parasite species									
Tetraphyllidean metacestodes*	0	0	0	0	0	0	0	3; 4 (4)	0
Acanthocolpid metacercaria*	6; 2 (1–2)	0	0	11; 2(1–3)	0	3; 14 (14)	10; 2 (1–3)	0	0
Bivesiculid type	0	0	0	4; 1(1)	0	0	7; 8 (1–14)	0	0
Conohelmins sp.	9; 2 (1–3)	27; 8 (1–23)	6; 5 (2–8)	46; 3 (1–6)	77; 13 (1–138)	71; 13 (2–33)	30; 26 (1–152)	9; 6 (2–12)	50; 9 (1–19)
Bomolochus bellones	19; 1 (1)	3; 1 (1)	21; 1 (1–4)	25; 2 (1–3)	45; 3 (1–7)	39; 2 (1–12)	40; 2 (1–3)	13; 1 (1)	54; 2 (1–2)
Opecoelid type	6; 3 (3)	0	0	0	0	0	6; 1 (1)	0	0
Axine sp.	13; 1 (1–2)	0	22; 1 (1–3)	14; 2 (1-3)	19; 2 (1–4)	10; 6 (1–17)	33; 4 (1–22)	38; 3 (1–6)	36; 4 (1–10)
Hysterothylacium sp.*	3; 1 (1)	0	0	39; 9 (1–46)	0	7; 1 (1)	3; 1 (1)	38; 1 (1–3)	0
Philometra sp.	0	3; 3 (3)	3; 1 (1)	43; 4 (1–16)	0	13; 1 (1–3)	10 ; 2 (1–4)	0	0
Micracanthorhynchina hemirhamphi	47; 7 (1–47)	73; 4 (1–10)	3; 1 (1)	7; 2 (1–2)	3; 1 (1)	3; 1 (1)	0	100; 14 (1–86)	0
Lernaeenicus hemirhamphi	0	7; 1 (1)	0	7; 1 (1)	19; 2 (1–6)	10; 1 (1–2)	13; 1 (1)	6; 1 (1)	14; 2 (1–4)
Mothocya halei	9; 2 (2)	37; 2 (1–2)	9; 1 (1)	11; 2 (2)	45; 2 (1–2)	32; 2 (1–2)	23; 1 (1–2)	0	61; 2 (1–3)

*Larval stages. Tetraphyllidean metacestodes includes types 1, 2 and 3.

Table 35. Statistical analysis of the abundance of parasite species of *Hyporhamphus melanochir* amongst sample sites in South Australia. One-way ANOVA (column one) identifies which parasite species differ significantly in abundance ($P < 0.01$) across sites sampled. Hochberg GT2 *post-hoc* values indicate those species which differ significantly from each other amongst sites (denoted by lowercase lettering combinations).

Region	Spencer Gulf					Gulf St Vincent		Kangaroo Island	
Site	Whyalla	Pt Pirie	Pt Broughton	Cowell	Corny Pt	Pt Wakefield	Pt Parham	American River	Bay of Shoals
Bomolochus bellones ($P < 0.001$)	1.226b	0.500b	0.313b	0.030a	0.156a	0.600b	0.871b	0.125a	0.929b
Acanthocolpid metacercaria ($P = 0.143$)	<0.001a	0.179a	<0.001a	<0.001a	0.094a	0.300a	0.452a	<0.001a	<0.001a
Conohelmins sp. ($P < 0.001$)	10.387c	1.393ab	0.313a	2.533ab	0.156a	7.700ab	9.032c	0.531a	4.393bc
Axine sp. ($P = 0.001$)	0.323ab	0.250ab	0.313ab	<0.001a	0.188ab	1.467b	0.613ab	0.969b	1.429b
Hysterothylacium sp. ($P <0.001$)	<0.001a	3.393b	<0.001a	<0.001a	0.031a	0.330a	0.650a	0.500a	<0.001a
Philometra sp. ($P < 0.001$)	<0.001a	1.857b	0.031a	0.100a	<0.001a	0.200a	0.258a	<0.001a	<0.001a
Micracanthorhynchina hemirhamphi ($P < 0.001$)	0.032a	0.107a	0.031a	2.700b	3.094b	<0.001a	0.032a	13.344c	<0.001a
Lernaeenicus hemirhamphi ($P = 0.031$)	0.452b	0.071ab	<0.001a	0.067ab	<0.001a	0.133ab	0.129ab	0.063ab	0.250ab
Mothocya halei ($P < 0.001$)	0.774cd	0.214ab	0.094ab	0.533bcd	0.188ab	0.333abc	0.484abc	<0.001a	1.143d

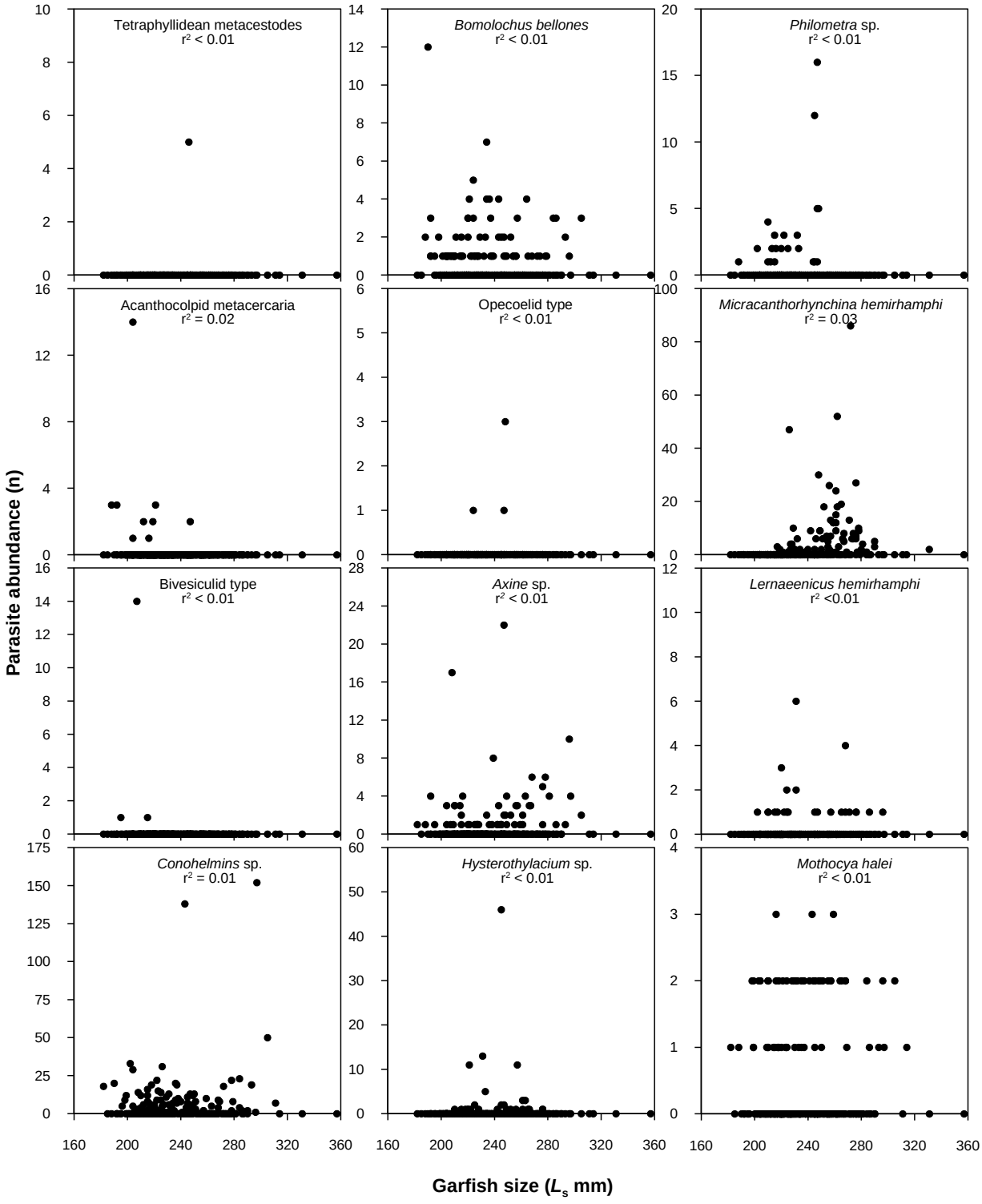

Figure 4. Scatterplots for individual abundance of parasite species against *Hyporhamphus melanochir* standard length (L_s).

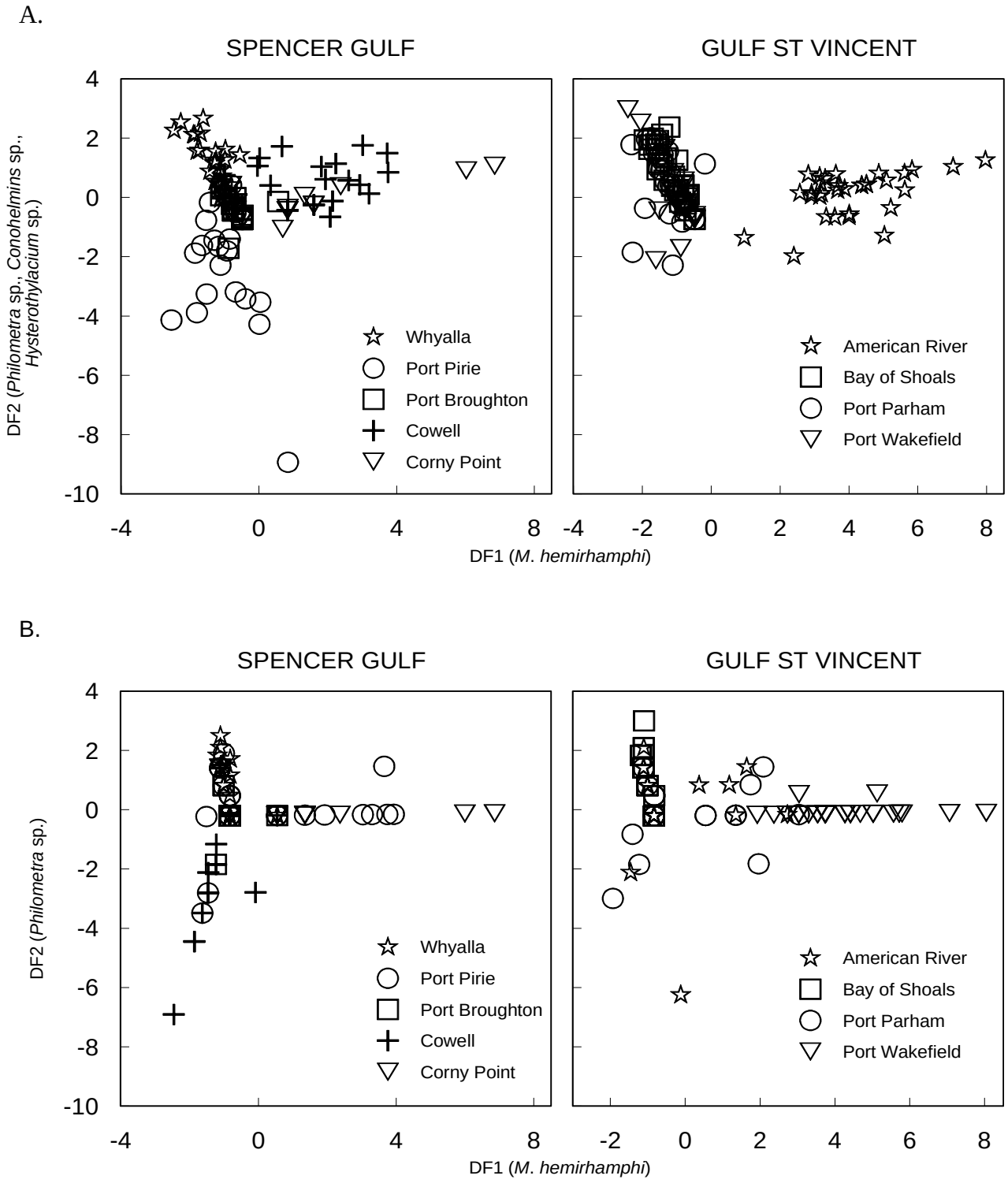

Figure 5. Results of discriminant function (DF) analyses (DF 1 v. DF2) of the entire parasite assemblage of *Hyporhamphus melanochir* (A) and permanent parasites only (B), from two regions Spencer Gulf and Gulf St Vincent (northern Kangaroo Island sites are combined with Gulf St Vincent).

Table 36. Reclassification table for the entire parasite assemblage of *Hyporhamphus melanochir* sampled from nine sites within three regions: Spencer Gulf (Whyalla, Port Pirie, Port Broughton, Cowell, Corny Point), Gulf St Vincent (Port Wakefield, Port Parham) and Kangaroo Island (American River, Bay of Shoals). Bold values indicate successful reclassification to the site of origin.

	Whyalla	Port Pirie	Port Broughton	Cowell	Corny Point	Port Wakefield	Port Parham	American River	Bay of Shoals
Whyalla	**35**	0	10	0	0	10	19	0	26
Port Pirie	4	**54**	25	0	0	0	10	0	7
Port Broughton	0	0	**82**	0	3	0	0	0	9
Cowell	0	0	17	**27**	23	7	6	17	3
Corny Point	0	0	41	3	**38**	3	0	6	9
Port Wakefield	10	7	40	0	0	**20**	10	0	13
Port Parham	13	6	19	0	0	3	**53**	0	6
American River	0	0	0	0	3	0	0	**97**	0
Bay of Shoals	14	0	14	0	0	14	18	0	**40**

Discriminant function analysis of assemblage data for permanent parasite species (e.g. *L. hemirhamphi*, *M. hemirhamphi*, *Mothocya halei* and *Philometra* sp.) detected significant variations in infection between sites (Wilk's $\lambda = 0.172$, $\chi^2 = 469.25$, df. =24, $P < 0.001$). Two discriminant functions explained 95.7% of the between-group variability, with 38.3% of the parasite infection successfully classified to the site of origin. Separation along the first discriminant function was mainly driven by differences in the infection of *Micracanthorhynchina hemirhamphi*, while the second discriminant function was mainly driven by infection of *Philometra* sp. (Figure 5(B)). The analysis gave a classification success range from 0% for Port Wakefield and Port Parham to 91% for American River (Table 37). In some cases *H. melanochir* from northern Spencer Gulf were incorrectly assigned as originating from a site in close proximity (e.g. 42% and 46% of *H. melanochir* collected from Whyalla and Port Pirie, respectively, were statistically assigned to Port Broughton) (Table 37).

Table 37. Reclassification table for permanent parasites of *Hyporhamphus melanochir* sampled from nine sites within three regions: Spencer Gulf (Whyalla, Port Pirie, Port Broughton, Cowell, Corny Point), Gulf St Vincent (Port Wakefield, Port Parham) and Kangaroo Island (American River, Bay of Shoals). Bold values indicate successful reclassification to the site of origin.

	Whyalla	Port Pirie	Port Broughton	Cowell	Corny Point	Port Wakefield	Port Parham	American River	Bay of Shoals
Whyalla	**19**	0	42	0	3	0	0	0	36
Port Pirie	0	**43**	46	0	0	0	0	0	11
Port Broughton	0	3	**84**	0	3	0	0	0	9
Cowell	7	0	7	**37**	17	0	0	20	13
Corny Point	0	0	44	22	**19**	0	0	6	9
Port Wakefield	13	10	60	0	0	**0**	0	0	17
Port Parham	7	10	55	0	3	0	**0**	0	26
American River	0	0	0	6	3	0	0	**91**	0
Bay of Shoals	14	0	36	0	0	0	0	0	**50**

3.5 Discussion

Multivariate discriminant function analyses of the entire parasite assemblage showed distinct groupings of *H. melanochir* according to their site of capture. Separation was mainly driven by differences in endoparasite infection, namely two nematode species (*Philometra* sp. and *Hysterothylacium* sp.), a trematode (*Conohelmins* sp.) and an acanthocephalan (*M. hemirhamphi*). *Micracanthorhynchina hemirhamphi* and *Philometra* sp. were suspected to be 'permanent' parasites and may be the most useful biological tags for *H. melanochir*.

Variation in endoparasite fauna may be a consequence of different biotic and abiotic features (e.g. temperature, salinity, currents) of the immediate environment and

corresponding availability of prey items infected with intermediate parasite stages. Most sites sampled appeared to exhibit similar habitats consisting of extensive seagrass meadows and patches of unvegetated soft bottoms (Table 32). American River was the notable exception, where the habitat is characterised as being predominantly reef with interspersed seagrass meadows (Bryars 2003). Shifts in feeding behaviour also strongly influence dietary constituents and consequent transmission of parasites through ingestion. Seasonal trophic shifts have been observed in the cape halfbeak off South Africa *Hyporhamphus capensis* (Thominot) which is an active herbivore during warmer months, shifting to invertebrates during winter months (Coetzee 1981) while *H. melanochir* exhibits diurnal trophic shifts, predominantly consuming seagrass during the day and switching to emergent benthic invertebrates at night (Robertson and Klump 1983; Earl et al. 2011). Seasonal variation in feeding behaviour demonstrates the importance of using parasite markers that persist in their host for prolonged periods as parasites that survive for less than a year are of little value for stock discrimination (MacKenzie and Abaunza 1998).

Acanthocephalans may permanently attach to the gut of the host (Pichelin 1997) and have been used previously to indicate host distribution over short distances (Cribb *et al.* 2000). Conversely, Zischke *et al.* (2009) predicted that the acanthocephalan *Neoechinorhynchus topseyi* Podder, 1937 only infected blue threadfin, *Eleutheronema tetradactylum* (Shaw, 1804) for relatively short periods. Live *M. hemirhamphi* were embedded in the gut lining of *H. melanochir* and were considered a 'permanent' parasite for this study (Table 33). Pennelids such as *L. hemirhamphi,* which anchor deep within fish muscle tissue, probably remain in their host until death, although there is no information available on longevity in wild fish hosts. Female cymothoids are also likely to be permanently attached to fish and some species may survive for more than one year on their host (Adlard and Lester 1994). Female *Philometra* sp. living in the body cavity were large and conspicuous, and cannot be easily lost compared to ectoparasites or gastrointestinal parasites such as *Axine* sp., *B. bellones*, *Conohelmins* sp. and *Hysterothylacium* sp.. Acanthocolpid metacercariae were labour intensive to remove from cysts, flatten, mount and identify and therefore are not considered useful as biological tags.

Accurate identification and knowledge of the biological attributes of a parasite species is fundamental to determine their application for host population differentiation. Although considerable care was taken in this study, identification can be severely limited by specimen quality and extensive taxonomic confusion in the literature. Selecting permanent parasites can be difficult because residence time in wild fish is generally unrecorded. In addition, reliance on commercial catch may bias observations. For example, *H. melanochir* collected in this study were confined to commercial catches and therefore the size of the fish was constrained by the legislated legal minimum length of 230 mm (L_T). Furthermore, the age structure of the commercial *H. melanochir* fishery is truncated to consist predominantly of 1 and 2 year-old fish (Fowler *et al.* 2008). These constraints precluded the reliable collection across a wide size and age spectrum. Fish sampled from Kangaroo Island were generally larger, which is likely to be a consequence of the commercial fishers selectively targeting larger fish with dip nets. If useful conclusions for management are to be drawn about population structuring in *H. melanochir*, then sampling must be more intensive.

A shallow inshore species with little tendency to migrate large geographical distances, such as *H. melanochir*, may exist as several discrete geographical populations in South Australian waters. Steer *et al.* (in press) provide evidence from otolith chemistry of a distinct *H. melanochir* population in northern Spencer Gulf that exhibits little inter-regional mixing. This hypothesis is further supported by the incorrect reassignment of fish sampled in northern Spencer Gulf to other localities within northern Spencer Gulf compared to southern Spencer Gulf (Table 37). In addition, the abundance of permanent *Micracanthorhynchina hemirhamphi*, did not differ significantly among locations sampled in northern Spencer Gulf, yet differed significantly between northern and southern Spencer Gulf localities (Table 37).

Parasite abundance in *H. melanochir* samples collected in close proximity off the north coast of Kangaroo Island varied considerably. For example, *H. melanochir* sampled from American River were infected by the permanent acanthocephalan *M. hemirhamphi*, whereas these parasites were not detected in fish from the adjacent (< 25 km) Bay of Shoals (Table 34). *Hyporhamphus melanochir* from the Bay of Shoals were infected by permanent isopod *Mothocya halei*, which was not detected in *H. melanochir* collected from American River (Table 34). Furthermore, *M. halei* was found to infect

fish across the size range sampled (Figure 4) and was recovered in all other locations (Table 34). These results imply that *H. melanochir* have limited local movement and that transmission of parasites may be concentrated locally. Certainly, parasites can indicate limited distributions of their hosts (e.g. Grutter 1998; Cribb *et al.* 2000; Vignon *et al.* 2008).

Some *H. melanochir* were classified as being broadly distributed amongst both gulfs, particularly those sampled from sites in northern Gulf St Vincent and northern Spencer Gulf (Figure 1). The incorrect assignment for these individuals is not clear, but suggests that sites that are separated by considerable distances may have comparable environments, resulting in similar parasite species abundance (Table 32 and 33). This discrepancy was also identified in recent otolith trace element studies and was attributed to environmental similarities rather than true migration pathways (Steer *et al.* 2009; Steer *et al.* in press).

The findings of this study provide further, supporting, evidence for a metapopulation theory where South Australian *H. melanochir* consists of a series of spatially separated sub-populations with limited levels of intermixing (Steer *et al.* 2009; Steer *et al.* in press). Species that are broadly distributed across large environmental gradients are generally partitioned into smaller component populations with different biological attributes (Secor 1999). These populations can be relatively discrete and self-replenishing but maintain sufficient exchange to preserve genetic homogeneity, as has been observed for *H. melanochir* by Donnellan *et al.* (2001).

High exploitation, combined with ineffective fisheries management, can result in depletion of fish stocks. Neglecting to account for a metapopulation structure, as suggested for *H. melanochir*, increases the risk of localised depletion. Steer *et al.* (2009) suggest that assessment and management of the *H. melanochir* fishery in South Australia may have to be restructured to align with smaller spatial units. Adopting a more holistic approach to population structure studies, which would include the assessment of the species' genetic diversity, meristic qualities, otolith microchemistry and biological markers has become the preferred option when there is a need to determine the relative size of management units (Begg and Waldman 1999). This study has identified four endoparasite species (including two 'permanent' species) that may be

used as biological markers for *H. melanochir*. Equivalent congeners could be useful for population studies of other hemiramphids worldwide.

4 PARASITE REDESCRIPTIONS

4.1 Redescriptions of two species of microcotylid monogeneans from three arripid hosts

This section has been published and may be cited as:
Catalano, S.R., Hutson, K.S., Ratcliff, R. & Whittington, I.D. 2010. Redescriptions of two species of microcotylid monogeneans from three arripid hosts in southern Australian waters. *Systematic Parasitology* **76**, 211-222.

4.1.1 Abstract

Microcotyle arripis Sandars, 1945 is redescribed from *Arripis georgianus* from four localities: Spencer Gulf, Gulf St. Vincent, off Kangaroo Island and Coffin Bay, South Australia. *Kahawaia truttae* (Dillon and Hargis, 1965) Lebedev, 1969 is reported from *A. trutta* off Bermagui, New South Wales and is redescribed from a new host, *A. truttaceus*, from four localities in South Australia: Spencer Gulf, Gulf St. Vincent, off Kangaroo Island and Coffin Bay. Phylogenetic analysis of the partial 28S ribosomal RNA gene (28S rRNA) nucleotide sequences for both microcotylid species and comparison with other available sequence data for microcotylid species across four genera contributes to our understanding of relationships in this monogenean family.

4.1.2 Introduction

All four species of *Arripis* Jenyns (Pisces: Arripidae), *Arripis georgianus* (Valenciennes), *A. trutta* (Forster), *A. truttaceus* (Cuvier) and *A. xylabion* Paulin, are endemic to the waters of southern Australia and New Zealand (NZ), and are caught by commercial and recreational fishers (see Paulin 1993 for a review). During a survey of the parasites of *A. georgianus*, *A. trutta* and *A. truttaceus*, two parasite species from the Microcotylidae Taschenberg, 1879 were recovered. *Microcotyle arripis* Sandars, 1945 is redescribed from *A. georgianus* as the original description by Sandars (1945) and subsequent redescriptions by Dillon *et al.* (1984) and Williams (1991) are based upon limited material. Our study also provides new geographic localities for *M. arripis* in South Australian (SA) waters. *Kahawaia truttae* (Dillon and Hargis, 1965) Lebedev,

1969, originally described as *Gonoplasius truttae* Dillon and Hargis, 1965 from *A. trutta* at Timuru, South Island, NZ (Dillon and Hargis, 1965), is reported from *A. trutta* off the coast of New South Wales (NSW) and from *A. truttaceus* from SA waters, providing new geographic localities for this species in southern Australian waters. This microcotylid is redescribed because the original description by Dillon and Hargis (1965) was based on limited material and the subsequent account by Lebedev (1969) was poorly illustrated. To improve our understanding of relationships within the Microcotylidae, newly sequenced partial 28S ribosomal RNA gene (28S rRNA) nucleotide sequence data for *M. arripis* from *A. georgianus* and *K. truttae* from *A. trutta* and *A. truttaceus* are compared with sequence data for other microcotylid species available in GenBank™.

4.1.3 Materials and Methods

4.1.3.1 Collection of hosts and parasites

Monogeneans were collected from the gills of *Arripis georgianus* and *A. truttaceus* at four localities in SA waters (Spencer Gulf, Gulf St. Vincent, off Kangaroo Island and Coffin Bay; see Results for geographical coordinates), from *A. trutta* from one location off NSW (Bermagui; see Results for geographical coordinates) and from *Sillaginodes punctatus* (Cuvier) in SA waters (specific locality unknown). Most fish were examined fresh; others were frozen immediately upon collection and processed later. Gills were removed, placed in individual Petri dishes of seawater and studied for parasites. Monogeneans were removed and preserved in either 10% formalin or 95% undenatured ethanol. Representative specimens were slivered along the body margin with the slivered section preserved in 95% ethanol and the remaining section preserved in formalin. Parasites preserved in formalin were washed twice in MiliQ water, stained in Mayer's haematoxylin, destained in 3% HCl, dehydrated in an ethanol series (70%, 90%, 95% and 100%), cleared in cedar wood oil and mounted on a slide beneath a coverslip in Canada balsam.

Tissue samples were collected from each fish, stored in undenatured ethanol and lodged with the Australian Biological Tissue Collection (ABTC) at the South Australian Museum Australia (SAMA), North Terrace, Adelaide, South Australia 5000, Australia (ABTC 108509–777).

4.1.3.2 Morphological methods

Drawings of preserved, mounted parasites were made with the aid of a drawing tube and measurements were made using a computerised digitising system similar to that described by Roff and Hopcroft (1986). The terminology of Williams (1991) for microcotylid monogeneans is adopted, using buccal organs in preference to buccal suckers, and ovary in preference to germarium. Unless stated otherwise, all measurements are presented in micrometres as the mean with the range in parentheses, followed in square brackets by the number of measurements made. A dash (-) indicates that measurements could not be made or were not available.

Measurements of lengths refer to the distance along the anteroposterior axis, except where noted otherwise. Length measurements of structures not orientated along the anteroposterior axis in some specimens, such as buccal organs, clamps, eggs and genital atrium armature, were measured along the longest axis of these structures. 'Middle clamps' are defined as being located halfway along the total haptor length and 'posterior clamps' are defined as being located at the posterior end of the haptor. Occasionally bodies and haptors were curved, so multiple linear measurements were made and summed to give total length. Body width was measured across the ovary and haptor width was measured across the anterior margin.

4.1.3.3 Molecular methods

Monogenean parasites for molecular analyses were preserved in 95% undenatured ethanol and collected from the gills of four fish species, *A. georgianus*, *A. trutta*, *A. truttaceus* and *S. punctatus* (Table 38). Additional monogenean sequences were obtained from GenBank™ (Table 38).

Total genomic DNA (gDNA) was extracted using the Gentra Kit (Gentra Systems) to digest tissue and release DNA, followed by the RNeasy Mini Kit (animal tissue protocol incorporating QIAshredder pretreatment, QIAGEN GmbH, Hilden, Germany) to extract the DNA. PCR amplification of partial 28S rRNA sequence was carried out using two universal primers (Jovelin and Justine, 2001), modified with a 5' M13 tag, C1'M13 (5'GTAAAACGACGGCCAGACCCGCTGAATTTAAGCAT-3') and reverse D2M13 (5'-CAGGAAACAGCTATGACTCCGTGTTTCAAGACGG-3'), corresponding to

positions 25 and 1126 of the complete *Mus musculus* 28S rRNA nucleotide sequence

Table 38. Polyopisthocotylean monogeneans: specimens amplified, sequenced and analysed (- = not applicable).

Family	Monogenean species	Host species	Number of individuals analysed	GenBank™ number	Source
Microcotylidae	*Kahawaia truttae*	*Arripis truttaceus*	52	GU263831	Present study
Microcotylidae	*Kahawaia truttae*	*Arripis trutta*	13	GU263832	Present study
Microcotylidae	*Microcotyle arripis*	*Arripis georgianus*	1	GU263830	Present study
Microcotylidae	*Polylabris sillaginae*[a]	*Sillaginodes punctatus*	10	GU289509	Present study
Microcotylidae	*Microcotyle erythrinii*	*Pagellus erythrinus*	-	AM157221	Badets *et al.* (unpublished)
Microcotylidae	*Microcotyle sebastis*	*Sebastes* sp.	-	AF382051	Olson and Littlewood (2002)
Microcotylidae	Microcotylidae sp. M10	*Sebastes* sp.	-	EF653385	Aiken *et al.* (2007)
Microcotylidae	*Diplostamenides sciaenae*	Not specified	-	FJ432589	Su (unpublished)
Discocotylidae	*Discocotyle sagittata*	*Salmo trutta*	-	AF382036	Olson and Littlewood (2002)

[a]Species identified from Hayward (1996)

(Hassouna *et al.*, 1984). Two additional primers were designed for improved amplification of fish fin clips, SC-FM13 (5'-GTAAAACGACGGCCAGACCTCAGATCAGACGAGACAAC-3') and reverse SC-RM13 (5'-CAGGAAACAGCTATGACCGTGCGTTAGACTCCTTGGTC-3').

Amplification reactions were conducted in a final volume of 25 µl containing: 2.5 µl of GeneAmp 10x PCR Buffer II (Applied Biosystems, Inc. [ABI], Foster City, CA, USA) 1.5 mM $MgCl_2$ (ABI), 200 µM of each dNTP, 10 pmol of each primer, 1.25 U of AmpliTaq Gold (ABI) and 1 µl gDNA extract. After enzyme activation at 94°C for 10 mins, the reactions were subjected to 60 cycles of amplification (94°C for 30 s, 50°C for 1 min and 72°C for 2 min). Negative controls were included to determine any potential contamination of the PCR products. PCR products were visualised with UV following agarose gel (1.5%) electrophoresis and staining with EZ-Vision Three, 6X (Amresco,

Solon, OH, USA). Products of the predicted size were cleaned (QIAquick PCR Purification Kit, QIAGEN) and sequenced in both directions using dye terminator chemistry (BigDye Terminator v3.1 cycle-sequencing kit, ABI) primed with M13 forward and reverse primers.

Sequence alignments, error correction and similarity analysis (neighbor-joining) were performed using Kodon (Applied Maths, St-Martens-Latem, Belgium) and PAUP* 4 (Swofford, 2003), including a bootstrap analysis (1000 replicates). Percentages over 50% were included on the neighbor-joining (NJ) tree. Representative sequences were deposited in GenBank (see Table 1).

4.1.3.4 Specimens requested and deposited

Five voucher specimens of *Kahawaia truttae* (as *Gonoplasius truttae,* W. 14952–56) lodged by Dillon and Hargis (1965) sampled from *A. trutta* in NZ, were borrowed from the Australian Museum (6 College Street, Sydney, NSW, Australia) for comparison with newly collected material. Voucher specimens of taxa reported in this paper are deposited in the Australian Helminthological Collection (AHC) of SAMA, Parasitology Section, North Terrace, Adelaide, South Australia 5000, Australia, the Parasitic Worms Collection of Natural History Museum (NHM), London, England and the United States National Parasitology Collection (USNPC).

4.1.4 Results

4.1.4.1 Microcotyle arripis Sandars, 1945

Class Monogenea Carus, 1863
Family Microcotylidae Taschenberg, 1879
Subfamily Microcotylinae Taschenberg, 1879
Genus *Microcotyle* van Beneden and Hesse, 1863

Type-host: Arripis georgianus (Valenciennes), Arripidae, "Australian herring, tommy ruff" (Sandars 1945; present study; Dillon *et al.* 1984; Williams 1991).

Site: Gills (Sandars 1945; Dillon *et al.* 1984); gill filaments (Williams 1991); primary gill filaments (present study).

Infection details: A total of 51 flukes collected from 50 fish; maximum intensity 4; mean intensity 2 (Sandars 1945); a total of 67 flukes (Dillon *et al.* 1984); a total of 12 flukes collected from 8 fish; prevalence 63%; maximum intensity 4 (Williams 1991); a total of 664 flukes collected from 183 fish; prevalence 72%; maximum intensity 21; mean intensity 5.1 (present study).

Geographic localities: North Beach, Swan River, Mandurah, Bunbury, Busselton, Albany, Woodman's Point, Scarborough and Whitford's Beach, Western Australia (WA), Australia (Sandars 1945); Ardrossan and Adelaide, South Australia (SA), Australia (Dillon *et al.* 1984); Swan River Estuary and Cockburn Sound, WA, Australia (Williams 1991); Spencer Gulf (Pt. Pirie [33.1770 S, 138.0102 E], Pt. Broughton [33.6001 S, 137.9315 E] and Tickera [33.7856 S, 137.7064 E]), Gulf St. Vincent (Edithburgh [35.0858 S, 137.7492 E], Stansbury [34.9135 S, 137.8082 E], Port Parham [34.4454 S, 138.2437 E] and Rapid Bay [35.5255 S, 138.1874 E]), Kangaroo Island (Kingscote [35.6576 S, 137.6439 E] and Emu Bay [35.6037 S, 137.5200 E]) and Coffin Bay [34.6224 S, 135.4628 E], SA, Australia (present study).

Specimens studied: 3 whole-mounts (Dillon *et al.* 1984); 6 whole-mounts (Williams 1991); 2 whole-mounts used for drawing (AHC 29751, AHC 29753), 10 whole-mounts measured, 5 voucher specimens deposited in SAMA (AHC 29751–3, AHC 29883–4), 2 voucher specimens deposited in NHM (NHM 2009.12.28.1–2) and 3 voucher specimens deposited in USNPC (USNPC 102673.00–102675.00) (present study).

Redescription (Figure 6, Table 39)

Body elongate, total length 1887 (867–2713) [10]; width 363 (155–564) [10]; body tapers anteriorly and posteriorly in most specimens (Figure 6A). Haptor weakly delineated, appears continuous with body, 569 (148–939) [10] long; width across anterior margin 161 (94–233) [10]. Haptor armed with total of 76 (44–96) [10] clamps, in two nearly equal ventrolateral rows (Figure 6E). Clamps of typical *Microcotyle*-type, similar in structure and size; middle clamps 22 (9–28) [10] long by 44 (26–54) wide (Figure 6D); posterior clamps 21 (14–43) [10] long by 36 (24–42) [10] wide.

Buccal organs 28 (20–35) [6] long by 28 (15–36) [6] wide; small, sclerotised, tooth-like papillae on rims. Pharynx 35 (29–40) [6] long by 38 (29–46) [6] wide; leading into oesophagus posteriorly. Oesophagus relatively long, 130 (115–144) [3], without diverticula. Genital atrium 38 (21–60) [9] long by 84 (39–113) [9] wide; located 208 (107–306) [9] from anterior end of body. Genital atrium arrangement and armature as in Figure 6B; boundary lined with large spines except posterior edge with spine points facing inwards; inner portion armed with numerous smaller, conical spines, points directed posteriorly; inner spines randomly distributed in some specimens, spaced somewhat evenly in others. Gut bifurcating immediately posterior to genital atrium forming intestinal caeca with medial and lateral branching (branches not shown in Figure 6A but note haematin residues); unclear whether caeca unite posteriorly.

Testes post-ovarian, 19 (15–21) [7] in number; of irregular shape 34 (17–52) [7] long by 55 (41–104) [7] wide. Ovary tubular, tapers posteriorly, difficult to trace course in most specimens. Vitelline ducts anterior, common vitelline duct posterior, complete structure Y-shaped. Vaginal pore not observed. Vitellarium commences at central margin of genital atrium, occupies both lateral fields, extends into haptor. Uterus situated in central field, proceeds to genital atrium. Eggs *in utero* fusiform with filaments at both ends (Figure 6C); eggs 122 (99–145) [2] long by 25 (22–28) [2] wide. Among live parasites collected, none was observed to lay eggs.

Remarks: Measurements for *M. arripis* collected in this study are presented in Table 39 for comparison with previously published measurements (Sandars, 1945; Dillon *et al.*, 1984; Williams, 1991). This species is redescribed here as new specimens show some minor differences to previous descriptions, which were based upon limited material. The present specimens differ from those described by Sandars (1945), Dillon *et al.* (1984) and Williams (1991) as follows: (1) body length shorter than described by Williams (1991), (2) haptor width larger than described by Sandars (1945), (3) oesophagus length smaller than described by Sandars (1945) and (4) middle clamp length, egg length and egg width smaller than all previous descriptions. However, despite these morphometric differences, the anatomy of the present material fits previous accounts of *M. arripis*.

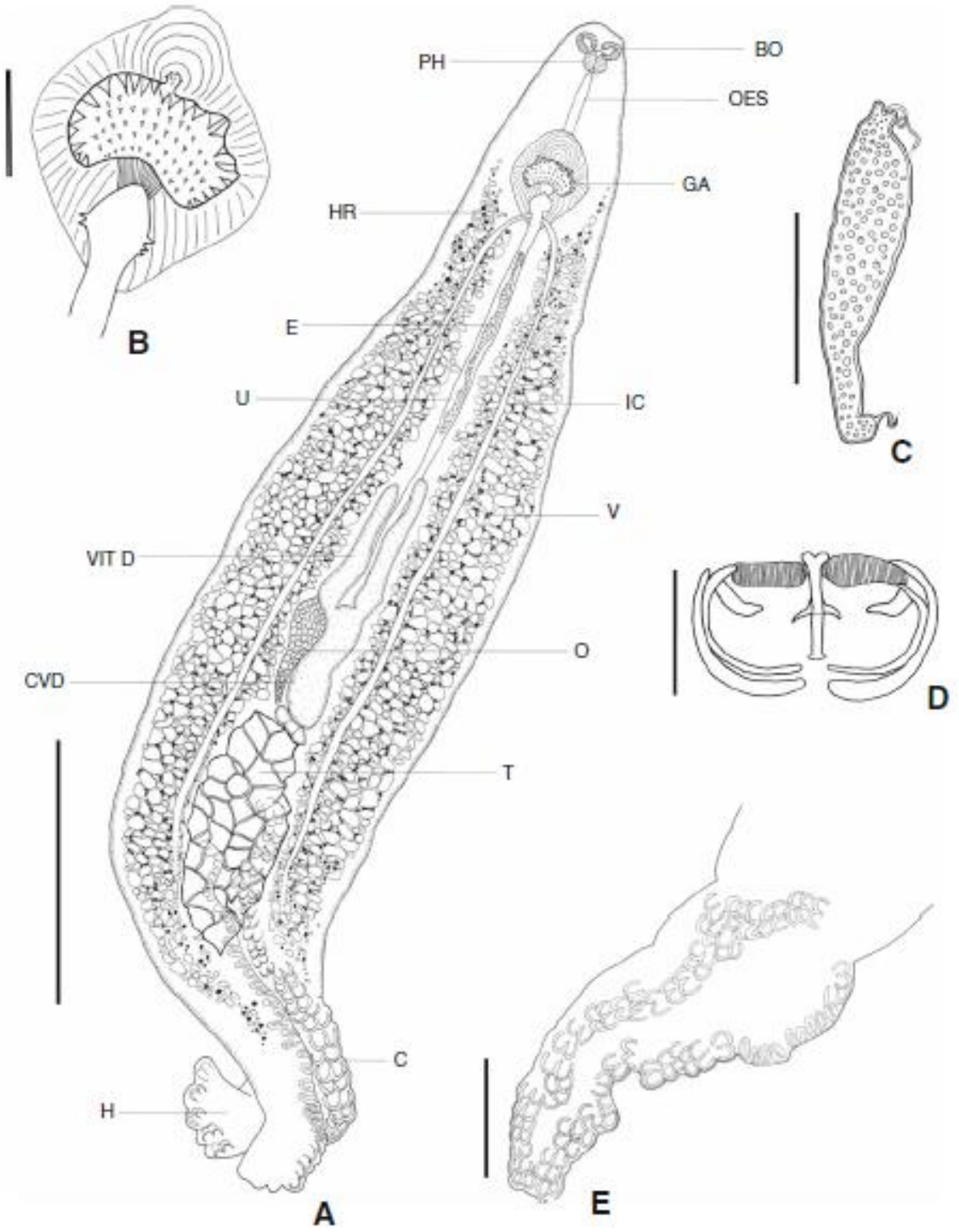

Figure 6. *Microcotyle arripis* Sandars, 1945. A. Whole-mount (ventral view), composite of two individuals (AHC 29751 and AHC 29753); B. Genital atrium drawn from AHC 29751; C. Egg, clearly distorted from normal shape, drawn in utero from AHC 29753; D. Clamp drawn from AHC 29751; E. Enlargement of unflattened, mopre typical symmetrical haptor showing opposing ventrolateral rows of equally-sized clamps dranw from AHC 29884. Abbreviations: BO, Buccal Organ; C, Clamp; CVD, Common Vitelline Duct; E, Egg; GA, Genital Atrium; H, Haptor; HR, Haematin Residues in medial and lateral gut branches (not drawn); IC, Intestinal Caeca; OES, Oesophagus; O, Ovary; PH, Pharynx; T, Testes; U, Uterus; V, Vitellarium; VIT D, Vitelline Duct. Scale bars: A = 0.5 mm, B, C = 50 µm, D = 25 µm, E = 200 µm.

Table 39. Comparative measurements of *Microcotyle arripis* Sandars, 1945 from *Arripis georgianus*, for specimens collected in this study and published data (measurements in micrometres; mean followed by range in parentheses; -, indicates no measurement available).

Characteristic	**Nine regions from Western Australia (WA) (Sandars, 1945)**	**Ardrossan and Adelaide, South Australia (SA) (Dillon *et al.*, 1984)**	**Swan River Estuary and Cockburn Sound, WA (Williams, 1991)**	**Spencer Gulf, Gulf St. Vincent and Kangaroo Island, SA (present study)**
Number of specimens	-	n=3	n=6	N=10
Body length	2080	2047 (1684–2530)	4347	1887 (867–2713)
Body width	720	500 (368–598)	539 (487–568)	363 (155–564)
Haptor length	480	797 (707–874)	1287	569 (148–939)
Haptor width	80	-	-	161 (94–233)
Total number of clamps	70	(68–108)	87 (78–96)	76 (44–96)
Middle clamps length	32	61 (56–70)	61 (56–66)	22 (9–28)
Middle clamps width	48	39 (34–45)	41 (37–46)	44 (26–54)
Posterior clamps length	-	38 (36–41)	-	21 (14–43)
Posterior clamps width	-	28 (26–30)	-	36 (24–42)
Buccal organ length	64	32 (29–37)	-	28 (20–35)
Buccal organ width	16	38 (37–45)	45 (41–49)	28 (15–36)
Pharynx length	-	43 (38–47)	-	35 (29–40)
Pharynx width	48	42 (38–47)	50 (46–56)	38 (29–46)
Oesophagus length	256	-	-	130 (115–144)
Genital atrium length	80	-	99 (92–103)	38 (21–60)
Genital atrium width	128	177 (164–201)	129 (114–137)	84 (39–113)
Distance from anterior end to anterior margin of genital atrium	352	190 (177–270)	395 (360–441)	208 (107–306)
Genital atrium spine length	12	12 (10–16)	-	8 (7–10)
Number of testes	23	(13–22)	18 (17–19)	19 (15–21)
Testes length	48	-	-	34 (17–52)
Testes width	96	-	-	55 (41–104)
Egg length	224	240 (233–248)	-	122 (99–145)
Egg width	80	83 (68–92)	-	25 (22–28)

4.1.4.2 Kahawaia truttae (Dillon and Hargis, 1965) Lebedev, 1969

Class Monogenea Carus, 1863
Family Microcotylidae Taschenberg, 1879
Subfamily Microcotylinae Taschenberg, 1879
Genus *Kahawaia* (Dillon and Hargis, 1965) Lebedev, 1969

Type-host: Arripis trutta (Forster), Arripidae, "eastern Australian salmon" (Dillon and Hargis 1965b; present study; Lebedev 1969).
Other hosts: A. truttaceus (Cuvier), Arripidae, "western Australian salmon" (present study - new host record).

Site: A. trutta- gills (Dillon and Hargis 1965; Lebedev 1969); primary gill filaments (present study). *A. truttaceus*- primary gill filaments (present study).
Infection details: A. trutta- a total of 24 flukes collected from 23 fish; prevalence 48%; maximum intensity 5; mean intensity 2.2 (present study). *A. truttaceus*- a total of 83 flukes collected from 67 fish; prevalence 39%; maximum intensity 14; mean intensity 3.2 (present study).

Geographic localities: A. trutta- Timaru, Canterbury Province, South Island, New Zealand (Dillon and Hargis 1965); Great Australian Bight, SA, Australia (Lebedev 1969); Bermagui [36.4202 S, 150.0822 E], NSW, Australia (present study); *A. truttaceus*- Spencer Gulf (Whyalla [33.0731 S, 137.6147 E]), Gulf St. Vincent (Cape Jervis [35.6082 S, 138.0961 E]), Kangaroo Island (Kingscote [35.6576 S, 137.6439 E] and Emu Bay [35.6037 S, 137.5200 E]) and Coffin Bay [34.6224 S, 135.4628 E], SA, Australia (present study).

Specimens studied: A. trutta- 2 (Dillon and Hargis 1965); 5 (Lebedev 1969); 10 whole-mounts measured, 5 voucher specimens deposited in SAMA (AHC 29781, 29782 [2 slides], 29887–8), 2 voucher specimens deposited in NHM (NHM 2009.12.28.5–6) and 3 voucher specimens deposited in USNPC (USNPC 102678.00 [2 slides], 102679.00) (present study); *A. truttaceus*- 2 whole-mounts used for drawings (AHC 29767, AHC 29882), 10 whole-mounts measured, 6 voucher specimens deposited in SAMA (AHC 29767–9, 29882, 29885–6), 2 voucher specimens deposited in NHM (NHM

2009.12.28.3–4) and 2 voucher specimens deposited in USNPC (USNPC 102676.00–102677.00) (present study).

Redescription (Figure 7, Table 40)

Measurements of *K. truttae* from two localities (SA and NSW) on two host species (*A. truttaceus* and *A. trutta*) are presented in Table 40, with previously published measurements. Body elongate, fusiform (Figure 7A). Haptor clearly delineated from body, wedge-shaped, lined on both sides by single column of clamps. Clamps of typical *Microcotyle*-type, similar in structure (Figure 7D), dissimilar in size: middle clamps longer and wider than posterior clamps (Table 40).

Buccal organs of similar width and length; rim armed with small, sclerotised, tooth-like papillae. Pharynx similar in length and width, smaller than buccal organs. Oesophagus relatively long. Genital atrium (Figure 7B) consists of two laterally placed pads (one to right and one to left of midline); each pad armed with a circle of spines with hooked tips; spines of similar structure, dissimilar in size. Gut bifurcated; intestinal caeca with medial and lateral branching (branches not shown in Figure 7A but note haematin residues); unclear whether caeca unite posteriorly.

Testes post-ovarian, abundant (number not determined), closely abutting but not touching; situated inter-caecally. Vas deferens wide; follows sinuous path anteriorly in midline to genital atrium. Vitellarium commences posterior to genital atrium, occupying both lateral fields, not extending into haptor. Ovary tubular, coiled in midline irregularly. Genito-intestinal canal present, connects right intestinal caecum with common vitelline duct. Vitelline ducts anterior, common vitelline duct posterior, complete structure Y-shaped. Mehlis' gland and vaginal pore not observed. Two dorsal cuticularised pits present, one on each side of midline (Figure 7A). Uterus proceeding anteriorly in midline to genital atrium, obscured from view by vas deferens. Eggs *in utero* fusiform, with filaments at both ends (Figure 7C). Among live parasites collected, none was observed to lay eggs.

Remarks: Measurements for *K. truttae* collected in this study are presented in Table 40 for comparison with previously published accounts by Dillon and Hargis (1965) and Lebedev (1969). The redescription given here is warranted because the original

description by Dillon and Hargis (1965) was based upon limited material, and the subsequent redescription by Lebedev (1969) was poorly illustrated. This study also provides a new host record for *K. truttae* from *A. truttaceus*. Specimens of *K. truttae* from *A. trutta* off the coast of NSW did not differ from specimens collected from *A. truttaceus* in SA waters based on morphometrics (Table 40). However, there were minor differences between *K. truttae* from *A. trutta* and *A. truttaceus* in our study and specimens described by Dillon and Hargis (1965) and Lebedev (1969) from *A. trutta* as follows: (1) middle clamp width larger than described by Dillon and Hargis (1965), but smaller than described by Lebedev (1969), (2) posterior clamp length smaller than described by Dillon and Hargis (1965), (3) number of spines on left pad less than all previous descriptions and (4) egg length smaller than all previous descriptions. Despite these differences in measurements and spine counts the morphology of the new material from *A. trutta* and *A. truttaceus* agrees with *K. truttae*.

Molecular analysis

The 28S rRNA fragments for the majority of taxa sequenced were 850–950 bp, but shorter sequences were occasionally obtained due to poor primer binding. Analyses included sequences for *Kahawaia truttae* from *A. truttaceus* in SA waters and *A. trutta* off NSW, *Microcotyle arripis* from *A. georgianus* in SA waters, and five other microcotylid taxa and one outgroup taxon in the Discocotylidae (Table 38). From the NJ analysis, the microcotylid taxa separated into four clades, *Diplostamenides sciaenae*, *Polylabris sillaginae*, *Microcotyle* spp. (*M. arripis*, *M. sebastis*, *M. erythrinii* and Microcotylidae sp. M10) and *Kahawaia truttae* (Figure 8). *Microcotyle arripis* from *A. georgianus* grouped within the Microcotyle spp. clade, yet was distinct from other species within this clade. The 10 individuals within the *P. sillaginae* clade had identical sequences. Replicated samples of *K. truttae* from *A. truttaceus* and *K. truttae* from *A. trutta* grouped together within a single clade, with strong support (Bootstrap value 100% after 1,000 replicates; Figure 8). Within the *K. truttae* clade, 52 individuals of *K. truttae* from *A. truttaceus* and 9 individuals of *K. truttae* from *A. trutta* were identical (Figure 8), providing confidence in the morphological study that the microcotylid species from A. *truttaceus* and *A. trutta* is a single taxon, *K. truttae*.

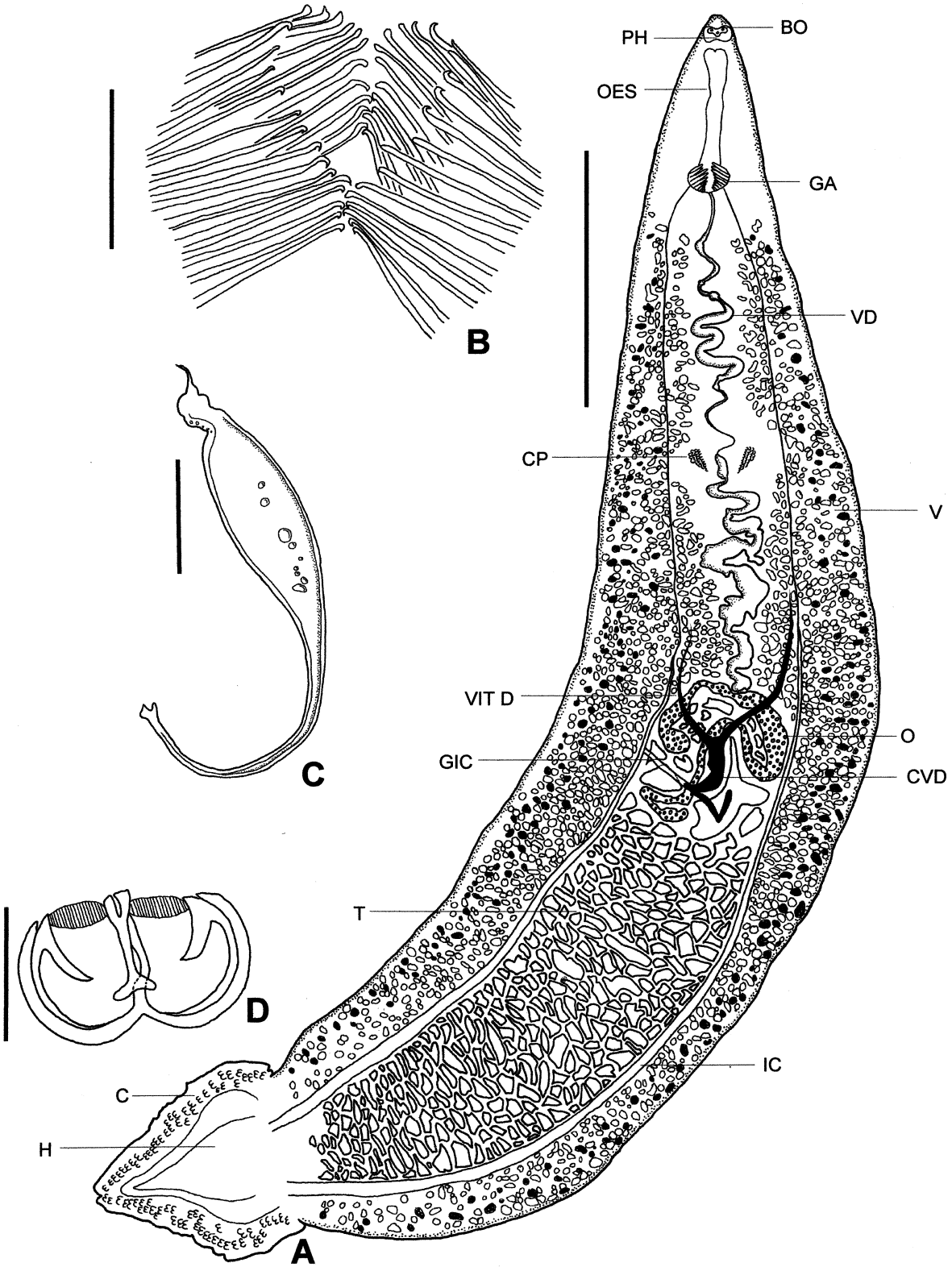

Figure 7. *Kahawaia truttae* (Dillon and Hargis, 1965) Lebedev, 1969 from *Arripis truttaceus*. A. Whole-mount (ventral view) drawn from AHC 29767; B. Genital atrium drawn from AHC 29767; C. Egg, clearly distorted from normal shape, drawn *in utero* from AHC 29882; D. Clamp drawn from AHC 29767. Abbreviations as for Figure 6. Additional abbreviations: CP, Cuticularised Pits; GIC, Genito-intestinal Canal; VD, Vas deferens. Scale bars: A = 1.5 mm, B = 75µm, C = 100µm, D = 35 µm.

Table 40. Comparative measurements of *Kahawaia truttae* (Dillon and Hargis, 1965) Lebedev, 1969, for specimens collected in this study and published data (measurements in micrometres; mean followed by range in parentheses; -, indicates no measurement available; R = right side, L = left side).

Characteristic	**New Zealand as *Gonoplasius truttae* (Dillon and Hargis, 1965)**	**Great Australian Bight, South Australia (SA) (Lebedev, 1969)**	**Spencer Gulf, Gulf St. Vincent and Kangaroo Island, SA (present study)**	**Bermagui, New South Wales (present study)**
Host species	*Arripis trutta*	*A. trutta*	*A. truttaceus*[a]	*A. trutta*
Number of specimens	n=2	n=5	n=10	n=10
Body length	11500 (11000–12000)	(7800–17100)	7867 (3897–11292)	10231 (5309–14455)
Body width	1480 (1440–1530)	(900–1900)	1320 (717–2322)	1361 (1239–1582)
Haptor length	1610 (1560–1650)	-	1078 (594–1349)	1649 (758–2268)
Haptor width	-	-	922 (471–1825)	993 (784–1288)
Number of clamps (R/L)	(56–57/51–52)	(48–57/48–57)	47/45 (36–60/38–55)	49/53 (41–59/44–63)
Middle clamps length	46 (45–47)	-	32 (25–41)	40 (30–49)
Middle clamps width	37 (35–39)	86	60 (50–65)	71 (66–75)
Posterior clamps length	75 (71–79)	-	25 (20–30)	30 (22–38)
Posterior clamps width	59 (56–62)	-	42 (38–53)	51 (41–60)
Buccal organ length	61 (58–65)	-	63 (53–77)	76 (63–98)
Buccal organ width	63 (61–65)	(50–90)	65 (54–76)	83 (56–97)
Pharynx length	43 (36–50)	-	54 (40–69)	59 (51–67)
Pharynx width	43 (36–50)	-	52 (38–68)	60 (55–64)
Genital atrium length	117 (115–119)	(113–288)	129 (106–151)	146 (127–169)
Genital atrium width	187 (184–191)	(163–363)	194 (135–310)	193 (163–231)
Number of spines (R/L)	27/34	28/33	28/28 (24–31/24–31)	27/28 (25–29/26–30)
Vaginal pore diameter	165	-	-	-
Vaginal pore distance from genital atrium	825	-	-	-
Egg length	241 (227–254)	275	175 (139–197)	190 (168–222)
Egg width	75 (68–83)	113	58 (31–72)	66 (59–83)

[a]New host record

4.1.5 Discussion

Based on morphology and host, the monogenean species from *Arripis georgianus* was identified as *Microcotyle arripis*, originally described by Sandars (1945). Phylogenetic analysis of partial 28S rRNA nucleotide sequence data using NJ supported the morphological conclusion, with *M. arripis* grouping within the *Microcotyle* spp. clade, but observed to be distinct from all other species (Figure 8). There is no published record of this parasite species from any other fish species, and therefore *M. arripis* likely exhibits host-specificity to *A. georgianus*. This is not unexpected as monogeneans are among the most host-specific of parasites in general (Rohde 1979; Whittington *et al.* 2000).

Kahawaia truttae was first recorded as *Gonoplasius truttae* Dillon and Hargis, 1965 from *A. trutta* at Timura, South Island, New Zealand (Dillon and Hargis 1965a), but after analysis of morphological structures, was re-classified by Lebedev (1969) into a new genus, *Kahawaia*. Morphological study suggested the monogenean species from *A. truttaceus* in SA waters was identical to the monogenean species from *A. trutta* in waters off the coast of NSW, namely *K. truttae*, which therefore represents a new host record for *K. truttae* from *A. truttaceus*. Molecular analyses supported this hypothesis, with 52 individuals of *K. truttae* from *A. truttaceus* and 9 individuals of *K. truttae* from *A. trutta* grouping together within the same clade with strong support (Bootstrap value 100% after 1000 replicates; Figure 8).

To enable further insight into the relationships within Microcotylidae established here, it would be preferable to analyse more sequence data from a number of nuclear genes including both coding and non-coding regions (McManus and Bowles 1996) and run analyses to incorporate these multi-gene data. Furthermore, incorporation of some sequence data from mitochondrial genes may shed further light on the phylogenetic relationships among microcotylids. An improved appreciation and understanding of informative morphological characters that separate microcotylid genera and species would contribute significantly to unravelling the evolutionary history of these higher polyopisthocotyleans.

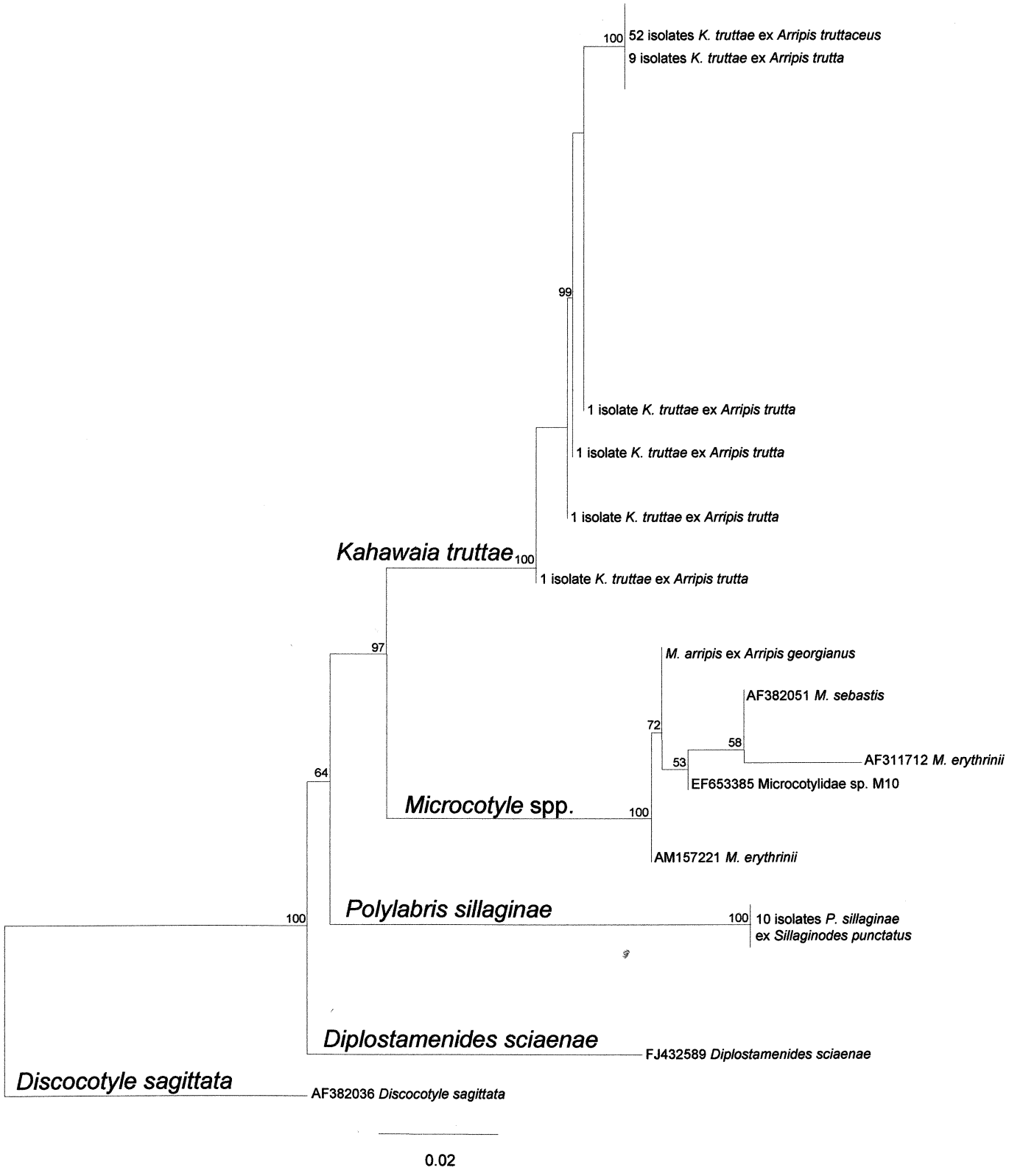

Figure 8. Neighbor-joining tree from analyses of the 28S rDNA nucleotide sequence data for Polyopisthocotylea including seven microcotylid taxa and one outgroup taxon (see Table 38). Note that the top branch represents a total of 61 identical *Kahawaia truttae* sequences from *Arripis trutta* and *A. truttaceus*. Bootstrap values (≥ 50) are shown above the branches.

4.2 Redescription of an atypical species of trebiid copepod

4.2.1 Abstract

The trebiid copepod *Kabataia ostorhinchi* Kazachenko, Korotaeva and Kurochkin, 1972 is redescribed from female specimens and male specimens are described for the first time from new material collected from the gills of the type-host *Oplegnathus woodwardi* Waite, 1900 [= *Ostorhinchus conwaii*] captured in the Southern Ocean, off Port MacDonnell, Australia. *Kabataia ostorhinchi* and *Innaprokofevnas orientcolae* Kazatchenko 2001 are the only trebiids known to infect actinopterygian (ray-finned) hosts, while all other members of the family infect elasmobranch fishes. Scanning electron micrographs confirm the atypical body plan of *Kabataia*, which exhibits functional articulation between the first pedigerous somite and second pedigerous segment. The second and third pedigerous somites are fused and exhibit no flexure and the fourth pedigerous somite is free. The unusual structure of *K. ostorinchi* is hypothesised to be progressive adaptation resulting from an ancestral host switch from elasmobranchs to teleost fish hosts. Deposition of specimens in a curated museum collection will enable future research into phylogenetic relationships within the Trebiidae.

4.2.2 Introduction

Parasitic copepods have modified structures from progressive adaptation to hosts and habitats. Cephalisation - the incorporation of successive segments into the cephalothorax, often associated with some loss of segmentation - appears to be the mechanism responsible for the development of the dorsal shield in siphonostome copepods (Kabata 1979). This structure has evolved as the means of adhering to the surface of fishes, without losing the ability to move over it freely. Progressive loss of segmentation can be observed among siphonostome copepods other than those with caligoid characteristics. Parasites that live permanently attached to gill filaments or other parts of the host have no need for it, hence, cephalisation is not usually observed in these species.

The process of cephalisation can be traced in a group of siphonostome families related to the Caligidae (Figure 9). The most primitive among them is the Dissonidae, exemplified by *Dissonus*, where only the first leg-bearing segment has been incorporated into the

cephalothorax and segmentation is exhibited between all four pedigerous somites (Kabata 1979; Boxshall *et al.* 2008) (Figure 9). This type of segmentation occurs in the majority of free-living copepods. *Trebius* spp. exhibit fusion of the first and second pedigerous somites, while *Caligus* spp. exhibit fusion of the first three predigerous somites (Figure 9). It is generally accepted that *Dissonus* is more primitive than *Trebius*, while the latter is less advanced than *Caligus*. This is exemplified by the fact that those families considered to be 'more primitive' are generally parasitic on 'more primitive' hosts. For example, *Dissonus* live on elasmobranch and teleost hosts and *Trebius* spp. live exclusively on elasmobranch hosts, while *Caligus* live predominantly on teleost hosts (Kabata 1979; Deets and Dojiri 1989).

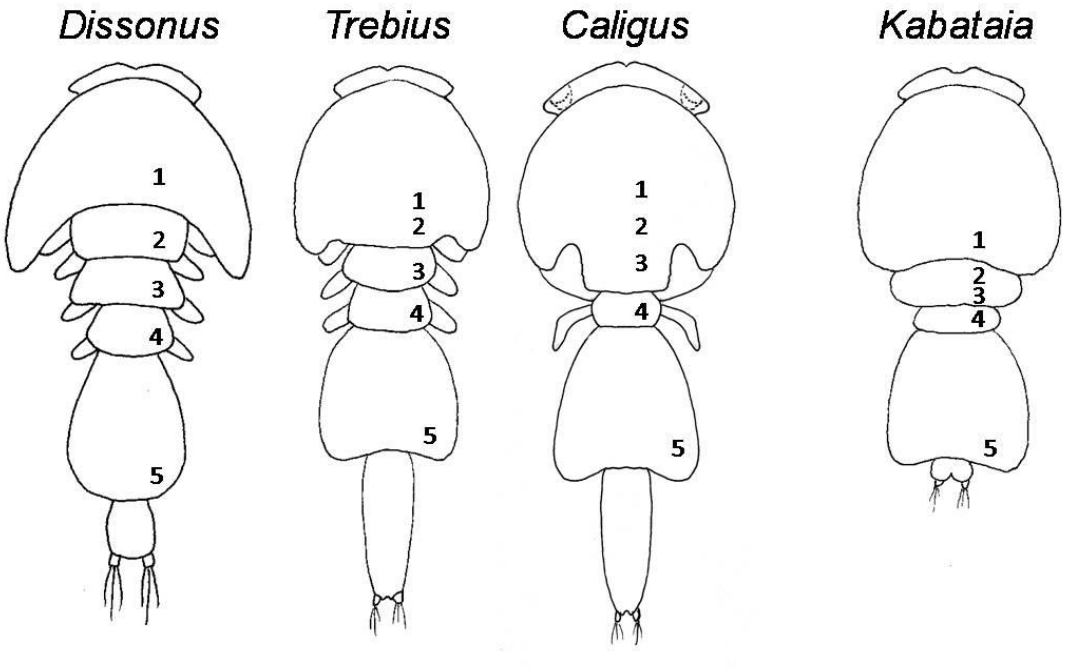

Figure 9. Progress of cephalisation in caligiform copepods, associated with development of the dorsal shield showing incorporation of the first (1), second (2), and third (3) pedigerous somites into the cephalothorax [Kabata, 1979; Boxshall & Halsey 2004].

Segmentation in trebiids is not consistent between genera. Trebiidae contains 3 genera, *Trebius* Krøyer, 1838 with 14 species, monotypic *Kabataia* Kazachenko, Korotaeva and Kurochkin, 1972 and monotypic *Innaprokofevnas* Kazatchenko, 2001. *Trebius* and *Innaprokofevnas* can be distinguished from *Kabataia* by the anterior cephalothorax incorporating the first and second pedigerous somites followed by 2 free pedigerous somites (segments 3 and 4; Figure 9). *Kabataia ostorhinchi* is atypical because it has a fused second and third pedigerous segment (Figure 9) that bears lateral plates. All species of *Trebius* infect

elasmobranchs, whereas *Innaprokofevnas orientcolae* is a parasite of a deep sea roughscale sole (*Clidoderma asperrimum*) (see Kabata 1979) and *K. ostorhinchi* is a parasite of a deep sea teleost fish (*Oplegnathus woodwardi*) (see Kazatchenko *et al.* 1972). The segmentation of *Kabataia ostorhinchi* is particularly curious, considering it lives on the gills of its teleost host, where cephalisation may not be as crucial compared to living on the body surface (Kabata 1979).

We provide the second ever record of *K. ostorhinchi* from the type-host *Oplegnathus woodwardi* [= *Ostorhinchus conwaii* see Gomon *et al.* (2008)] and report and describe male specimens for the first time. Fusion and segmentation in the body plan of *K. ostorhinchi* is confirmed using scanning electron microscopy of female and male specimens.

4.2.3 Materials and Methods

Four knifejaw, *Oplegnathus woodwardi,* were captured by recreational fishers on the slopes of the continental shelf off Port MacDonnell, South Australia, in May 2007 and May 2008. Copepods were recovered from the gills using a dissecting microscope and fixed in 70% ethanol. Specimens were cleared in lactophenol prior to morphological examination and drawings were made using a drawing tube. Selected specimens were measured using a calibrated eyepiece micrometer under a compound microscope and drawings were made with the aid of a camera lucida. Anatomical terminology follows Dojiri & Cressey (1987) and Huys & Boxshall (1991). Parasite prevalence and intensity, followed by the range of parasites recovered in parentheses, are given in whole numbers and follow Bush *et al.* (1997). The total length (L_T) range of parasitised hosts is presented in millimetres, followed in parentheses by the total length range of all fish examined and the total number of hosts studied. Five female and three male specimens were prepared for scanning electron microscopy (SEM). Specimens were dehydrated in an acetone series (70; 80; 90; 95; 100; 100%) before critical point drying and gold-platinum coating. Representative specimens were deposited in the Natural History Museum, London (BMNH).

4.2.4 Results

4.2.4.1 Kabataia ostorhinchi *Kazachenko, Korotaeva and Kurochkin, 1972*

Descriptions

Trebiidae

Kabataia Kazachenko, Korotaeva and Kurochkin, 1972

Kabataia ostorhinchi Kazachenko, Korotaeva and Kurochkin, 1972

Host and locality: *Oplegnathus woodwardi* (Waite), Oplegnathidae. Slopes of the continental shelf, off Port MacDonnell, South Australia, Australia.
Site: Gills; attached to secondary gill lamellae.
Prevalence and intensity: 100, 38 (6-89); L_T 401 (385-430) mm; n = 4.
Material examined: 21♀♀; 6♂♂ (including 3♂♂ and 5♀♀studied by SEM).
Material accessioned: BMNH 2007.947=6♀♀; BMNH 2009.263-264=2♂♂; BMNH 2009.265-274=10♀♀.

Adult female (Figures 10-13): Body dorsoventrally flattened, comprising caligiform anterior cephalothorax in form of dorsal shield (Figures 10A, 11A,D). Dorsal shield and lateral plates with thin marginal membranes (Figure 10A, 11A). Cephalothorax incorporating first pedigerous somite (ventrally bearing leg 1) (Figures 10A, 11B-E) and fused second and third pedigerous segment (ventrally bearing legs 2 and 3). Surface suture dorsal (Figures 10A, 11B). One free pedigerous somite following, ventrally bearing leg 4 (Figure 11B). Genital complex apple-shaped (Figure 10A, B), consisting of fused fifth pedigerous and genital somites, ventro-laterally bearing leg 5 (Figures 12F, 13D). Caudal ramus approximately as long as wide, medial margin bearing setules and armature consisting of 3 + 2 pinnate seta following single maginal naked seta (Figure 10C).

First antenna 2-segmented, first segment with 29 setae (27 anteroventral plumose setae and 2 dorsal plumose seate); second segment with 13 setae (1 on base). Second antenna three segmented; second segment largest, with blunt projection (Figure 12A); third segment with a simple hamulus with two naked setae (Figures 10D, 12A). Basal part of postantennary process carrying two clusters of sensory papillae, another similar, but isolated cluster, on the sternum (Figure 12B). Mouth cone (Figure 12C) caligoid, labrum and labium each with marginal membrane about buccal orifice. Maxilla (Figure 10E) single dentiform process composed of 2 papillae, of approximately equal length. Second maxilla branchiform; flabellum absent; callamus with 3 rows of spiralling serrated

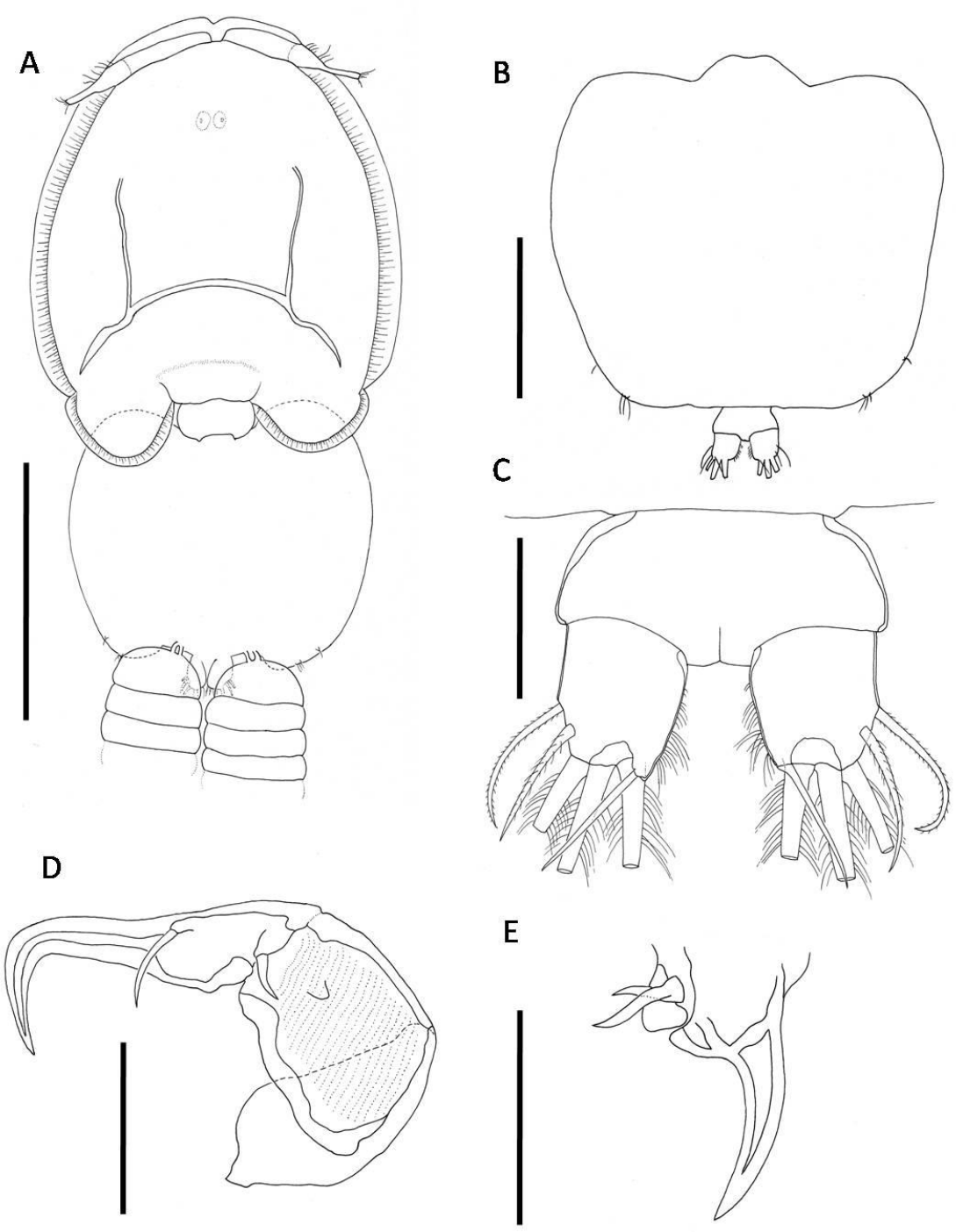

Figure 10. *Kabataia ostorhinchi*, adult female A-D. A. Dorsal view, with egg sacs, scale = 1mm. B. Ventral view of genital complex, scale = 500µm C. Caudal ramus, ventral view, scale = 100µm. D. Second antenna, scale = 100 µm. E. Maxilla, scale = 50µm.

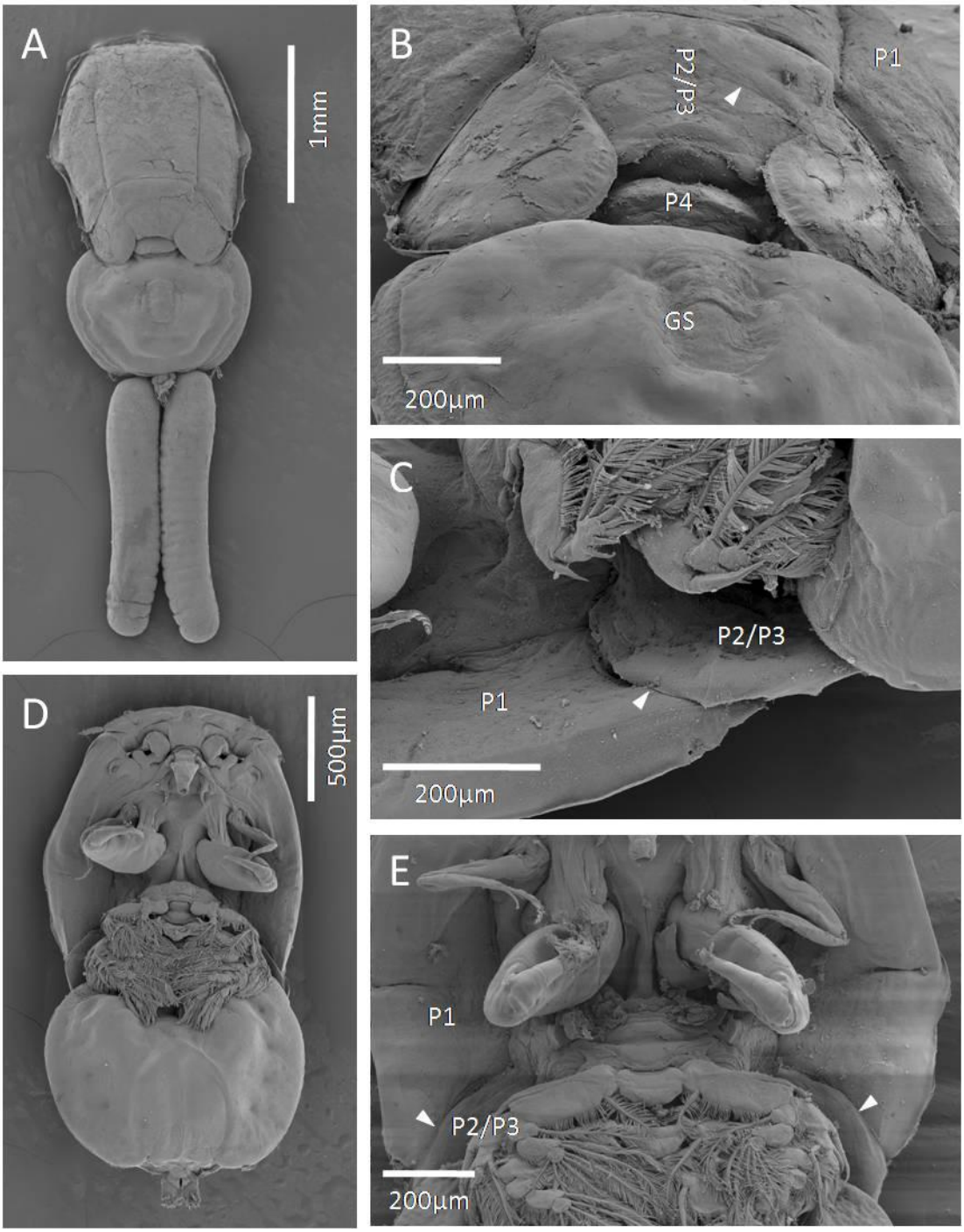

Figure 11. Scanning electron micrographs A-E. *Kabataia ostorhinchi*, adult female. A. Dorsal view, whole specimen with egg sacs. B. Dorsal view showing free first pedigerous somite (P1), fused second and third pedigerous segment (P2/P3) and free fourth pedigerous somite (P4); arrowhead indicates surface suture. C. Ventral view, showing boundary (arrowhead) between the first free pedigerous somite (P1) and fused second and third pedigerous segment (P2/P3). D. Ventral view, whole specimen without egg sacs. E. Ventral view, first leg removed, showing definition between first and second pedigerous segment (arrowheads).

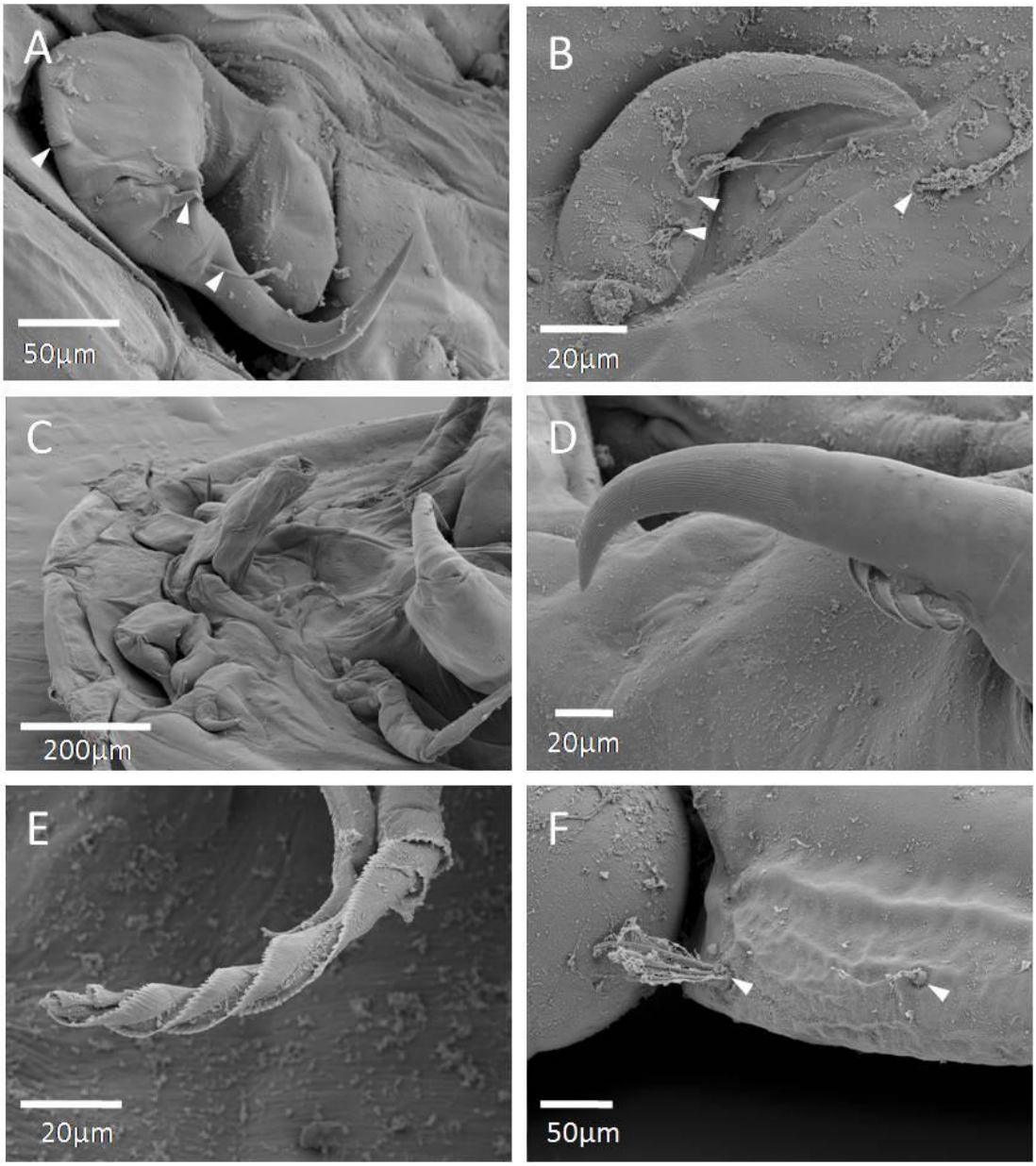

Figure 12. Scanning electron micrographs A-F. *Kabataia ostorhinchi*, adult female. A. Second antenna with blunt projection (left hand arrowhead) and setae (right hand arrowheads). B. Postantennal process showing three clusters of sensory papillae (arrowheads). C. Ventral view showing mouth tube opening. D. Ventral view, maxilliped, showing ridged tip of claw. E. Ventral view, tip of maxilla. F. Lateral view of female with eggs showing seta (right hand arrowhead) and fifth leg (lefthand arrowhead) on the genital complex.

membranes (Figure 12E); canna with single finely serrated membrane. Maxilliped 2-segmented, robust corpus with 2 large patches of minute denticals on distal outer margin; claw denticulated at tip (Figure 12D). Sternal furca absent.

Swimming legs 1–4 biramous (Figure 5). Armature on rami of legs 1–4 as follows (Roman numerals = spines; Arabic numerals = setae):

	Coxa	Basis	Endopod	Exopod
leg 1	0-1	0-1	0-0; 0-3	I-0; II-5
leg 2	0-1	0-1	0-1; 0-2; 0-6	I-1; I-1; III-4
leg 3	0-1	0-1	0-1; 0-1; 0-4	I-1; I-1; III-4
leg 4	0-1	0-1	0-1; 0-3	I-1; I-1; III-4

Leg 1 (Figure 13A) biramous. Coxa with small lateral plumose seta. Basis with small inner plumose seta. Endopod 2- segmented. First segment unarmed, second segment with 3 plumose seta and lateral setules. First exopodal segment with large distolateral denticultated spine and medial setules. Distal segment smaller, armed with medial setules followed by 5 pinnate setae (3 large + 2 small) and 2 denticulated spines. Interpodal bar narrow and lacking marginal membrane.

Leg 2 biramous. Coxa with small lateral plumose seta. Basis with large pinnate medial seta, medial setules. Endopod 3-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with lateral setules and 2 distomedial pinnate seta. Third segment smallest, with six pinnate setae. Exopod 3-segmented. First segment largest, with large distolateral denticulated spine and distomedial pinnate seta. Second segment with distolateral denticulated spine medial and 1 distomedial pinnate seta. Third segment with 3 denticulated spines, increasing in size with the third being the largest, followed by 4 long pinnate setae. Interpodal bar with wide marginal membrane.

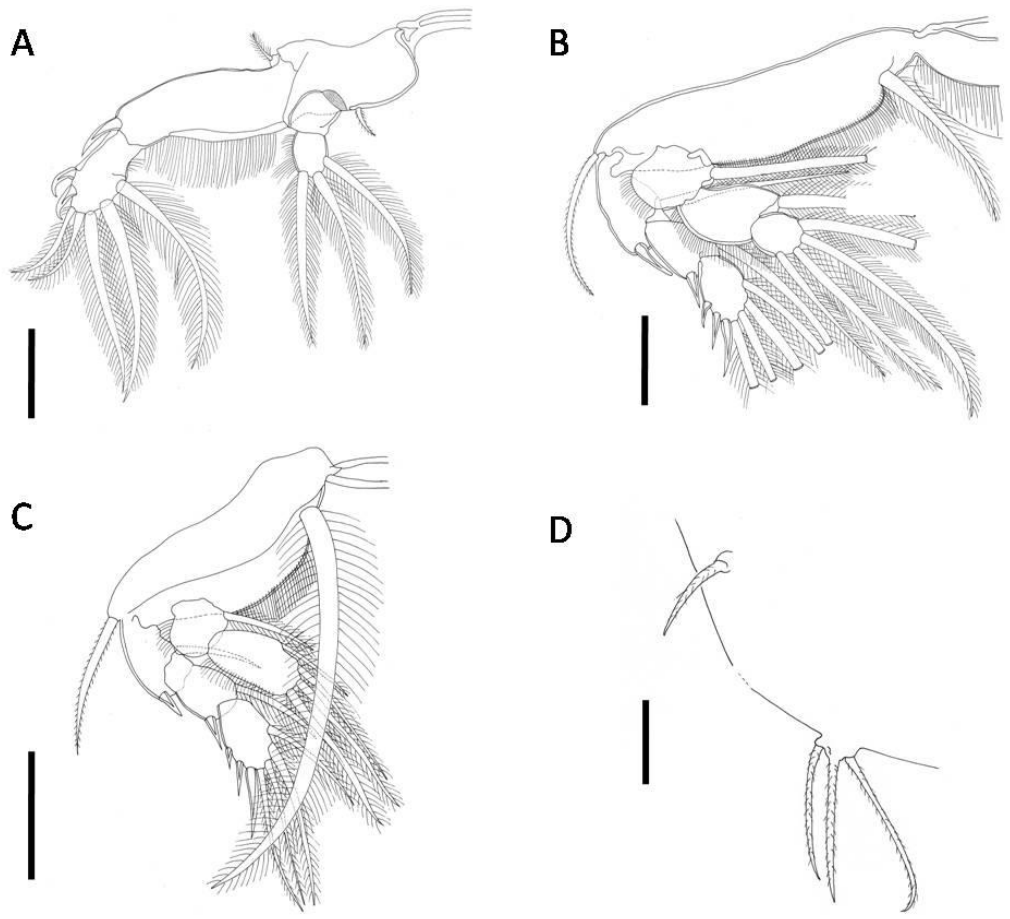

Figure 13. *Kabataia ostorhinchi*, adult female A-D. A. Leg 1, scale = 100 µm. B. Leg 3, scale = 100 µm. C. Leg 4, scale = 100 µm. D. Leg 5, scale =50 µm.

Leg 3 biramous (Figure 13B). Coxa with large, distolateral plumose seta. Basis with large pinnate medial seta, medial setules. Endopod three-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with lateral setules and distomedial pinnate seta. Third segment smallest, with lateral setules and 4 pinnate setae. Exopod 3-segmented. First segment largest with large distolateral denticulated spine and distomedial pinnate seta. Second segment with distolateral denticulated spine and distomedial pinnate seta. Third segment with 3 denticulated spines, increasing in size with the third being the largest, followed by 4 long pinnate setae. Interpodal bar with wide marginal membrane.

Leg 4 (Figure 13C) biramous. Coxa with large, distolateral plumose seta. Basis with extremely large pinnate medial seta, medial setules. Endopod 2-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with 4 pinnate setae. Exopod 3-segmented. First segment largest with large distolateral denticulated spine and distomedial pinnate seta. Second segment with with lateral setules and distolateral

denticulated spine and distomedial pinnate seta. Third segment with 3 denticulated spines, increasing in size with the third being the largest, followed by 4 pinnate setae. Interpodal bar lacking marginal membrane.

Leg 5 (Figures 10C, 12F) vestigial, represented by two setiferous lobes. Anterior lobe bearing single, small plumose seta, posterior lobe bearing 3 small plumose seta.

Adult male (Figures 14-16): Body, dorsoventrally flattened, comprising caligiform anterior cephalothorax in form of dorsal shield (Figure 14A). Dorsal shield and lateral plates with thin marginal membranes (Figures 14A, 15A). Cephalothorax incorporating first pedigerous somite (ventrally bearing leg 1) and fused second and third pedigerous segment (ventrally bearing legs 2 and 3) (Figures 15A-E). Surface suture dorsal, more prounced than in female (Figures 14A, 16D). One free predigerous somite following, ventrally bearing leg 4 (Figure 14A). Round genital complex (Figure 14B), ventrally bears legs 5 and 6 (Figure 15F); 2 segmented abdomen. Caudal ramus armed as in female (Figures 14A, B).

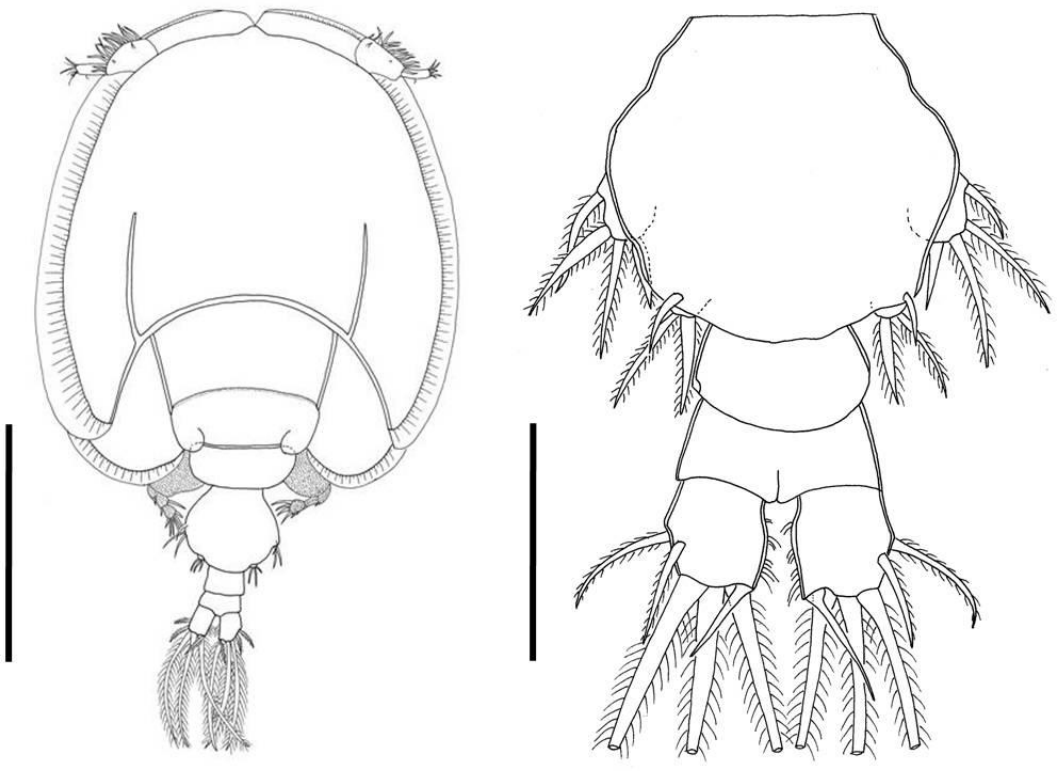

Figure 14. *Kabataia ostorhinchi*, adult male A-B. A. Dorsal view, scale = 1mm. B. Dorsal view of genital complex, including legs 5 and 6, scale = 200µm.

First antenna 2-segmented, first segment with 29 setae (27 anteroventral plumose setae and 2 dorsal plumose seate); second segment with 13 setae (1 on base). Second antenna 3-segmented, second segment with 5 large crenulated adhesion pads (Figures 16A, B) and corrugated processes; third segment a curved claw bearing 2 large naked setae and corrugated adhesion pad. Basal part of postantennary process carrying two clusters of sensory papillae, another isolated cluster on the sternum. Mouth cone (Figure 16D) caligoid, labrum and labium each with marginal membrane about buccal orifice. Maxilla (Figure 16E) robust projection, dentiform with 1 large and 3 small accessory processes; base composed of 2 large and 1 medium setae. Second maxilla branchiform; flabellum absent; callamus with 3 rows of spiralling, serrated membranes; canna with 2 rows of serrated membranes; base with long, ribbon-like extension (Figure 16A). Maxilliped 2- segmented, robust corpus with 3 large patches of scales on distal outer margin (Fig 16F); claw ridged at tip; myxal process opposing the tip of the claw (Figure 16G). Sternal furca absent.

Legs 1–4 biramous armature on rami of legs 1–4 as follows (Roman numerals = spines; Arabic numerals = setae):

	Coxa	Basis	Exopod	Endopod
leg 1	0-1	0-1	I-0; II-5	0-0; 0-3
leg 2	0-1	0-1	I-1; I-1; III-4	0-1; 0-1; 0-6
leg 3	0-1	0-1	I-1; I-1; III-4	0-1; 0-1; 0-4
leg 4	0-1	0-1	I-1; I-1; III-4	0-1; 0-1; 0-3

Leg 1 as in female. Biramous. Coxa with small lateral plumose seta. Basis with small inner seta. Endopod 2- segmented. First segment unarmed, second segment with 3 plumose seta and lateral setules. First exopodal segment with large distolateral denticultated spine and medial setules. Distal segment smaller, armed with medial setules followed by 5 pinnate setae (3 large + 2 small) and 2 large denticulated spines. Leg one interpodal bar narrow and lacking marginal membrane.

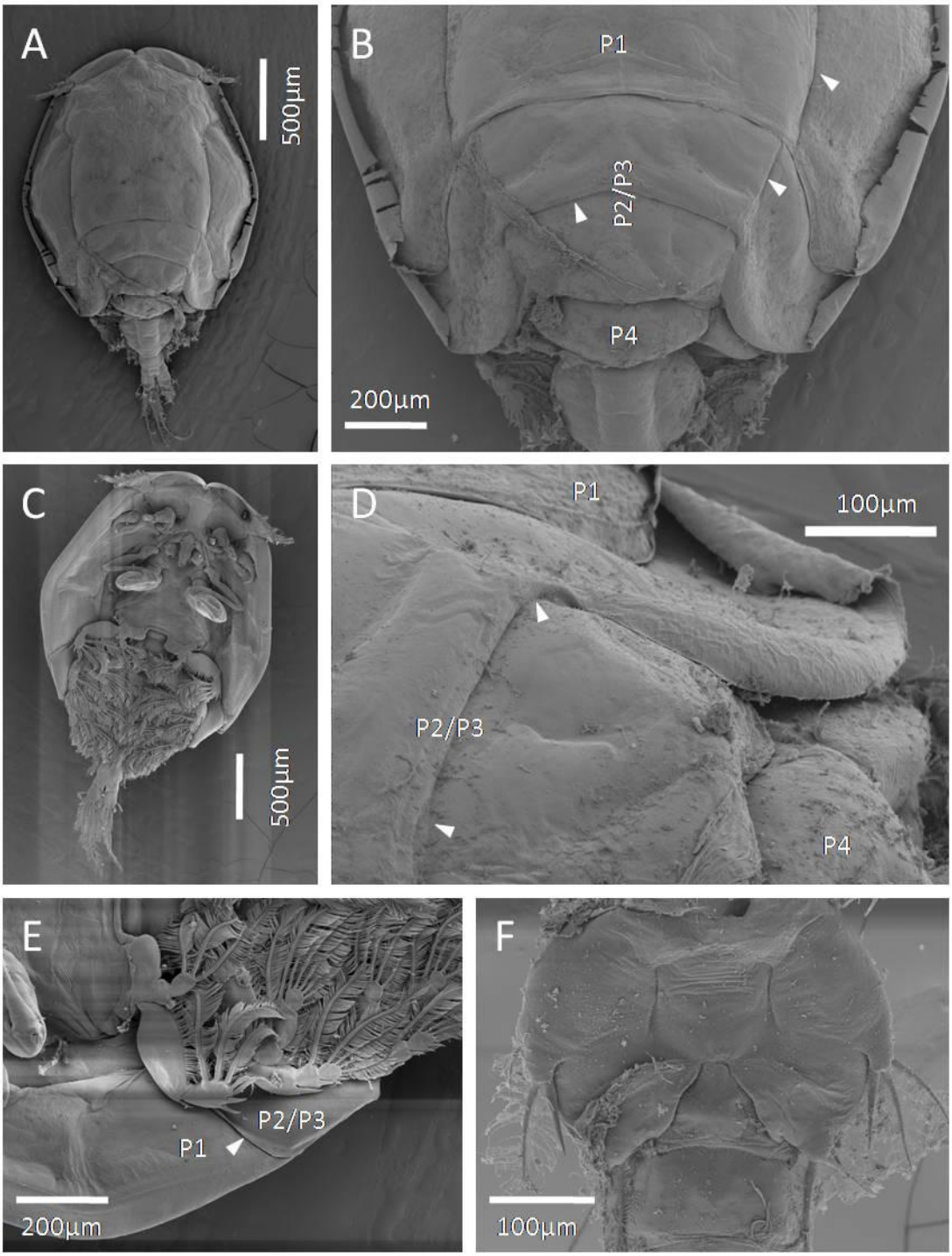

Figure 15. Scanning electron micrographs A-F. *Kabataia ostorhinchi*, adult male. A. Whole specimen, dorsal view. B. Dorsal view showing surface sutures indicated by arrowheads and segmentation of pedigerous somites C. Whole specimen, ventral view. D. Dorsal view, showing functional and non-functional parts of pedigerous somites; suture lines between second (P2) and third (P3) pedigerous somites indicated by arrowheads. E. Ventral view showing segmentation (arrowhead) between first (P1) fused second and third pedigerous segment (P2/P3). F. Leg 5 and leg 6.

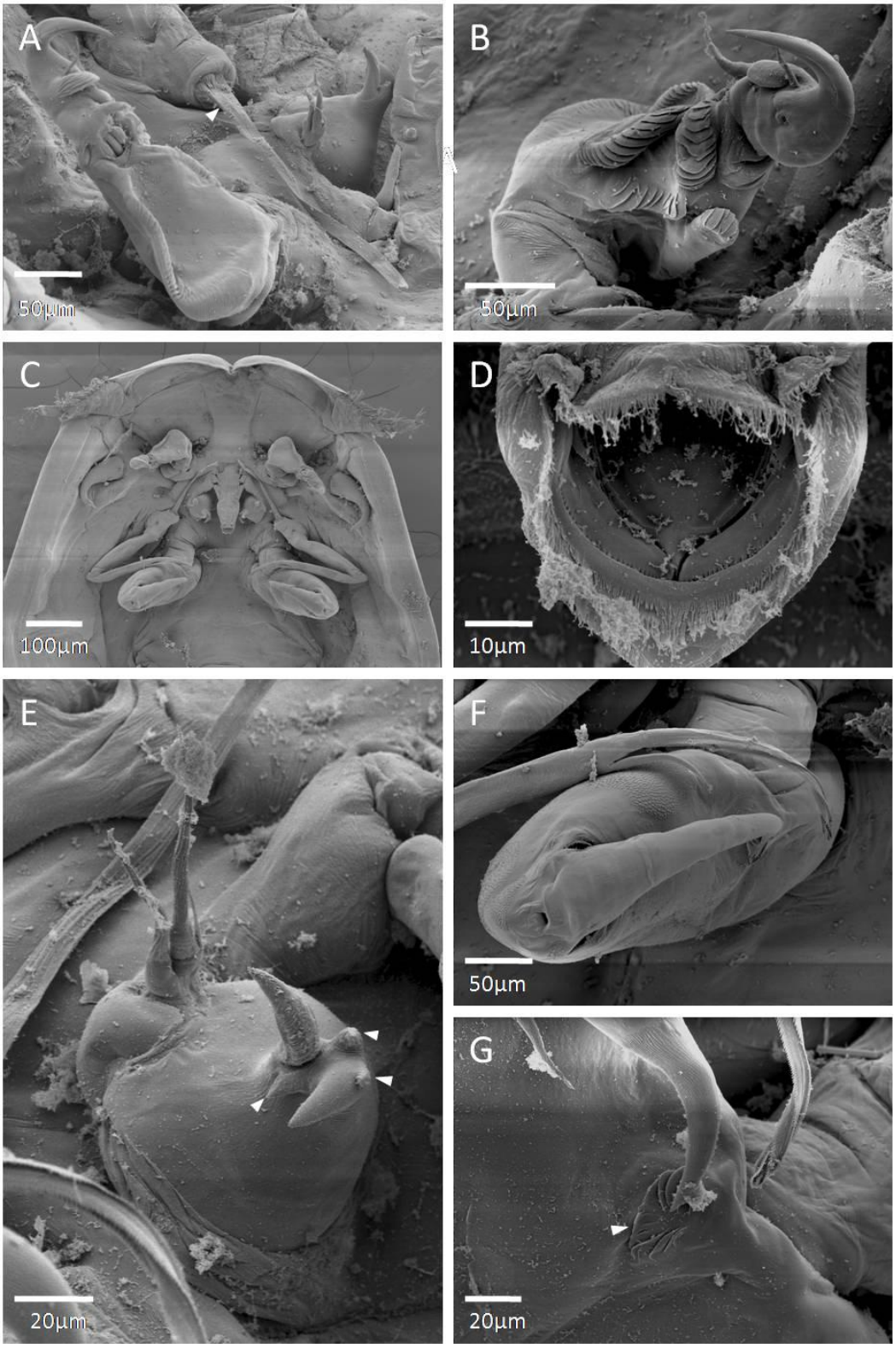

Figure 16. Scanning electron micrographs A-G. *Kabataia ostorhinchi*, adult male. A. Base of maxilla showing ribbon extension (arrowhead); second antenna in foreground; maxilla in background. B. Second antenna detail showing adhesion pads. C. Ventral view of male feeding structures. D. Mouth cone opening. E. Maxilla showing 3 small accessory processes (arrowheads); base composed of 2 large and 1 medium setae. F. Maxilliped and scale detail. G. Claw of maxilliped showing myxal process (arrowhead) opposing the tip of the claw.

Leg 2 biramous. Coxa with small lateral plumose seta. Basis with large pinnate medial seta, medial setules. Endopod 3-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with lateral setules and distomedial pinnate seta. Third segment smallest, with lateral setules and 6 pinnate setae. Exopod 3-segmented. First segment largest, with large distolateral denticulated spine and distomedial pinnate seta. Second segment with distolateral denticulated spine and 1 distomedial pinnate seta. Third segment with 3 denticulated spines, the third being the largest, followed by 4 long pinnate setae. Interpodal bar with wide marginal membrane.

Leg 3 as in female. Biramous. Coxa with small lateral plumose seta. Basis with large pinnate medial seta, medial setules. Endopod 3-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with lateral setules and distomedial pinnate seta. Third segment smallest, with 4 pinnate setae. Exopod 3-segmented. First segment largest, with large distolateral denticulated spine and distomedial pinnate seta. Second segment with distolateral denticulated spine and 1 distomedial pinnate seta. Third segment with 3 denticulated spines, the third being the largest, followed by 4 long pinnate setae. Interpodal bar with wide marginal membrane.

Leg 4 biramous. Coxa with lateral plumose seta. Basis with large pinnate medial seta, medial setules. Endopod 3-segmented. First segment with lateral setules and distomedial pinnate seta. Second segment with lateral setules and distomedial pinnate seta. Third segment smallest with 3 pinnate setae. Exopod 3-segmented. First segment largest, with large distolateral denticulated spine and distomedial pinnate seta. Second segment with distolateral denticulated spine and 1 distomedial pinnate seta. Third segment with 3 denticulated spines, the third being the largest, followed by 4 long pinnate setae. Interpodal bar lacking marginal membrane.

Leg 5 and 6 (Figure 14B, 15F) vestigial. Leg 5 represented by two lobes. Anterior lobe bearing single, plumose seta, posterior lobe bearing 3 plumose seta. Leg 6 represented by a single lobe bearing 2 large and 1 small plumose setae.

4.2.4 Discussion

Until the early 1970s, the Trebiidae contained only its type genus *Trebius* and the family diagnosis was identical with that of the genus. Changes in the diagnosis were required on the discovery of *K. ostorhinchi* which differs from *Trebius* spp. in having lateral plates on the second, fused pedigerous segment and in lacking the sterna furca. Scanning electron micrographs in this study show fusion of the second and third pedigerous segments in male and female specimens, confirming the atypical body plan of *K. ostorhinchi* compared with other species of the family.

Like many caligiform copepods, *Trebius* spp. are typically found on the general body surface or in the branchial chambers of their hosts (Kabata 1979; Deets and Dojiri 1989). Interestingly, Nagasawa *et al.* (1998) found that *Trebius shiinoi* is an endoparasite of the uterine linings and on embryos within the uterus of adult female clouded angel shark (*Squatina nebulosa*). The authors comment that this phenomenon is particularly unusual given that *T. shiinoi* seems little modified, if at all, to lead an endoparasitic existence. Fusion of the second and third pedigerous somites observed in *K. ostorhinchi* has enabled extension of the dorsal plate to incorporate the fused segment. It is likely that this atypical body plan of *K. ostorhinchi* may benefit its ability to manoeuvre and attach to the secondary gill lamellae of its teleost host.

The atypical body plan observed in *K. ostorhinchi* could be a result of a host-switch event. A host-switch could have been made by a common copepod ancestor infecting deep water elasmobranchs (i.e. species of *Dissonus* or *Trebius*) to a deep sea teleost host, or perhaps even *Innaprokofevnas orientcolae* from its pleuronectid host to *Oplegnathus woodwardi* (Figure 9). Specimens accessioned in the Natural History Museum in the current study will be valuable for future molecular studies to investigate the phylogeny of trebiids and their relatives. In view of the under-exploration of the deep sea environment (Webb *et al.* 2010), it is likely that more trebiids will be discovered in future that may contribute further to this hypothesis.

5 MOLECULAR TOOLS TO DISTINGUISH PARASITE SPECIES

5.1 *Paradeontacylix* n. sp. (Digenea: Aporocotylidae) infects three *Seriola* species (Perciformes: Carangidae) in three oceans

5.1.1 Abstract

Three wild *Seriola* species including amberjack *Seriola dumerili* (Risso, 1810), Samson fish *S. hippos* Günther, 1876 and yellowtail kingfish *S. lalandi* Valenciennes, 1833, (Perciformes: Carangidae), were examined for parasitic fish blood flukes in the Indian, Southern and Pacific Oceans off the southern coast of Australia. Four *Paradeontacylix* species (Digenea: Aporocotylidae) were found: *P. sanguinicoloides* McIntosh 1934, *P. godfreyi* Hutson & Whittington 2006, *P.* cf. *kampachi* and a new species. *Paradeontacylix* n. sp. was detected in all *Seriola* spp. examined and is described from the heart of *S. hippos* Günther, 1876 (type host) from the Indian Ocean off Rottnest Island, Western Australia (type locality). A key to the species of *Paradeontacylix* is provided. The new species is most easily distinguished from other *Paradeontacylix* species by a combination of: (1) a body length of ~2,800–3,650 μm; (2) enlarged posterior tegumental spines; (3) a bi-lobed posterior margin; (4) a maximum number of 15 marginal tegumental spines in rows; (5) 46–48 intercaecal testes; (6) a testicular field occupying 30–41% of the total body length; (7) a portion of the uterus (58–105 μm) descending posterior to the seminal receptacle. The new species most closely resembles *P. sanguinicoloides*, which differs by having short, rose-thorn shaped posterior tegumental spines. New locality records are provided for *P. godfreyi* at Sir John Young Banks, New South Wales and *P. sanguinicoloides* and *P.* cf. *kampachi* off Rottnest Island, Western Australia. A phylogenetic analysis is provided for *Paradeontacylix* spp. based on molecular sequence data from two genes (nuclear ribosomal 28S and mitochondrial cytochrome oxidase I (COI)). Monophyly of *Paradeontacylix* was supported strongly in the Bayesian inference analyses of COI data (Posterior Probability [PP] 95%) and 28S rDNA data (PP 97%). The Aporocotylidae was not monophyletic in the analyses of COI or 28S rDNA data. In the 28S rDNA analysis inclusion of numerous outgroups resulted in two non-aporocotylid taxa (*Clinostomum* spp.) being included with all aporocotylid species, but this relationship was not strongly supported (PP 57%). Following submission of this Chapter for publication, the new species will be formally named and described, type and voucher specimens will be accessioned in museums and sequences will be lodged in GenBank™.

5.2 Introduction

Recent collections of aporocotylids (see Bullard 2009) from marine fishes indicate that there is a diversity of undescribed species (Nolan and Cribb 2006a, b; Bullard 2010). This is undoubtedly true for fish blood flukes in *Paradeontacylix*, of which four new species have been described recently (Hutson and Whittington 2006; Lakshmi and Madhavi 2007; Repullés-Albelda *et al.* 2008). *Paradeontacylix* species can be insidious pathogens and have been associated with mass mortalities of cultivated carangid fish species worldwide (see Bullard and Overstreet 2008 for review). We provide a description of a new species of *Paradeontacylix* from the heart of wild Samson fish *Seriola hippos* Günther, 1876 caught near Rottnest Island, Western Australia. Phylogenetic analyses are provided for several *Paradeontacylix* spp. from specimens obtained in this study and analysed with data available through GenBank™, based on molecular sequence data from two genes (nuclear ribosomal 28S and mitochondrial cytochrome oxidase I (COI)).

5.3 Materials and Methods

5.3.1 Host and parasite collection

Sources of host specimens are listed in Table 41 and their locations are mapped in Figure 17. Tissue from the majority of host individuals examined was stored in 95% AR grade ethanol and accessioned in the Australian Biological Tissue Collection (ABTC) at the South Australian Museum (Table 41). Hosts were sampled from fresh catches made by recreational, commercial and charter fishers. The heart was removed, opened, flushed with saline and the settled contents examined under a dissecting microscope. Other parts of the circulatory system were not examined. Parasite specimens were aspirated by a pipette and killed in almost boiling saline before fixation in 10% formalin or fixed directly in 95% AR grade ethanol for sequencing. Formalin-fixed parasites were placed in distilled water before being stained in Mayer's haematoxylin, then destained in 1% HCl in 70% ethanol. Parasites were dehydrated in a graded ethanol series, cleared in cedar wood oil and mounted on a slide beaneath a coverslip in Canada balsam.

Mounted parasites were studied using a compound microscope and drawings were made with the aid of a drawing tube. Parasite prevalence and mean intensity follows Bush *et al.* (1997). The fork length (L_F) range of parasitised hosts is presented in millimetres (mm), followed in

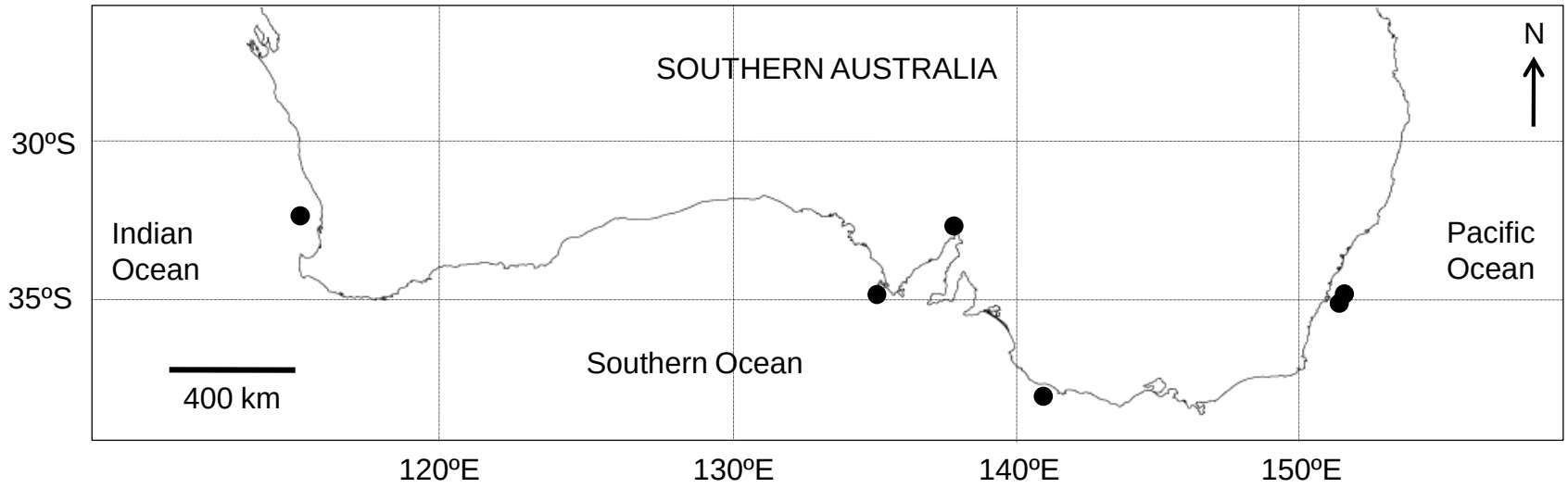

Figure 17. Capture localities for *Seriola* spp. in southern Australian waters. From west to east; Rottnest Island, Western Australia (32º2′36″ 115º26′ 24″E, *S. hippos* and *S. dumerili*); Greenly Island, South Australia (34º38′29″S, 134º47′28″E, *S. hippos*); Port Augusta, South Australia (32º32′41″S, 137º46′45″E, *S. lalandi*); Killarney, Victoria (38°23′36″S, 142°20′24″E, *S. lalandi*); Culburra Cliffs (35°1′5″S, 150°50′52″E, *S. lalandi*) and Sir John Young Banks (34º56′52″S, 150º55′45″E, *S. lalandi*), New South Wales. Greenly Island and Killarney represent localities sampled in a previous study (Hutson and Whittington 2006).

Table 41. *Paradeontacylix* spp. infections in *Seriola dumerili*, *S. hippos* and *S. lalandi* off southern Australia in the Indian Ocean (Western Australia (WA)), the Southern Ocean (South Australia (SA) and Victoria (Vic)) and the Pacific Ocean (New South Wales (NSW)); see also Figure 17). Collections before 2006 represent data from Hutson and Whittington (2006).

Host species	Site	Date	No. hosts examined	ABTC host tissue accession no.	Parasite species	Accession no.
S. dumerili	Rottnest Island, WA	Jan 2009	2	109663-109664	*Paradeontacylix* n. sp.	Not accessioned
					P. cf. *kampachi*	Not accessioned
S. hippos	Greenly Island, SA	Apr 2005	4	102982-102984, 84002	*Paradeontacylix* n. sp.	AHC 28912
					P. sanguinicoloides	AHC 28910
	Rottnest Island, WA	Jan 2007	9	Not accessioned	*Paradeontacylix* n. sp.	Not accessioned
					P. sanguinicoloides	Not accessioned
		Jan 2009	2	109665-109666	To be confirmed*	Not accessioned
S. lalandi	Port Augusta, SA	Sept 2004-Oct 2005	32	108439-108448	*P. godfreyi*	AHC 28903-08; USNPC 097276
		Sept 2007-Sept 2008	18	108449-108465	*P. godfreyi*	Not accessioned
	Kilarney, Vic	Jan 2005	25	82932-82945	*Paradeontacylix* n. sp.	AHC 28911
					P. godfreyi	Not accessioned
	Sir John Young Banks, NSW	June 2003	25	76755-76762, 81991-82003	*P. sanguinicoloides*	AHC 28909
		Feb 2008	131	109649	*Paradeontacylix* n. sp.	Not accessioned
					P. godfreyi	Not accessioned
	Culburra Cliffs, NSW	Jan 2009	14	109650-109666	To be confirmed*	Not accessioned

*Specimens have not been mounted and are maintained as 'wet' preparations in the event that further molecular work is required for publication of this Chapter and/or to make specimens available for future molecular studies.

parentheses by the fork length range of all fish examined and the total number of hosts studied. Measurements of parasite specimens were made using a computerised digitising system similar to that described by Roff and Hopcroft (1986). All measurements are given in micrometres (µm) as the mean followed in parentheses by the range and number of structures measured. Parasite material collected in this study is deposited in the Australian Helminthological Collection (AHC) at the South Australian Museum (SAMA) and the United States National Parasite Collection (USNPC).

5.3.2 DNA preparation, PCR amplification and sequencing

Aporocotylid specimens fixed in ethanol were cut anterior to the caecal bifurcation under a dissecting microscope using a new scalpel blade. The anterior fragment of the worm was returned to ethanol, while the posterior fragment was stained and mounted as described above. DNA was extracted from 22 specimens (including *Paradeontacylix* n. sp., *P. sanguinicoloides*, *P. godfreyi*, *P.* cf. *kampachi* and a *Paradeontacylix* sp. to be confirmed; Table 41) according to the Gentra Kit (Gentra Systems) protocol for animal tissues preserved in ethanol. Extracted DNA was stored in hydration solution at 4 ºC. PCR amplification of partial 28S rDNA and COI sequences was carried out with published primers and additional primers designed using OLIGO 4.0 (Rychlik, 1992) listed in Table 42. For amplification of the 28S rDNA dataset, primer combinations used were G1820/G1821 (approx. 800 bp) and G1842/G1843 (approx. 800 bp). For amplification of the COI dataset the M1174/M1175 primer combination generated a product of approximately 340 bp. Primers used for PCR were also used for sequencing. PCR amplifications were performed in 25 µL reactions using the following cycle conditions: denaturation at 94 ºC for 45 s, annealing at a minimum 50 ºC and maximum 65 ºC (dependent on primers being used) for 45 s and extension at 72 ºC for 1 min; this was repeated for 34 cycles and increased to 38–40 cycles when PCR product yield was low. Each 25 µL PCR contained a final concentration of: 0.5 U AmpliTaq Gold® (5 U/µL), 0.2 µM of each primer, 200 µM of each dNTPs, 2–4 µM $MgCl_2$, 1 x AmpliTaq Gold® buffer. Annealing temperature and $MgCl_2$ concentration were varied to produce optimal amplification. PCR products were cleaned using Agencourt® AMPure® PCR purification kit and were cycle sequenced using the BigDye Terminator v3.1 cycle-sequencing kit (Applied Biosystems). The cycling protocol consisted of 25 cycles of denaturation at 96

ºC for 30 s, annealing at 50 ºC for 15 s and extension at 60 ºC for 4 min. All samples were sequenced on an Applied Biosystems 3730 DNA sequencer.

Table 42. Primers used for PCR amplification.

Gene	Primer ID	Sequence (5' – 3')	Forward / Reverse	Source
28S rRNA	G1820	GATTCCCTTAGTAACGGCGA	F	Chen *et al.* 2007
	G1821	TCGGCAGTGAGTTGTTACAC	R	Chen *et al.* 2007
	G1842	CTTAGTAACGGCGAGTGAACAGGGATGAGC	F	Elizabeth Perkins
	G1843	CGGCAGTGAGTTGTTACACACTCCTTAGCG	R	Elizabeth Perkins
COI	M1174	TTTTTTGGGCATCCTGAGGTTTAT	F	Repulles-Albelda *et al.* 2008
	M1175	TAAAGAAAGAACATAATGAAAATG	R	Repulles-Albelda *et al.* 2008

5.3.3 Phylogenetic analyses

Sequence chromatograms were edited using SeqEd version 1.0.3 and aligned initially using Clustal X (Thompson *et al.*, 1997). Adjustments to alignments were made manually in SeAl version 2.0a11 (Rambaut, 1996) using inferred amino acid sequences where applicable (COI). Monte Carlo Markov Chain (MCMC) Bayesian phylogenetic analyses were run using MrBayes 3.1.1 (Huelsenbeck and Ronquist, 2001). This analysis method allowed the data to be partitioned and optimal models of nucleotide substitution applied to each partition. The model of nucleotide substitution for each gene was assessed using the Akaike Information Criteria (AIC – Akaike, 1985) in ModelTest version 3.7 (Posada and Crandall, 1998). The General Time Reversible (GTR) model with a proportion of invariable sites and a gamma distribution for rates across sites was selected. The final MCMC analyses were run for 10,000,000 generations with a sample frequency of every 100 generations. Tracer v1.4 (Rambaut and Drummond, 2007) was used (to plot the generation number against the log likelihood value) to identify the point at which log likelihood values became stable and all trees generated before this point were discarded. A 50% majority rule consensus tree of the remaining trees was computed.

In analyses of COI data, newly generated sequences for *Paradeontacylix* spp. collected in the present study were aligned and analysed with aporocotylid sequence data on GenBank™ and outgroup taxa selected from other basal trematodes in the Strigeidae and

Schistosomatoidea. In analyses of 28S rDNA data, newly generated sequences for *Paradeontacylix* spp. collected in the present study were aligned and analysed with aporocotylid sequence data on GenBank™ and numerous available sequences for other basal trematodes in the Brachylaimoidea, Diplostomoidoidea and Schistosomatoidea (order Diplostomida of Olson *et al*. 2003).

5.4 Results

5.4.1 *Paradeontacylix* n. sp.

Aporocotylidae Odhner, 1912
Paradeontacylix McIntosh, 1934
Paradeontacylix n. sp. (Figures 18, 19A)
Synonym: *Paradeontacylix* sp. of Hutson andWhittington (2006)
Type-host: *Seriola hippos* Günther, 1876 (Carangidae).
Other hosts: *Seriola dumerili* (Risso, 1810); *Seriola lalandi* Valenciennes, 1833.
Type-locality: off Rottnest Island, offshore from Perth, Western Australia (32º2′36″S 115º26′ 24″E).
Other localities: Sir John Young Banks, New South Wales (34º56′52″S, 150º55′45″E, *S. lalandi*). Localities of previously deposited material (as *Paradeontacylix* sp.) from Hutson and Whittington (2006): Greenly Island, offshore from Port Lincoln, South Australia (34º38′29″S, 134º47′28″E, *S. hippos*), SAMA AHC 28912; Killarney, Victoria (38º23′36″S, 142º20’24″E, *S. lalandi*), SAMA AHC 28911.
Site: Heart.
Infection details: to be confirmed (see Table 41).
Previous infection details (as *Paradeontacylix* sp.) in Hutson and Whittington (2006): *S. lalandi*, Killarney, Victoria: number of infected fish = 1; prevalence 4%; intensity 1; host sizes 760 FL (460–790 FL, n = 25) SAMA AHC 28911; *S. hippos*, Greenly Island, South Australia: number of infected fish = 1; prevalence 25%; intensity 1; host sizes 1160 FL (1120–1160 FL, n = 4) SAMA AHC 28912.
Etymology: The species is not named in this report. The species will be named and described formally when this Chapter is published in the scientific literature.
Specimens deposited: Specimens not deposited at time of submission of this report.

5.4.1.1 Description

Paradeontacylix sensu Smith (2002), Figure 18. Description and measurements based on 7 whole mounted, adult specimens, comprising the type series. Body slender, dorsoventrally flattened, 3,177 (2,821–3,657, n = 7) long by 333 (289–397, n =7) wide; approximately 10 times longer than wide, width consistent throughout specimen only narrowing at anterior and posterior extremities. Lateral body margins slender, ribbon–like, bearing numerous transverse rows of marginal tegumental spines spanning entire length of parasite except for anterior extremity. Marginal tegumental spines ventrolateral, arranged in 584 (555–631, n = 10) transverse rows on both sides of body, rows regularly spaced; number of spines per row increasing from anterior and posterior ends towards middle of body, 4-6 anteriorly, 14-15 at level of caecal bifurcation, 3-4 posteriorly. Posteriorly, 12, rarely 13, enlarged tegumental spines, conspicuous, tips in short tight curve, slightly curved distally, arranged in 4 longitudinal rows each comprising 3 spines. Spine length within rows increasing posteriorly; anterior spines 20 (10–35, n = 28) long; centre spines 25 (17–32, n = 28) long; posterior spines 29 (22–37, n = 28) long. Body margin extending posterior to base of elongated spines, bi-lobed, forming wide inverted 'm' shape. Additionally, 0–3 rows of medium–sized spines 12 (5–17, n = 24) long, arranged on either side of large spines. Nerve cords conspicuous, laterally extended; nerve commissure 131 (122–138, n = 4) from anterior end.

Mouth subterminal. Oesophagus narrow anteriorly, widening posteriorly, 1,160 (909–1,729, n = 5) long, ~35% of total body length, surrounded by gland cells from level posterior to nerve commissure to intestinal bifurcation; larger gland cells forming compact mass in posterior portion. Short anterior and elongate posterior intestinal caeca forming distinct H–shape, 775 (487–974, n = 4) from anterior end of body or ~23% of total body length. Left and right anterior caeca equivalent in length, 83 (78–100, n = 6) long extending anterolaterally from midline; right and left posterior caeca equivalent in length 1,356 (1,077–1510, n = 11), ~42% of body length, approximately 16 times longer than anterior caeca, terminating blindly, abutting ovary.

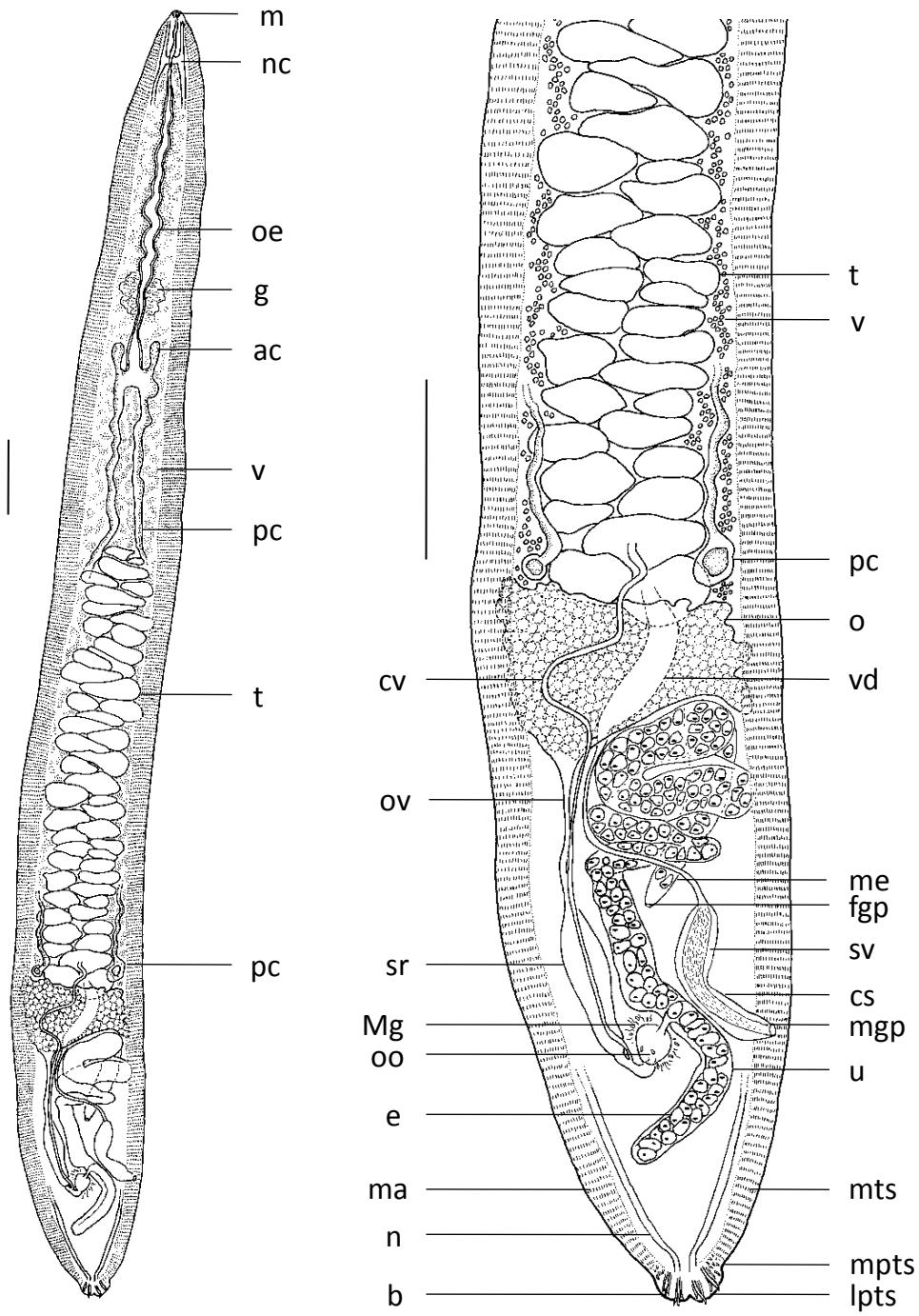

Figure 18. *Paradeontacylix* n. sp. A. Whole adult parasite, composite drawing, ventral view. B. Posterior region, composite drawing, ventral view, showing testicular field and post–ovarian region. *Abbreviations*: ac, anterior caecum; b, bi-lobed posterior margin; cs, cirrus sac; cv, common vitelline duct; e, egg; fgp, female genital pore; g, gland cells; lpts, large posterior tegumental spines; m, mouth; ma, margin; me, metraterm; Mg, Mehlis' gland; mgp, male genital pore; mpts, medium posterior tegumental spines; mts, marginal tegumental spines; n, nerve cord; nc, nerve commissure; o, ovary; oe, oesophagus; oo, oötype; ov, oviduct; pc, posterior extremity of posterior caecum; sr, seminal receptacle; sv, seminal vesicle; t, testis; u, uterus; v, vitellarium; vd, vas deferens. Scale bars = 200µm.

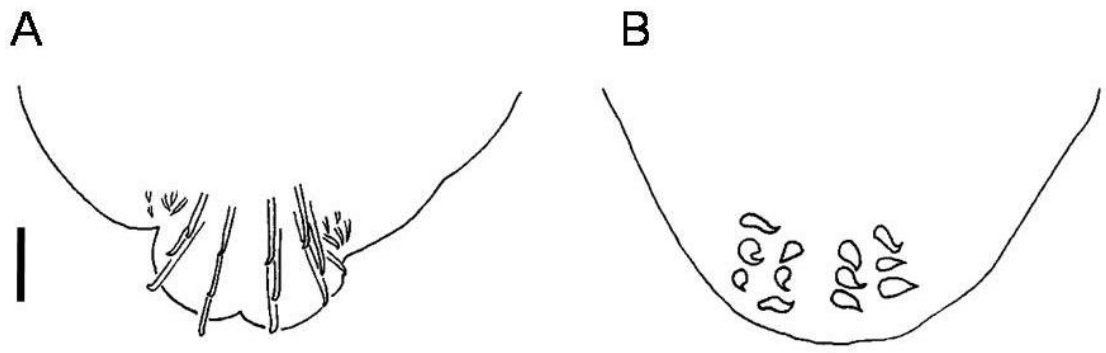

Figure 19. Large posterior tegumental spines of *Paradeontacylix* n. sp. and *P. sanguinicoloides* from southern Australian waters (ventral view). A. *Paradeontacylix* n. sp, showing bi-lobed posterior margin and pointed, slightly curved large posterior tegumental spines arranged in 4 longitudinal rows each comprising 3 spines. B. *P. sanguinicoloides* specimen showing rose-thorn-like posterior tegumental spines arranged in 4 longitudinal rows each comprising 3 spines. Scale bar = 30μm.

Testes 47 (46–48, n = 5), transversely elongate, stacked irregularly forming testicular field, 1-2 testes wide, mostly between posterior extremities of caeca. Testicular field 1,145 (990–1,278, n = 5) long, representing 37 (30–41)% of total body length; posteriorly contiguous to ovary. Vas deferens descending at mid–line from posterior region of testicular field, passing dorsal to ovary, following curved path to form seminal vesicle filling entire cirrus sac. Cirrus cylindrical, male genital pore dorsal, near left body margin, 474 (305–499, n = 5) from posterior end of body. Ovary 159 (116–196, n = 6) long and ~5% of body length, 23 (18–27, n = 6) wide or 72% of body width, located 640 (529–820, n = 6) or ~2% of body length from posterior end of body. Oviduct originating from right side of ovary, passing posteriorly, dilating to form seminal receptacle, 175 (160–221, n = 6) long, 42 (39–49, n = 6) wide. Posteriorly, seminal receptacle narrows, receives common vitelline duct, turns left to join oötype, ovoid, near level of cirrus sac, surrounded by Mehlis' glands. Uterus descending 81 (58–105, n = 4) posterior to oötype, then ascending through several coils filling space immediately posterior to ovary, finally descending to form metraterm. Female genital pore opening dorsally, anterior to male pore, near level of dilated seminal receptacle. Eggs ovoid 20 (14–31, n = 40), measured *in utero*. Vitellarium follicular, follicles extending from nerve commissure posteriorly to anterior margin of ovary. Anteriorly, common vitelline duct noted medianly, just anterior to testicular field, passing posteriorly ventral to testes and ovary, terminating at level of oötype. Excretory vesicle and pore not observed.

5.4.1.2 Remarks

Paradeontacylix n. sp. infected all three *Seriola* spp. at all localities sampled, except for Port Augusta (Table 41; Culburra Cliffs to be determined). The new species is most easily distinguished from other *Paradeontacylix* species by a combination of: (1) a body length of ~2,800–3,650 μm; (2) enlarged posterior tegumental spines; (3) a bi-lobed posterior margin; (4) a maximum number of 15 marginal tegumental spines in rows; (5) 46–48 intercaecal testes; (6) a testicular field occupying 30–41% of the total body length; (7) a portion of the uterus (58–105 μm) descending posterior to the seminal receptacle (Figures 18, 19A).

5.4.2 *Paradeontacylix sanguinicoloides* McIntosh, 1934

Type-host: *Seriola lalandi* Valenciennes, 1833
Other hosts: *S. hippos* Günther, 1876.
Type-locality: Atlantic Ocean off Miami, Florida USA.
Other localities: off Rottnest Island, offshore from Perth, Western Australia, Australia (32º2′36″ 115º26′ 24″E).
Site: Heart.
Infection details: to be confirmed (see Table 41).
Specimens deposited: Specimens not deposited at time of submission of this report.
GenBank™ accession numbers: Not accessioned at time of submission of this report.
Previous records: *S. lalandi*: blood vessels of the gills, Atlantic Ocean, off Miami, Florida, USA (Holotype: USNPC 34329, McIntosh 1934); heart, Sir John Young Banks, New South Wales, Australia (34º56′52″S, 150º55′45″E) (voucher: SAMA AHC 28909, Hutson & Whittington 2006). *S. hippos*, heart, Greenly Island, offshore from Port Lincoln, South Australia (34º38′29″S, 134º47′28″E) (voucher: SAMA AHC 28910, Hutson and Whittington 2006).

5.4.2.1 Remarks

Paradeontacylix sanguinicoloides infected *Seriola lalandi* and *S. hippos* and was present at all locations sampled, except for Port Augusta, SA and Killarney, Victoria (Table 41; Culburra Cliffs to be determined). The additional material collected in this study provides a new locality record for this species in the Indian Ocean. The Australian specimens differ from the single type specimen (collected from the Atlantic Ocean, off Miami,

Florida, USA, Holotype: USNPC 34329 of McIntosh (1934) (damaged specimen)) in that they exhibit a smooth, rounded margin (Figure 19B) whereas a bi-lobed posterior margin was observed in photographs of the type specimen kindly provided by the USNPC. This feature was not included in the original drawing or description by McIntosh (1934). It is possible that Australian specimens of *P. sanguinicoloides* exhibit additional features that may distinguish them from the type, but this cannot be determined until more specimens are collected from the type-host from the type-locality.

5.4.3 *Paradeontacylix godfreyi* Hutson and Whittington 2006

Type-host: *Seriola lalandi* Valenciennes, 1833.
Type-locality: Port Augusta, northern Spencer Gulf, South Australia (32º 42′04″S, 137º46′17″E).
Other localities: present study, Sir John Young Banks, New South Wales (34º56′52″S, 150º55′45″E, *S. lalandi*).
Site: Heart.
Infection details: to be confirmed (see Table 41).
Specimens deposited: Specimens not deposited at time of submission of this report.
GenBank™ accession numbers: Not accessioned at time of submission of this report.
Previous records: Hutson and Whittington 2006, Killarney, Victoria (38º23′36″S, 142º20’24″E, *S. lalandi*)

5.4.3.1 Remarks

Paradeontacylix godfreyi was not found in any specimens of *S. dumerili* or *S. hippos* studied. This species of *Paradeontacylix* infected *S. lalandi* at all localities where this host species was sampled (Figure 17, Table 41; Culburra Cliffs to be determined).

5.4.4 *Paradeontacylix* cf. *kampachi*

Type-host: *Seriola dumerili* (Risso, 1810)
Type-locality: for *Paradeontacylix kampachi* Ogawa and Egusa 1986; Nomi Bay, Japan.
Locality*:* for *Paradeontacylix* cf. *kampachi*, off Rottnest Island, offshore from Perth, Western Australia, Australia (32º2′36″ 115º26′ 24″E) (present study)
Site: Heart.

Infection details: to be confirmed (see Table 41).
Specimens deposited: Specimens not deposited at time of submission of this report.
GenBank™ accession numbers: Not accessioned at time of submission of this report.

5.4.4.1 Remarks

A single specimen was recovered showing strong morphological similarities to *Paradeontacylix kampachi*. This specimen was cut and used in molecular analyses and the posterior section was stained and mounted for morphology. The specimen was not of sufficient quality to enable definitive identification but is designated here as *P*. cf. *kampachi*.

5.4.5 Key to the species of *Paradeontacylix*

1. Body length 1,300–4,200 μm……...2
- Body length 9,800 μm…….*P. odhneri*
2. Posterior tegumental spines small, inconspicuous……3
- Posterior tegumental spines conspicuous, larger than marginal tegumental spines……...5
3. Testes arranged in two or three irregular longitudinal rows, extending laterally to posterior intestinal caeca……..4
- Testes circular, in median longitudinal row, more or less contained within space between posterior intestinal caeca……...*P. megalaspium*
4. Testicular field occupying 19-34% of total body length; male genital pore 440-730 from posterior extremity…….*P. kampachi*
- Testicular field occupying ~22% of total body length; male genital pore 211-401 from posterior extremity……*P. ibericus*
5. Body length 1,300–3,650 μm; posterior caeca terminating blindly close to, or abutting, ovary; testicular field occupying > 25% of total body length; < 18 spines in marginal rows at level of caecal bifurcation…….6
- Body length 3,700–4,200 μm; posterior caeca terminating blindly within testicular field; testicular field occupying < 25% of total body length; ≥ 18 spines in marginal rows at level of caecal bifurcation…….*P. godfreyi*
6. Maximum of 13 tegumental spines in marginal rows; fewer than 35 testes; portion of uterus descending to level of seminal vesicle……...7

- More than 13 tegumental spines in marginal rows; greater than 35 testes; portion of uterus, descending posterior to level of seminal vesicle……..8
7. Body approximately 7 times longer than wide; testicular field occupying 27-38% of total body length; male genital pore 320-550 from posterior extremity…...*P. grandispinus*
- Body approximately 10 times longer than wide; testicular field occupying 34% of total body length; male genital pore 97-193 from posterior extremity……...*P. balericus*
8. Posterior spines increasing in length posteriorly, posterior spines 22-37 μm long…….*Paradeontacylix* n. sp.
- Posterior spines mostly uniform in length, posterior spines 13–19 μm long……...*P. sanguinicoloides*

5.4.5.1 Remarks

The description of *P. odhneri* Layman, 1930 does not provide sufficient detail of important characters for species discrimination (see Layman 1930) and additional material of this species is required to aid a more detailed comparison with other species. *Paradeontacylix sinensis* Liu, 1997, was reassigned to *Psettarium* Goto and Ozaki, 1929 by Holzer *et al.* (2008) and is therefore not included in the key.

5.4.6 Phylogenetic analyses

We present the Bayesian 50% majority rule consensus tree for the COI data in Figure 20 along with posterior probabilities (PP). Monophyly of *Paradeontacylix* was supported strongly (PP 95%). A clade comprising two undescribed aporocotylid (=sanguinicolid) species formed the sister group to *Paradeontacylix* (PP 99%). Due to the exclusion of the aporocotylid *Chimaerohemecus trondheimensis*, an aporocotylid from a chimaera, the Aporocotylidae are not monophyletic based on the taxon representation analysed. Sister to all *Paradeontacylix* spp. and the two undescribed sanguinicolid spp. were species from the Strigeidae (*Apatemon* spp. and *Ichthyocotylurus* spp.). The most important result for the purposes of this study is that a clade of monophyletic *Paradeontacylix* species all of which are likely to be *P. godfreyi*, was strongly supported (PP 100%) using COI data. The remaining *Paradeontacylix* spp. (representatives from the Mediterranean (*P. balearicus* and *P. ibericus*) and one from southern Australia (*P.* cf. *kampachi*)) were distributed in two clades. *Paradeontacylix kampachi* did not form a monophyletic group

with representatives in two different clades and *P*. cf. *kampachi* was also separate in this analysis.

We present the Bayesian 50% majority rule consensus tree for the 28S rDNA data in Figure 21 along with posterior probabilities. Inclusion of many more fish blood fluke taxa and numerous basal trematode outgroups from a variety of vertebrate hosts resulted in two non-aporocotylid taxa (*Clinostomum* spp. [Clinostomatidae]) being included with all aporocotylid species but this relationship was not strongly supported (PP 57%). The sister group to the clades containing aporocotylids and two clinostomatid taxa is a large group containing representatives of diplostomatids, brachylaimids, spirorchiids and some schistosomatids. Important for this study is the discovery that *Paradeontacylix* spp. were monophyletic (Figure 21, PP 97%). Species in another aporocotylid genus, *Cardicola*, were not monophyletic with two representatives, *C. aurata* and *C. forsteri*, separated in the tree.

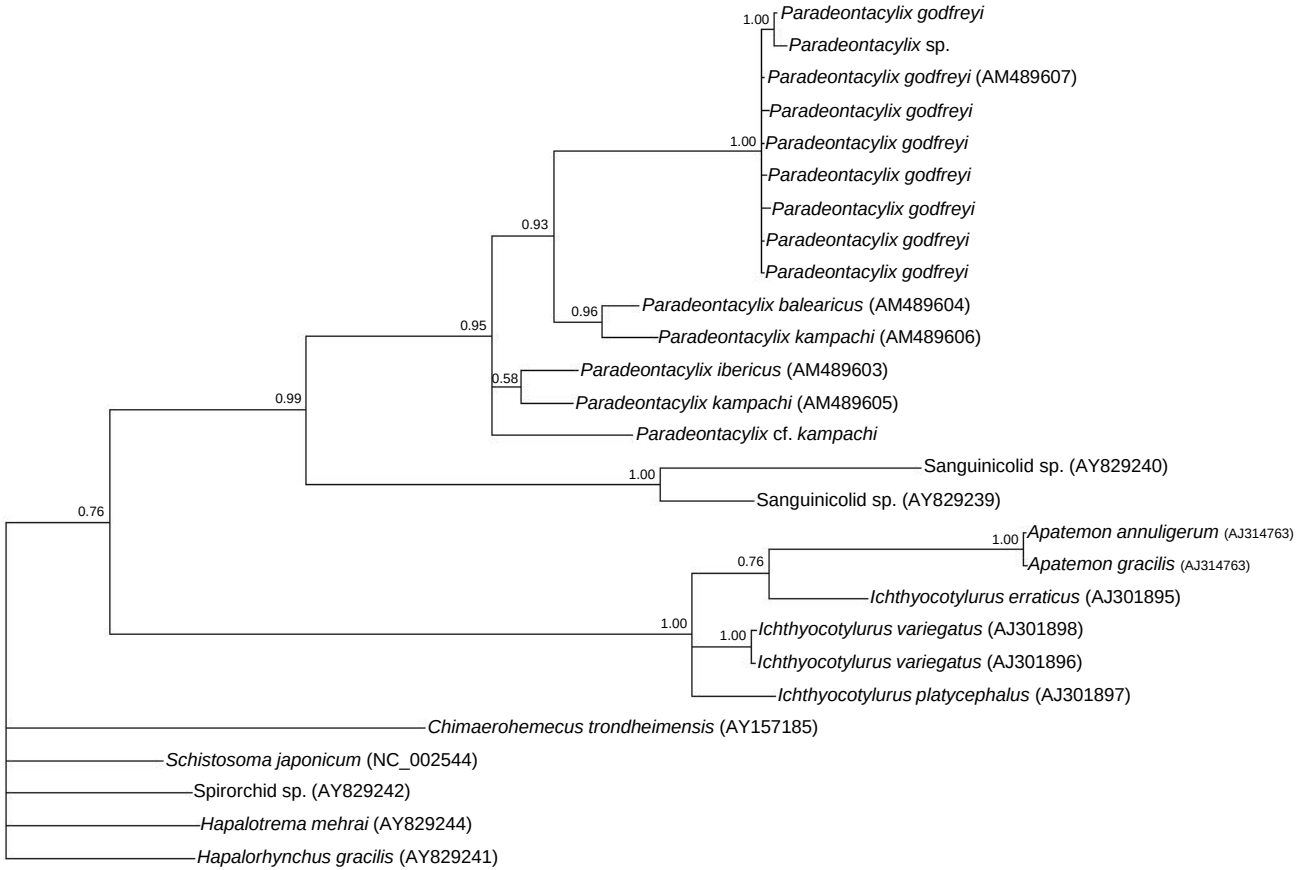

Figure 20. A 50% majority rule consensus tree produced from Bayesian inference analyses of the CO1 data for the Aporocotylidae and outgroup taxa. Posterior probabilities are indicated above each node. The *Paradeontacylix* sp. is a presently unidentified species from *Seriola lalandi* *Paradeontacylix* n. sp. was not sequenced for CO1. GenBank™ accession numbers are shown in brackets for each taxon sequenced previously for CO1. All other taxa were newly sequenced in the present study.

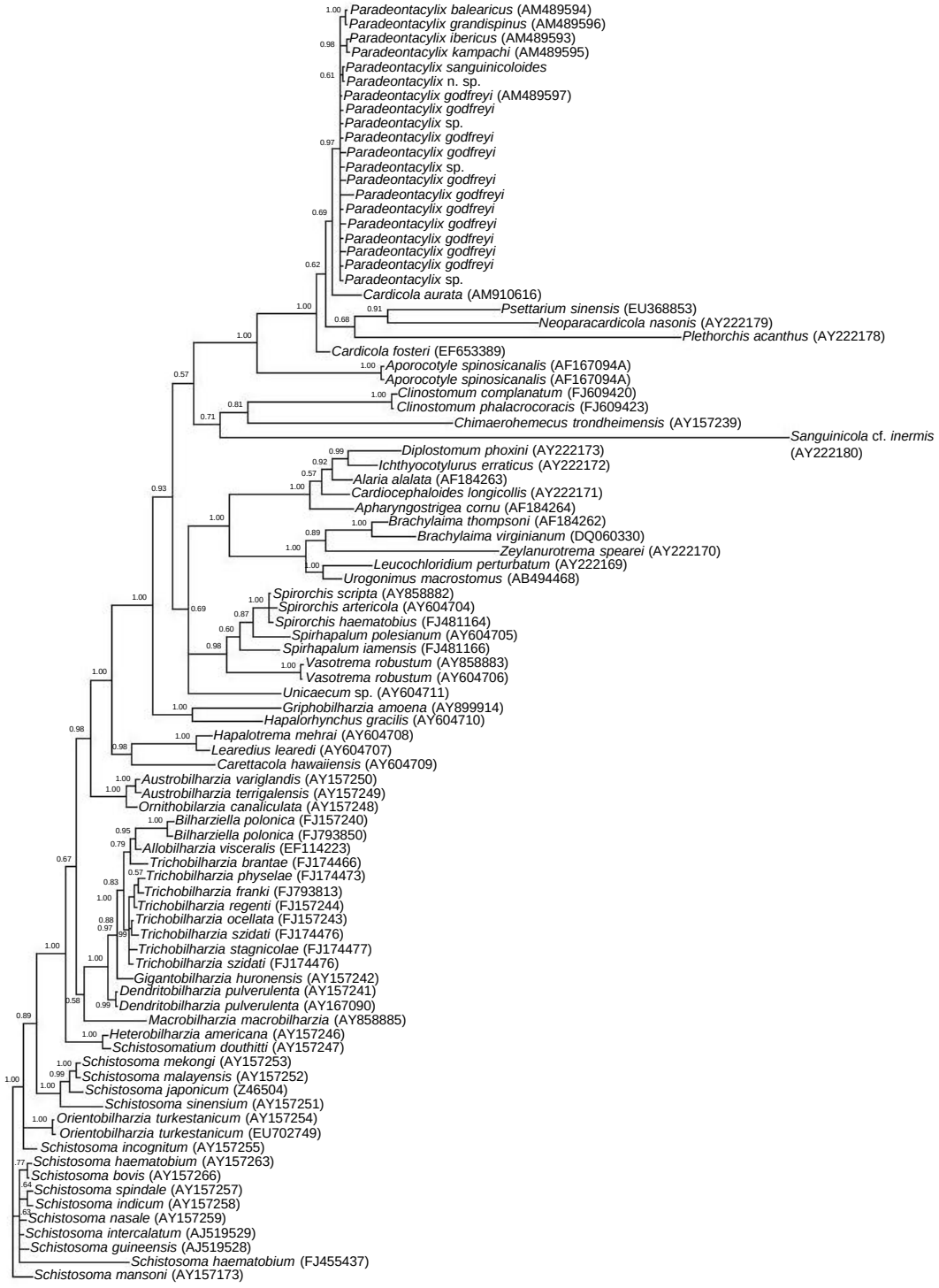

Figure 21. A 50% majority rule consensus tree produced from Bayesian inference analyses of the 28S rDNA data for the Aporocotylidae and numerous outgroup taxa. Posterior probabilities are indicated above each node. The *Paradeontacylix* sp. is a presently unidentified species from *Seriola lalandi*. GenBank™ accession numbers are shown in brackets for each taxon sequenced previously for 28S rDNA. All other taxa were newly sequenced in the present study.

5.5 Discussion

5.5.1 *Paradeontacylix* n. sp.

Paradeontacylix n. sp. differs from other species assigned to *Paradeontacylix* by a combination of: (1) a body length of ~2,800–3,650 μm; (2) enlarged posterior tegumental spines 20-29 μm long; (3) a bi-lobed posterior margin; (4) a maximum number of 15 marginal tegumental spines in rows; (5) 46–48 intercecal testes; (6) a testicular field occupying 30–41% of the total body length and; (7) a considerable portion of the uterus descending posterior to the seminal receptacle. The new species most closely resembles *P. sanguinicoloides* and *P. grandispinus*. *Paradeontacylix* n. sp. and *P. grandispinus* share the same host species (*Seriola dumerili*), possess enlarged posterior spines and a similar testicular field relative to body length. However, *P. grandispinus* has fewer testes (19-32) and the uterus does not descend posterior to the seminal receptacle. *Paradeontacylix* n. sp. and *P. sanguinicoloides* share two host species (*S. hippos* and *S. lalandi*) and exhibit a similar number of testes, extent of testicular field, number of rows of tegumental marginal spines and the maximum number of tegumental marginal spines in rows. The dissimilarities between these two species are largely observed in the size and shape of the posterior spines and the shape of the posterior margin of the body. *Paradeontacylix* n. sp. exhibits elongate posterior spines compared with *P. sanguinicoloides* where the spines are wider at the base and shorter in length (Figure 19). The posterior tegumental spines are more similar in *Paradeontacylix* n. sp. and *P. grandispinus* where the posterior spines measure 22-37 and 21-26 μm in length, respectively.

The new species exhibits a bi-lobed posterior margin which has not been noted previously in *Paradeontacylix*. Although this feature was not observed in Australian *P. sanguinicoloides*, a bi-lobed margin is evident in photographs of the (damaged) single type specimen collected from the Atlantic Ocean, off Miami, Florida, USA (Holotype: USNPC 34329, described by McIntosh 1934). It is possible that Australian specimens of *P. sanguinicoloides* exhibit additional features that may differ from the type, but this cannot be verified until further specimens are collected from the type-host at the type-locality.

5.5.2 Phylogenetic relationships

Aiken *et al.* (2007) hypothesised that platyhelminth parasites of large pelagic fishes are common around the world. Their comparisons of ITS2 and partial 28S rDNA sequence data indicated two *Cardicola* species from multiple scombrid hosts and localities. Some *Seriola* spp. also exhibit worldwide distributions (e.g. *S. dumerili* and *S. lalandi*) but it appears that *Paradeontacylix* species may not necessarily share their host's distribution in the same oceans or hemispheres. Repullés-Albelda *et al.* (2008) proposed two new species of *Paradeontacylix* from *S. dumerili* off Spain, namely *P. ibericus* and *P. balearicus*, which are similar morphologically to *P. kampachi* and *P. grandispinus*, respectively, from the same host species off Japan. They reported low sequence variability for three genes studied for *P. ibericus* and *P. kampachi* (0.2% for 28S; 4.7% for ITS2; 7% for COI) and for *P. balearicus* and *P. grandispinus* (0.2% for 28S; 2.5% for ITS2; 6.3% for COI).

Similarly, *Paradeontacylix* cf. *kampachi* infected Australian *S. dumerili* (Table 41) and forms a sister clade to *P. ibericus* and *P. kampachi* based on COI data (Figure 20). Further sampling of *S. dumerili* in Australia may reveal whether this taxon represents a new species that exhibits minor morphological differences to *P. ibericus* and *P. kampachi*. *Paradeontacylix* n. sp. showed 99% sequence similarity for 28S rDNA data to *P. sanguinicoloides* and 98% similarity to *P. grandispinus*.

Paradeontacylix kampachi did not form a monophyletic group based on COI data with representatives in two different clades (Figure 20; *P. kampachi* AM489606). This is very likely to be the result of an error made when depositing sequence data in GenBank™ . It seems highly probable that data for *P. grandispinus* was incorrectly ascribed to *P. kampachi* (see Repullés-Albelda *et al*. 2008; Holzer *et al.* 2008). This demonstrates the value of sequencing a cut, anterior segment of the parasite and accessioning the posterior section to a museum. The posterior section of *Paradeontacylix* spp. display the most valuable morphological characters for species identification. Alternatively, the lack of monophyly may indicate the presence of a cryptic species or that the authors amplified nuclear copies of mitochondrial DNA containing COI sequence data.

Cardicola spp. were not monophyletic, with two representatives, *C. aurata* and *C. forsteri*, separated in the 28S rDNA tree (Figure 21). Similarly, Holzer *et al.* (2008) also reported that

C. aurata formed a sister clade to *Paradeontacylix* spp. using ITS2 rDNA sequences. In our analyses of available 28S rDNA sequence data, the extensive number of outgroups included represents the significant volume of research on medically important trematodes such as human blood flukes (*Schistosoma* spp.) and some of their relatives that infect reptiles, birds and primates (Schistosomatidae), turtles (Spirorchiidae) plus other basal trematodes representing Diplostomidae, Brachylaimidae and Strigeidae, members of which infect a range of aquatic and terrestrial vertebrate final hosts. By incorporating our new sequence data for several *Paradeontacylix* spp. into a broader sample of taxa, our results support the analysis of Olson *et al*. (2003) because the three trematode families that inhabit the circulatory systems of their final vertebrate hosts, the Aporocotylidae, Schistosomatidae and Spirorchiidae, do not form a monophyletic group unless the Clinostomatidae, a family whose species do not inhabit blood, are included. Of particular significance from our analyses is that the two included clinostomatid species nest within the Aporocotylidae. It is evident that quality specimens are required for morphological and molecular research to examine relationships within *Paradeontacylix* and between fish blood fluke genera.

5.5.3 Coexistence of congeneric species

Modification of the posterior spines and margin morphology in *Paradeontacylix* n. sp. may be a consequence of site specificity in the circulatory system and may account for the coexistence of congeneric species in the same host species. Montero *et al.* (2009) determined that the main habitat for adult *P. ibericus* were the blood vessels supplying the thoracic and pelvic girdle muscles, with parasite intensity gradually decreasing in those sites in the blood system located furthest from the girdles. *Paradeontacylix* spp. presumably anchor to the lining of the host's circulatory system using posterior and marginal spines. Complicating the matter of site specificity is that at least some aporocotylid adults are motile and may relocate within the fish's vascular system (Bullard 2010).

Some fish blood fluke species may infect multiple fish host species and an individual host fish specimen can be infected by multiple congeners (e.g. Hutson and Whittington 2006; Aiken *et al*. 2007; present study, Table 41). The occurrence of the same parasite species in more than one host species can indicate the maintenance of single parasite species, following host speciation, resulting from continued gene flow between parasite populations in different host species. It can also result from host switching, or colonisation of new host species. This

is more likely for parasites that have direct transmission pathways compared with those transmitted through trophic interactions (Cribb *et al.* 2001; Poulin 2007). The co-existence of congeneric species in the same host species may indicate that intra-host speciation through microhabitat selection is driving species richness in fish blood flukes (i.e. sympatric speciation). Puzzlingly, microhabitats of congeneric adult fish blood flukes may overlap in their final fish host (present study; Ogawa *et al*. 1993). Studies of parasitic helminths have shown that congeners in the same host species may not necessarily be sibling species (Littlewood *et al.* 1997; Nolan & Cribb 2006a), indicating this system may be best explained by phylogenetic co-speciation, where parasite radiation parallels that of the host species.

5.5.4 Diagnotic testing capabilities and life cycle elucidation

Exploring phylogenetic relationships of congeners in final hosts and examining patterns of host use in final vertebrate (e.g. Montero *et al.* 2009) and intermediate invertebrate hosts will extend our ability to understand biodiversity and determine whether parasites have radiated with their final fish and/or intermediate invertebrate hosts. Knowledge of parasite life cycles may also infer the sequence, acquisition and loss of life cycle traits (Cribb *et al.* 2003). Diagnostic testing capabilities will be enabled for *Seriola* spp. in Australia when all fish blood fluke sequences from this study have been accessioned in GenBank™ (see Chapter 11 Planned Outcomes). These adult parasite sequences may be matched to asexual parasite stages recovered from invertebrate intermediate hosts. This will facilitate our growing understanding of the specificity of fish blood flukes to their intermediate hosts, the role these invertebrate hosts play in the environment and whether it is feasible to relocate sea-cages to areas where intermediate hosts are absent or less abundant and therefore reduce the severity of aporocotylid infections (Chapter 7).

6 POTENTIAL PARASITE THREATS TO SOUTHERN AUSTRALIAN FINFISH AQUACULTURE

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6.1 Abstract

Parasitic crustaceans are responsible for severe disease outbreaks in finfish aquaculture. We provide the first report of five marine ectoparasitic crustacean species including *Argulus diversicolor* Byrnes, 1985 (Branchiura: Argulidae), *Caligus bonito* Wilson, 1905, *C. longipedis* Bassett-Smith, 1898, *C. pelamydis* Hewitt, 1963 and *C. punctatus* Shiino, 1955 (Copepoda: Caligidae) on wild arripid hosts, *Arripis georgianus* (Valenciennes, 1841), *A. trutta* (Forster, 1801) and *A. truttaceus* (Cuvier, 1829) (Perciformes: Arripidae) in southern Australian waters. *Caligus pelamydis* and *C. punctatus* are new Australian records. All five crustacean species exhibit low host-specificity and *Argulus* spp., *C. longipedis*, *C. pelamydis* and *C. punctatus* have been associated with mass mortalities in cultured fishes outside Australia. Given the propensity for arripids to aggregate at sea-cage aquaculture sites, awareness of these five parasitic crustacean species may allow health managers to identify and anticipate potential outbreaks on southern Australian fish farms.

6.2 Introduction

Sea-cage farms are highly attractive to wild fishes and attract large and diverse aggregations in their immediate vicinity. Increased availability of food due to waste feed and faeces from caged fish (Fernandez-Jover *et al.* 2008) and their use as a refuge from predators (Castro *et al.* 2002), may explain the attraction of fish to floating structures. Aggregations of wild fish at individual farms have been documented in warm temperate (Mediterranean Sea; Dempster *et al.* 2002) and cool temperate (Norway; Dempster *et al.* 2009, Australia; Dempster and Kingsford 2004a) environments. Many parasite taxa may be shared between farmed and wild fishes. Parasites can have severe impacts on farmed fish production, while high densities of farmed fish may elevate parasite prevalence and intensity in surrounding wild populations, as

has been observed for sea lice infestations of wild salmonids in the northern hemisphere (e.g. MacKenzie and Abaunza 1998; Morton *et al.* 2005; Krkošek *et al.* 2007).

Parasitic crustaceans are widely recognised to cause serious problems in the culture of many fish species (see Johnson *et al.* 2004a for review). They are likely to establish and proliferate in aquaculture because most have direct life-cycles consisting of free-living, free-swimming and attached parasitic stages. They also may reproduce rapidly and directly reinfect their hosts. Parasitic crustaceans may also act as vectors of bacterial and viral infections as well as protozoans and metazoans (e.g. Overstreet *et al.* 2009). Research is required to identify sources of parasitic crustaceans, to recognize taxa with low host-specificity and to document potentially harmful species.

In Australia, fish farming is intensifying in Spencer Gulf, South Australia (SA), where southern bluefin tuna *Thunnus maccoyii* (Castelnau, 1872), yellowtail kingfish *Seriola lalandi* Valenciennes, 1833 and mulloway *Argyrosomus japonicus* (Temminck & Schlegel, 1843) are farmed in sea-cages. Fish species in the Arripidae have a tendency to aggregate near structure (Dempster and Kingsford 2004a; Neira 2005b) and have been observed in, and around, sea-cages in SA (KSH pers. obs.). The Arripidae comprises a single genus *Arripis* (Jenyns) which includes four species. Three species comprise important commercial and recreational fisheries in southern Australian waters; Australian herring or tommy ruff *Arripis georgianus*; eastern Australian salmon *A. trutta*; and western Australian salmon *A. truttaceus*. *Arripis georgianus* and *A. truttaceus* share similar distributions, found in temperate waters from Western Australia (WA) through to New South Wales, and around Tasmania (Gomon *et al.* 2008). *Arripis trutta* overlaps in distribution with these two species in waters off South Australia, Victoria, New South Wales and Tasmania, but is also found off the coast of NZ (Smith *et al.* 2008).

In a broad parasitological survey of the Arripidae in southern Australian waters, we made a remarkable discovery of five species of parasitic crustaceans not previously known from arripid fishes, including two new Australian parasite records. The majority of crustacean parasite species reported here have been associated with mass mortalities in aquaculture overseas. This study documents five new parasite-host records from three species of *Arripis* in southern Australian waters, reviews associated problems with these parasite species

elsewhere and draws attention to the potential threat of these species for southern Australian finfish aquaculture.

6.3 Materials and Methods

A total of 183 *Arripis georgianus* (mean [range] = 179 [154–220] mm fork length (FL)) and 67 *A. truttaceus* (350 [185–601] mm FL) obtained from Gulf St Vincent, Spencer Gulf, northern Kangaroo Island and Coffin Bay in SA waters was examined for ectoparasites (Figure 22). Nineteen *A. trutta* (483 [400–545] mm FL) were examined from Bermagui, New South Wales (NSW) (Figure 22). Fish were identified using the key by Gomon *et al.* (2008). Fish were collected by line fishing or from fish markets, namely Safcol fish market, Mile End, Adelaide, SA, and the Sydney Fish Market, Pyrmont, NSW (University of Adelaide, Animal Ethics Committee Science project S-098-2007). Tissue samples were collected from each fish, stored in undenatured ethanol and lodged with the Australian Biological Tissue Collection (ABTC 108509–108777) at the South Australian Museum (SAMA), North Terrace, Adelaide, 5000, SA.

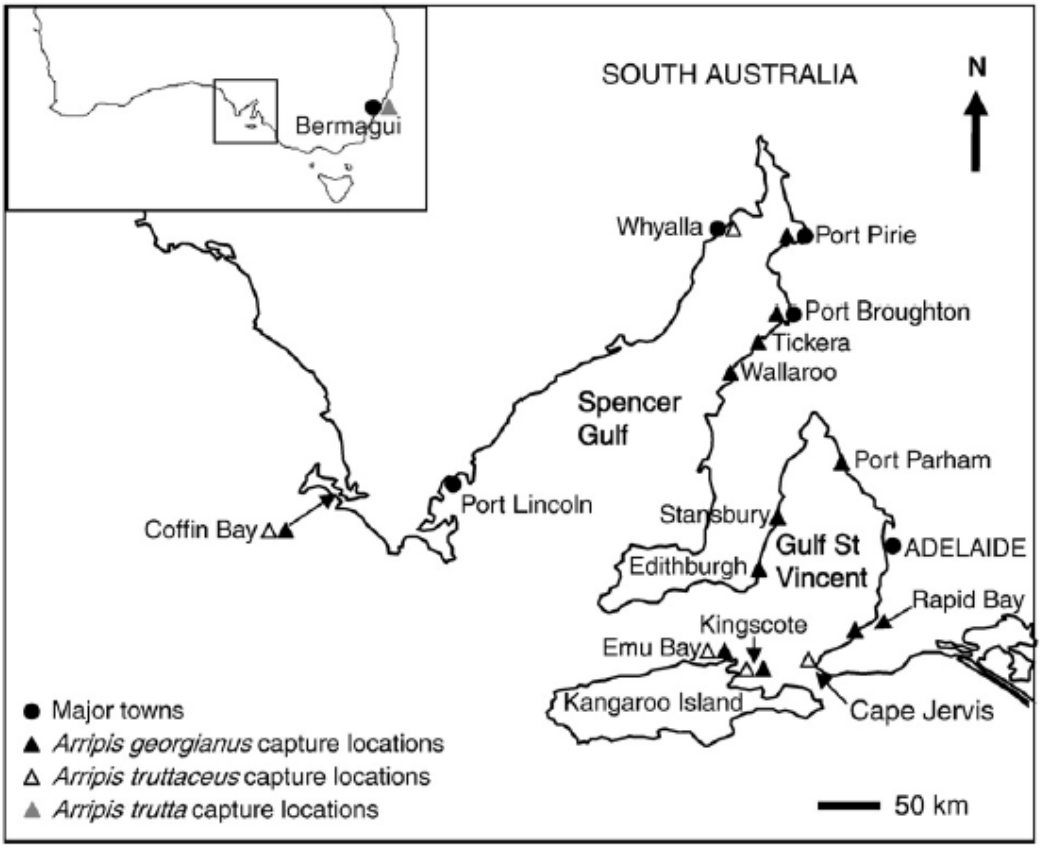

Figure 22. Collection localities for three Arripis spp. in southern Australian waters and one site off the coast of New South Wales.

Fish were examined for ectoparasites with the naked eye on all exterior surfaces, including branchiostegal membranes, fins, fin sulcus, mouth and operculum. The gills were removed, placed in seawater and examined under a dissecting microscope for ectoparasites. Parasitic crustaceans were removed with fine forceps, fixed in 70% ethanol and cleared in lactophenol prior to morphological examination using a compound microscope. All parasitic crustaceans were identified from their original published descriptions, redescriptions and reference to information in Ho and Lin (2004). Identifications were confirmed by Prof Geoff Boxshall, Natural History Museum, London (NHM). Voucher specimens were lodged in the SAMA Crustacean collection (SAMA C) and at the NHM.

6.4 Results and Discussion

Five species of parasitic crustaceans were recorded from arripid hosts; *Argulus diversicolor* (Branchiura: Argulidae), *Caligus bonito*, *C. longipedis*, *C. pelamydis* and *C. punctatus* (Copepoda: Caligidae). Microhabitat, host species, host localities and museum accession numbers are shown in Table 43. This is the second record of *A. diversicolor* which was originally described from the sparid *Acanthopagrus latus* (Houttuyn, 1782) from Point Samson, Western Australia (Byrnes 1985). Although *C. pelamydis* and *C. punctatus* are cosmopolitan caligid species (see Hewitt 1963; Ho and Lin 2004), this is the first time they have been recorded in Australian waters.

Argulus spp. infect fish hosts distributed on and around all continents, except Antarctica, in marine and estuarine (n=44) and freshwater (n=85) habitats (Poly 2008). *Argulus* spp. are problematic ectoparasites in freshwater fish culture and several epizootics have occurred in wild freshwater fisheries (e.g. Menezes *et al.* 1990; Hakalahti-Siren *et al.* 2008; Hewlett *et al.* 2009). *Argulus japonicus* (Thiele, 1900) has been introduced all over the world, primarily due to stockings of goldfish *Carassius auratus* (Linnaeus, 1758) and carp *Cyprinus carpio* Linnaeus, 1758 but may also parasitize brown trout *Salmo trutta* Linnaeus, 1758, as well as stickleback *Gasterosteus aculeatus* Linnaeus, 1758, roach *Rutilus rutilus* (Linnaeus, 1758), perch *Perca fluviatilis* Linnaeus, 1758, tench *Tinca tinca* (Linnaeus, 1758), pike *Esox lucius* Linnaeus, 1758 and bream *Abramis brama* (Linnaeus, 1758**)** (see Poly 2008). *Argulus* species have also been reported from marine fish farming facilities in Chile, Canada and Norway and can cause mortality in farmed salmonid stocks (Boxshall 2005; Schram *et al.* 2005).

Table 43. Parasitic crustaceans of *Arripis georgianus*, *A. trutta* and *A. truttaceus* from southern Australian waters, indicating microhabitat, host locality and museum accession number.

Parasite	Microhabitat	Host species	Locality	Accession no.
Branchiura				
Argulidae				
Argulus diversicolor	Skin	[b]*A. georgianus*	[c]SA	NHM 2009.261
		[b]*A. truttaceus*	[c]SA	SAMA C6823
Copepoda				
Caligidae				
Caligus bonito	Gills	[b]*A. trutta*	NSW	SAMA C6821
		[b]*A. truttaceus*	[c]SA	SAMA C6819
Caligus longipedis	Gills	[b]*A. truttaceus*	[c]SA	SAMA C6815-16
[a]*Caligus pelamydis*	Gills	*A. trutta*	[c]NSW	SAMA C6822
[a]*Caligus punctatus*	Skin	[b]*A. georgianus*	[c]SA	SAMA C6814
		[b]*A. trutta*	[c]NSW	SAMA C6820
		[b]*A. truttaceus*	[c]SA	SAMA C6817-18

[a]=new Australian record; [b]=new host record; [c]=new state locality record; SA=South Australia; NSW=New South Wales.

Argulids attach to fish skin by means of suckers and spines and feed on blood and external tissues (Boxshall 2005). They are capable of changing their position on their host as well as changing their host individual (Pasternak *et al.* 2000; Pasternak *et al.* 2004), and may enhance severity of bacterial infections (Cusack and Cone 1986; Bandilla *et al.* 2006). Although there is no published record of *A. diversicolor* from farmed fishes in SA, a photograph of this species (or a similar species) is presented on the cover of an illustrated guide to the parasites of wild and captive southern bluefin tuna *Thunnus maccoyii* (Scombridae) from SA (Rough 2000). However, there is no further reference to the cover image or to an *Argulus* species in the publication. The presence of *A. diversicolor* on arripids is a cause for concern for farmed southern Australian fishes given the likely similarities in pathology to other *Argulus* spp. and its report from two fish families (i.e. Arripidae and Sparidae).

Of the four caligid copepod species identified here, three are recognised as being problematic in aquaculture: *Caligus longipedis, C. pelamydis* and *C. punctatus*. *Caligus longipedis*, which infected *A. truttaceus* in SA, can cause bruising to the body of farmed striped jack *Pseudocaranx dentex* (Bloch & Schneider, 1801) (Carangidae) in Japan and make it unmarketable (Ogawa 1992). Also, Madinabeitia *et al.* (2009) demonstrate that *C. longipedis*

could act as a potential vector for the transmission of the bacterium *Lactococcus garvieae*, which has caused severe mortalities to *Seriola* spp. (Carangidae) and *P. dentex* in Japan. Although *C. longipedis* primarily infects carangids, it has been reported previously from 18 fish species representing 10 fish families including acanthurids, carangids, haemulids, monacanthids, ostraciids, paralichthyids, pomacanthids, scarids, serranids and trichiurids (see Ho and Lin 2004). We provide the second record of this species in Australian waters. *Caligus longipedis* was documented previously by Heegaard (1962) in NSW (as *C. lucidus*; verified by Ho and Lin 2004) from the Chinaman-leatherjacket *Nelusetta ayraud* (Quoy & Gaimard, 1824) (Monacanthidae) (as *Cantherhines ayraud*).

Caligus pelamydis, documented on *A. trutta* in this study, has been associated with mass mortalities of cultured Japanese sea perch *Lateolabrax japonicus* (Cuvier, 1828) (Lateolabracidae) in South Korea (Choi *et al.* 1995). This widely distributed species has been reported from seven fish species in four families (gempylids, lateolabracids, sciaenids and scombrids) from New Zealand, the Mediterranean, the British Isles, the east coast of the United States and South Africa (see Hewitt 1963). This is the first time this species has been recorded in Australian waters.

Caligus punctatus, which we documented on all three arripid species, has caused mass mortality of numerous cultured fishes in Taiwan including carangids, charids, cichlids, lateolabracids, latids, mugilids, serranids, sparids, and terapontids (see Ho and Lin 2004). This parasite species is particularly problematic due to its low host-specificity (21 species in 14 families) and its ability to survive in the plankton (Ho and Lin 2004). This species is known from Taiwanese, Japanese and Korean waters and is reported here from Australian waters for the first time.

Caligus bonito is a cosmopolitan caligid that primarily infects scombrids (10 species) but has also been recorded from a carangid, coryphaenid, mugilid, pomatomid, serranid and two lutjanid species (see Ho and Lin 2004). In Australia, it has been previously reported from three wild scombrids including *Euthynnus affinis* (Cantor, 1849) and *Sarda australis* (Macleay, 1881) in NSW and *E. alleteratus* (Rafinesque, 1810) from Queensland (reported as *C. kuroshio* by Kabata (1965), verified by Ho and Lin 2004). There is no recorded pathology associated with *C. bonito* species in the literature.

Although the parasite species identified in this study have not been recorded on farmed Australian fishes to date, an epizootic of *Caligus chiastos* Lin & Ho, 2003 was recently documented on farmed southern bluefin tuna *Thunnus maccoyii* (Scombridae) in southern Spencer Gulf, SA (Hayward *et al.* 2008). Infestations by *C. chiastos* were associated with eye damage of farmed *T. maccoyii* and peaks in infection occurred in summer months (Hayward *et al.* 2009b). *Caligus chiastos* has also been detected on the gills of farmed *Argyrosomus japonicus* (Sciaenidae) and *Seriola lalandi* in the same region (Hayward *et al.* 2007a). As no attached chalimus stages of any caligids have been detected on *T. maccoyii*, Hayward *et al.* (2008) suggested that *C. chiastos* infections are transmitted to tuna from other species of infected wild fish that are attracted to tuna cages. Indeed, the only previous record of *C. chiastos* in Australia is on wild snapper *Chrysophrys auratus* (Forster 1801) (Sparidae) from eastern Australia (Ho and Lin 2004), a fish species that is known to associate with sea-cage sites in SA (KSH pers. obs.).

Given the low degree of host-specificity exhibited by parasitic crustaceans detected in this study and the tendency for arripids to aggregate at sea-cage farms, it is highly likely that, in suitable conditions, the argulid and caligid species reported here may establish and proliferate in farmed fish populations in southern Australia.

6.5 Conclusion

Discovery and documentation of parasite fauna of wild and farmed fish should be incorporated into an ongoing sampling program for effective parasite management and risk assessment. Effective mitigation of parasite species infecting fishes in sea-cage farms can only be achieved through reliable parasite identification, knowledge of their biology and assessment of appropriate management methods. Recognition of parasite species that may decrease profitability through mortality, morbidity and reduced marketability of stocks is crucial. Considering the previous history of *Argulus* spp., *Caligus longipedis, C. pelamydis* and *C. punctatus* in finfish farming overseas, these parasites are of immediate priority for research into their biological attributes. This will enable development of the most suitable management strategies to avoid outbreaks in southern Australian aquaculture.

7 ASSESSING PARASITE RISK IN FINFISH AQUACULTURE AND IDENTIFYING APPROPRIATE HUSBANDRY

7.0 Parasite risk assessment for sea-cage aquaculture of mulloway (*Argyrosomus japonicus*) and barramundi (*Lates calcarifer*)

7.1 Abstract

Reliable parasite identification and assessment of the risk that parasite species present is a vital pre-requisite for effective management of parasites in sea-cage farms. A qualitative risk assessment for parasite species known to infect mulloway (*Argyrosomus japonicus*) and barramundi (*Lates calcarifer*) was undertaken to recognise parasite species that present serious risks for fish farmed in sea-cages and their wild conspecifics. Risk was estimated by considering the likelihood and consequence of parasite establishment and proliferation in sea-caged stock. The monogenean *Benedenia sciaenae* was considered to present a high risk to *A. japonicus* while two monogenean species, *Neobenedenia melleni* and *B. epinepheli,* were considered a high risk to *L. calcarifer*. Future research on the life cycle parameters of moderate to high risk parasite species under various environmental conditions will enable identification of management strategies that can break life cycles and minimise the potential for outbreaks appropriate to the biology of the parasite species.

7.2 Introduction

The early 1990s marked the expansion of Australia's aquaculture sector. This was underpinned by innovation in ranching southern bluefin tuna (*Thunnus maccoyii*), growth in existing aquaculture industries and significant development of new finfish industries including mulloway (*Argyrosomus japonicus*), barramundi (*Lates calcarifer*) and yellowtail kingfish (*Seriola lalandi*). These three species are grown from fertilised eggs in land-based hatcheries where, through standard biosecurity practices, fish are usually isolated from infection by parasites. Wild marine organisms and infected farmed fish may act as an initial source of parasite infection when fingerlings are transferred from the hatchery to sea-cages for grow out. The natural occurrence of wild conspecifics and other wild organisms at locations where fish are farmed provides an opportunity for transfer of host-specific and generalist parasites from wild to farmed populations. The detailed parasite survey and review

of published parasite records in Chapter 2 indicate that more than 26, 32 and 56 parasite species can infect *A. japonicus* (Table 12), *L. calcarifer* (Table 10) and *S. lalandi* (Table 6), respectively. While parasite surveys and risk assessments for sea-cage aquaculture of *S. lalandi* and *T. maccoyii* have been made previously (Nowak 2004; Deveney *et al.* 2005; Hutson *et al.* 2007a; Hutson *et al.* 2007b) parasite risks for sea-cage aquaculture of *A. japonicus* and *L. calcarifer* have not been assessed.

Aquaculture of *A. japonicus* in Australia began in 1992 with successful production of fish under hatchery conditions (Battaglene and Talbot 2004). In recent years, production has steadily increased. Sea-cage farms in South Australia and Western Australia (and until recently New South Wales) and pond culture on the east coast of Australia produce small commercial quantities, with production of over 600 t in 2006/07 (valued at ~AUD$4.9 million) (O'Sullivan and Savage 2009). In a global context, production of sciaenids in 2007 was >115,000 t valued at over USD$156 million. In terms of the volume and value of global production, Australia's sciaenid production is third behind China and the USA (FAO 2008; O'Sullivan and Savage 2009).

Aquaculture of *L. calcarifer* constitutes the third most valued finfish industry in Australia after Atlantic salmon (*Salmo salar*) and southern bluefin tuna (*T. maccoyii*). *Lates calcarifer* aquaculture commenced in north Queensland with a hatchery operation in 1982 and spread to terrestrial ponds and/or tank operations in all mainland states. Sea-cage aquaculture occurs in Queensland, Northern Territory and Western Australia. In 2006/07 >3,000 t was produced, valued at approximately AUD$26.8 million (O'Sullivan and Savage 2009).

Argyrosomus japonicus and *L. calcarifer* are susceptible to a number of parasite species which present considerable concerns for economical sea-cage farming (Deveney *et al.* 2001; Hayward *et al.* 2007b). Intensive sea-cage aquaculture may also amplify pathogens in the environment and is a concern for the health of wild fishes that may associate with, or move past, farm sites (e.g. McKibben and Hay 2004; Krkošek *et al.* 2005). According to the Standards Australia/Standard New Zealand (2004) for risk management, risk is 'the chance of something happening that will have an impact on objectives'. For aquaculture and wild fisheries, risk generally applies to the potential impact on long-term sustainability (Fletcher *et al.* 2004). Therefore, a risk assessment is needed that accommodates identification of

parasite species 1) that are likely to establish and proliferate in aquaculture and 2) that may decrease host profitability through reduced marketability, morbidity and mortality.

The aim of this study was to assess the risks to sea-cage aquaculture of *A. japonicus* and *L. calcarifer* posed by parasites reported from these species in Australia and from overseas. This risk assessment identifies: 1) the most likely parasite species to establish and proliferate in sea-cage aquaculture, 2) parasite species with potentially negative consequences and 3) the overall risk of the likelihood and consequence of parasite establishment. Husbandry practices that may be useful to manage infection by moderate and high risk parasite species are reviewed.

7.3 Materials and Methods

7.3.1 Fish and parasite collection

Table 44 indicates sample locations for wild and farmed fishes collected during this study. Methods for parasite sampling were described previously (Chapter 2, page 12).

Table 44. Sample locations for wild and farmed *Argyrosomus japonicus* and farmed *Lates calcarifer* surveyed in this study.

Species	Origin	Location	Latitude and longitude	*n*	L_F range (mm)
A. japonicus	wild	Port Adelaide, SA	34°45′49″S 138°29′45″E	2	990 & 1500
	sea-cage	Botany Bay, NSW	34°0′10″S 151°12′32″E	14	340 - 370
L. calcarifer	sea-cage	Port Hinchinbrook, QLD	18°23′31″S 146°8′12″E	26	515 - 650

7.3.2 Risk analyses

Several risk analyses provide frameworks to identify hazards and quantify risks posed by metazoan parasites in aquaculture (Diggles 2003; Fletcher *et al.* 2004; Nowak 2004; Murray and Peeler 2005; Hutson *et al.* 2007b). We devised an assessment to determine the likelihood of parasite establishment in farmed fish by considering the biological pathway necessary for parasite species to infect farmed fish species. A framework was developed to assess the potential negative consequence of establishment and proliferation of parasite species. This risk assessment estimated two parameters, likelihood and consequence, that were considered independently. Likelihood and consequence estimates were then combined in a risk matrix to determine overall risk. Risk was estimated for each parasite species identified from our

survey and for parasites from previously published records for these two fish species in the scientific literature.

7.3.3 Likelihood

The likelihood of parasite establishment and proliferation on or in *A. japonicus* and *L. calcarifer* cultured in sea-cages was estimated by reference to the biological pathways necessary for each parasite species to infect and reproduce on the farmed species. Five likelihood criteria were used to estimate the likelihood of parasite establishment and proliferation based on parasite biology. Likelihood definitions ranged from negligible to extreme (based on Fletcher *et al.* 2004; Table 45).

7.3.4 Consequence

Information was gathered from scientific literature on parasites (genus and/or species if available) to indicate any potential negative **consequence** of establishment and proliferation. Data are sought that directly related to four risk criteria including: 1) potential to cause mortality, 2) potential to cause pathology or disease, 3) potential impact on marketability and 4) potential impact on consumer health. Parasites were scored for each of these four criteria (Tables 47, 49). Parasites that met all four criteria were assigned an ***extreme*** consequence, parasites that met three criteria were assigned a ***high*** consequence, parasites that met two criteria were assigned a ***moderate*** consequence, parasites that met one criterion were assigned a ***low*** consequence and parasites that met no criteria were assigned a ***negligible*** consequence. Using this information, we determined the consequence of parasite establishment and proliferation as adapted from risk criteria by Fletcher *et al.* 2004 (Table 45). Note that risk criteria proposed by Fletcher *et al.* (2004) do not address human zoonoses.

7.3.5 Overall risk

In order to identify the most serious parasite risks for sea-cage aquaculture of *A. japonicus* and *L. calcarifer* (Table 48 & 50), total risk was estimated by considering the likelihood and consequence of parasite establishment and proliferation in sea-cage farming following the Australian Quarrantine Inspection Service (AQIS) (AQIS 1999) risk estimation matrix (Table 46).

Table 45. Definitions for likelihood and consequence (based on Fletcher *et al.* 2004).

Likelihood level	Descriptor
Extreme	Parasites with direct infective stages that have been recorded on aquaculture fish
High	Parasites with direct life cycles not previously recorded in sea-cage culture
Moderate	Parasites with complex life cycles that exhibit direct infective stages
Low	Parasites with two or more host species in the life cycle, that require the primary host to consume an infected intermediate host
Negligible	Parasites that have not been previously recorded from the host fish species
Consequence and risk level	
Extreme	Catastrophic consequences to the entire industry, total mortality or eradication of fish is considered, trade implications at the national level
High	Establishment and proliferation of parasites could have serious biological consequences, prolonged high mortality rates, enterprise survival is questioned, significant economic concern to the industry
Moderate	Substantial seasonal morbidity and mortality rates with significant cost to the farmer to warrant intermittent concern by the industry
Low	Establishment and proliferation of parasite species is manageable with low economic significance to the industry
Negligible	Establishment and proliferation of parasite species could have no significant consequences, with low or no measurable economic effect at the enterprise level

Table 46. Risk estimation matrix following the Australian Quarantine and Inspection Service (AQIS 1999).

	Consequence (2)				
Likelihood (1)	**Negligible**	**Low**	**Moderate**	**High**	**Extreme**
Extreme	Negligible	Low	Moderate	High	Extreme
High	Negligible	Low	Moderate	High	High
Moderate	Negligible	Negligible	Low	Moderate	High
Low	Negligible	Negligible	Negligible	Low	Moderate
Negligible	Negligible	Negligible	Negligible	Negligible	Low

7.4 Results

7.4.1 Parasite species

Parasites detected on wild and farmed *A. japonicus* and farmed *L. calcarifer* in this survey and from previously published records are shown in Chapter 2 (Tables 12 and 10, respectively). Care should be taken in the interpretation of these results where few fish were sampled. Low sample sizes can severely underestimate parasite diversity, especially for parasites that exhibit low prevalence. Nevertheless, this risk assessment is based on all current data available, not only from fish examined during this study but from the wider literature.

Some parasites we found could not be identified to species, a result of a combination of factors including limited number of parasite specimens, potentially undescribed parasite species and inability to identify larval parasite life stages definitively. We detected cysts in the muscle tissue of wild *A. japonicus* but they did not contain any cestode larva (Chapter 2, Table 12). It is likely that the cysts were produced by *Poecilancistrium caryophyllum* which has been recorded previously from this microhabitat in wild *A. japonicus* (see Robinson 1965).

7.4.2 Risk assessment for sea-cage farming of Argyrosomus japonicus

Most monogenean species, two copepod species and one isopod species presented an extreme likelihood of establishment and proliferation (Table 48). The consequence of parasite establishment and proliferation ranged from negligible to high (Table 47). No taxon was classed as an extreme risk. *Benedenia sciaenae* was determined to present the highest risk for *A. japonicus* farmed in sea-cages and for wild conspecifics nearby (Table 48).

7.4.3 Risk assessment for sea-cage farming of Lates calcarifer

All protozoan, monogenean, aporocotylid trematodes, copepod and isopod species presented an extreme likelihood of establishment and proliferation (Table 50). The consequence of parasite establishment and proliferation ranged from negligible to high (Table 49). No taxon was classed as an extreme risk. Two species (*Neobenedenia melleni* and *Benedenia epinepheli*) were determined to present a high risk for sustainability of *L. calcarifer* sea-cage aquaculture (Table 50).

Table 47. Consequence of parasite establishment and proliferation in sea-cage aquaculture of *Argyrosomus japonicus*.

Parasite taxa	Mortality	Pathology	Marketability	Consumer health	Consequence
Cestoda					
Empty cysts	-	-	x	-	Low
Dasyrhynchus pacificus	-	-	-	-	Negligible
Poecilancistrium calyophyllum	-	-	x	-	Low
Pterobothrium sp.	-	-	-	-	Negligible
Digenea					
Stephanostomum bicoronatum	-	x	-	-	Low
Pleorchis sciaenae	-	-	-	-	Negligible
Monogenea					
Calceostoma glandulosum	-	-	-	-	Negligible
Benedenia sciaenae	x	x	x	-	High
Diplectanum spp.	x	x	-	-	Moderate
Sciaenacotyle sciaenicola	x	x	-	-	Moderate
Nematoda					
Philometra sp.	-	x	-	-	Low
Ascaris sp.	-	-	-	-	Negligible
Anisakis sp.	-	-	-	x	Low
Hysterothylacium marinum	-	-	-	x	Low
Terranova sp.	-	-	-	-	Negligible
Contracaecum legendrei	-	-	-	x	Low
Acanthocephala					
Neoechinorhynchus dorsovaginatus	-	-	-	-	Negligible
Copepoda					
Caligus sp. (male)	x	x	-	-	Moderate
Caligus cf. *elongatus*	x	x	-	-	Moderate
Caligus chiastos	x	x	-	-	Moderate
Isobranchia scianophila	-	-	-	-	Negligible
Lernanthropus gisleri	x	x	-	-	Moderate
Isopoda					
Ceratothoa sp.	x	x	-	-	Moderate
Nerocila sigani	-	-	-	-	Negligible

Parasites were scored for four criteria, (denoted with an 'x') including: 1. previous mortality in aquaculture, 2. potential pathology or disease, 3. potential negative impact on marketability/consumer acceptance and 4. potential negative impact on consumer health. Four *Diplectanum* spp. (Chapter 2, Table 12) are represented as '*Diplectanum* spp.'

Table 48. Parasite risk analysis for sea-cage aquaculture of *Argyrosomus japonicus*.

Parasite taxa	Likelihood	Consequence	Risk
Cestoda			
Empty cysts	Low	Low	Negligible
Dasyrhynchus pacificus	Low	Negligible	Negligible
Poecilancistrium calyophyllum	Low	Low	Negligible
Pterobothrium sp.	Low	Negligible	Negligible
Digenea			
Stephanostomum bicoronatum	Low	Low	Negligible
Pleorchis sciaenae	Low	Negligible	Negligible
Monogenea			
Calceostoma glandulosum	Extreme	Negligible	Negligible
Benedenia sciaenae	Extreme	High	**High**
Diplectanum sp.	Extreme	Moderate	Moderate
Diplectanum spp.	High	Moderate	Moderate
Sciaenacotyle sciaenicola	Extreme	Moderate	Moderate
Nematoda			
Philometra sp.	Low	Low	Negligible
Ascaris sp.	Low	Negligible	Negligible
Anisakis sp.	Low	Low	Negligible
Hysterothylacium marinum	Low	Low	Negligible
Terranova sp.	Low	Negligible	Negligible
Contracaecum legendrei	Low	Low	Negligible
Acanthocephala			
Neoechinorhynchus dorsovaginatus	Low	Negligible	Negligible
Copepoda			
Caligus sp. (male)	High	Moderate	Low
Caligus cf. *elongatus*	Extreme	Moderate	Moderate
Caligus chiastos	Extreme	Moderate	Moderate
Isobranchia scianophila	High	Negligible	Negligible
Lernanthropus gisleri	High	Moderate	Moderate
Isopoda			
Ceratothoa sp.	Extreme	Moderate	Moderate
Nerocila sigani	Moderate	Low	Negligible

Table 49. Consequence of parasite establishment and proliferation in sea-cage aquaculture of *Lates calcarifer*.

Parasite taxa	Mortality	Pathology	Marketability	Consumer health	Consequence
Myxozoa					
Myxozoa gen et. sp. indet	-	-	-	-	Negligible
Cestoda					
Scolex pleuronectis	-	-	-	-	Negligible
Nybelinia indica	-	-	-	-	Negligible
Digenea					
Prosorhynchus sp.	-	-	-	-	Negligible
Pseudometadena celebesensis	-	-	-	-	Negligible
Erilepturus hamati	-	-	-	-	Negligible
Metacercariae gen et. sp. indet	-	-	-	-	Negligible
Cruoricola lates	-*	-*	-	-	Moderate
Parasanguinicola vastispina	-*	-*	-	-	Moderate
Monogenea					
Benedenia epinepheli	x	x	x	-	High
Capsalid gen. et sp. Indet	-	-	-	-	Negligible
Neobenedenia melleni	x	x	x	-	High
Diplectanum spp.	x	x	-	-	Moderate
Laticola spp.	x	x	-	-	Moderate
Pseudorhabdosynochus spp.	x	x	-	-	Moderate
Nematoda					
Hysterothylacium sp.	-	-	-	x	Low
Raphidascaris sp.	-	-	-	x	Low
Raphidascaris sp. II	-	-	-	x	Low
Terranova sp.	-	-	-	x	Low
Acanthocephala					
Serrasentis sagittifer	-	-	-	-	Negligible
Copepoda					
Caligus epidemicus	x	x	-	-	Moderate
Lernanthropus kroyeri	x	x	-	-	Moderate
Lernanthropus latis	x	x	-	-	Moderate
Isopoda					
Cymothoa indica	x	x	-	-	Moderate

For scored criteria see footnote to Table 47. Four *Diplectanum* spp., 3 *Laticola* spp. and 3 *Pseudorhabdosynochus* spp. (Chapter 2, Table 10) are represented. *See Discussion for comment.

Table 50. Parasite risk analysis for sea-cage aquaculture of *Lates calcarifer*.

Parasite taxa	Likelihood	Consequence	Risk
Myxozoa			
Myxozoa gen. et. sp. indet	Moderate	Negligible	Negligible
Cestoda			
Scolex pleuronectis	Low	Negligible	Negligible
Nybelinia indica	Low	Negligible	Negligible
Digenea			
Prosorhynchus sp.	Low	Negligible	Negligible
Pseudometadena celebesensis	Low	Negligible	Negligible
Erilepturus hamati	Low	Negligible	Negligible
Metacercariae gen et. sp. indet	Extreme	Low	Low
Cruoricola lates	Extreme	Moderate	Moderate
Parasanguinicola vastispina	Extreme	Moderate	Moderate
Monogenea			
Benedenia epinepheli	Extreme	High	**High**
Capsalid gen. et sp. Indet	Extreme	Negligible	Negligible
Neobenedenia melleni	Extreme	High	**High**
Diplectanum spp.	Extreme	Moderate	Moderate
Laticola spp.	Extreme	Low	Low
Pseudorhabdosynochus spp.	Extreme	Low	Low
Nematoda			
Hysterothylacium sp.	Low	Low	Negligible
Raphidascaris sp.	Low	Low	Negligible
Raphidascaris sp. II	Low	Low	Negligible
Terranova sp.	Low	Low	Negligible
Acanthocephala			
Serrasentis sagittifer	Low	Negligible	Negligible
Copepoda			
Caligus epidemicus	Extreme	Moderate	Moderate
Lernanthropus latis	Extreme	Moderate	Moderate
Isopoda			
Cymothoa indica	Extreme	Moderate	Moderate

7.5 Discussion

This risk assessment determined the potential likelihood, consequence and risk for sea-cage aquaculture posed by parasite species known to infect wild and/or farmed mulloway (*A. japonicus*) and barramundi (*L. calcarifer*). When considering this risk assessment, it is important to note that the likelihood of parasite establishment is dependent upon the farm location and the distribution of parasite species (including geographic distribution and host-specificity). As new information on parasite distributions in wild and farmed fish populations becomes available, assessments of risks will be better informed. Information on parasite species infecting wild organisms that associate with sea-cages is largely unavailable. Consequently, this risk assessment is unable to account for risk presented by generalist parasite species, or species that may be capable of host switches. Information on host-

specificity changes rapidly and some parasite species previously considered host-specific have been found to attach to a wide range of hosts under experimental conditions (Bricknell *et al.* 2006). The presence of new parasite records on Australian sea-caged *A. japonicus* and *L. calcarifer* in this study highlights the need for ongoing surveillance of wild and captive fishes.

7.5.1 Parasite risks to sea-cage aquaculture of Argyrosomus japonicus

High risk

Monogeneans exhibit a direct life cycle and most have free swimming larval stages, enabling them to complete their life cycle in the sea-cage environment. *Benedenia sciaenae* is an epithelial-feeding monopisthocotylean that infects wild and sea-caged *A. japonicus* in Australia (Chapter 2, Table 12; Table 48; Whittington 1996). If left untreated, high numbers of *B. sciaenae* on the body surface cause scale loss, haemorrhage and localised lesions (Tokşen *et al.* 2007). High infections on the body surface may also render fish unappealing to consumers (Chapter 2, Table 12; Table 47). There are no published records of host mortality associated with *B. sciaenae* (Table 47), but other *Benedenia* spp. are associated with mortality in sea-cage aquaculture, largely through facilitating secondary infections by bacteria (Egusa 1983; Whittington *et al.* 2001a; Ernst *et al.* 2002).

Moderate risk

This study provides the first record of a *Diplectanum* sp. infecting farmed *A. japonicus* (Chapter 2, Table 12). Previously these monogeneans were only known from wild conspecifics (see Williams 1989; Hayward *et al.* 2007b). *Diplectanum* spp. are oviparous monogeneans likely to be associated with pathology (Table 47) because congeners are known to cause epithelial hyperplasia and haemorrhage (Oliver 1977). *Diplectanum* spp. are significant pathogens to sea bass (*Dicentrarchus labrax*) culture in the Mediterranean (Whittington and Chisholm 2008). Effects on fish health likely largely depend on the attachment mechanism possessed by different diplectanid species and whether their squamodisc acts as a friction pad that may provoke a host response (e.g. *Diplectanum aequans*, see Oliver 1977) or as a series of several overlapping sclerites that create suction (e.g. *Furnestinia echeneis*, see Desdevises *et al.* 2001).

High burdens of the blood-feeding polyopisthocotylean *Sciaenacotyle sciaenicola* have been associated with pale gills in sea-caged *A. japonicus* in South Australia (Hayward *et al.* 2007b; Table 47). An outbreak of a similar species, *Sciaenacotyle panceri* on sea-caged *A. regius* in the Mediterranean was associated with host lethargy, emaciation, gill anaemia and mortality (Merella *et al.* 2009; Table 47).

Caligid copepods have direct life cycles consisting of free-living, free-swimming stages and attached parasitic stages. Severe ectoparasitic copepod infestations in aquaculture have been associated with mortalities through host osmoregulatory failure, anaemia, ulcerations or through facilitating secondary infections (Finstad *et al.* 2000). Hayward *et al.* (2007b; 2008) proposed that *Caligus chiastos* infecting farmed *A. japonicus* in South Australia may cause damage in high numbers because similar species are well-known to be associated with pathology (damage to epidermal and dermal tissue) among cultured salmonids. Although *C. chiastos* is not known to cause mortalities in sciaenid aquaculture, this species which has low specificity has been associated with gross corneal damage and epizootics in sea-caged southern bluefin tuna (*Thunnus maccoyii*) in South Australia (see Hayward *et al.* 2008; Table 47).

Lernanthropus gisleri infects wild *A. japonicus* in South Australia and New South Wales, but has not been documented from sea-caged fish in these regions (Chapter 2, Table 12). A similar species, *L. kroyeri*, has been responsible for high mortalities of Mediterranean sea bass, *Dicentrarchus labrax* as a result of host asphyxia and anaemia (Manera and Dezfuli 2003; Henry *et al.* 2009; Table 47). Considering the high likelihood of establishment in sea-cages and disease-induced mortality of similar species, *L. gisleri* presents a moderate risk for sea-cage culture of *A. japonicus* (Table 48).

This study provides the first record of *Ceratothoa* sp. from the tongue of farmed *A. japonicus* in New South Wales (Chapter 2, Table 12). A similar species, *Ceratothoa gaudichaudii*, is an economically important isopod parasite of marine sea-caged fish in Chile (Sievers *et al.* 1996). The species has been reported from a wide range of native hosts and low host specificity has enabled it to successfully infect farmed coho (*Oncorhynchus kisutch*) and Atlantic salmon (*Salmo salar*). *Ceratothoa* spp. feed on host blood when attached to the inner mouth surfaces, and less frequently to the gills. Damage to the host includes ulcers on the

gill arch and inside the mouth. Sievers *et al.* (1996) reported reduced body weight of *S. salar* with increasing burdens of *C. gaudichaudii.*

Low risk

A single male *Caligus* sp. infected wild *A. japonicus* in South Australia (Chapter 2, Table 12). Species in *Caligus* should be treated with caution when assessing risk because host-specificity of *Caligus* spp. is not fully understood and some species may infect numerous host families (see Johnson *et al.* 2004a for review).

Negligible risk

Cestodes transfer to piscivorous fish when infected intermediate hosts are eaten. *Poecilancistrium caryophyllum* causes large, white cysts in the muscle tissue of wild *A. japonicus* in New South Wales (Robinson 1965) but were not observed in farmed fish (Chapter 2, Table 12). While the cysts have implications for marketability, it is not clear whether they are associated with a pathological host response (Adjei *et al.* 1986; Table 47). In Japan, farmed *Seriola quinqueradiata* fed parasitised raw fish became infected with a larval cestode, *Callitetrarhynchus nipponica*, which altered the appearance and reduced the marketability of the flesh (Ogawa 1996). However, when raw fish was replaced with frozen food, the parasite disappeared from farm sites (Ogawa 1996).

Digenean infections, with the exception of aporocotylids, generally do not appear to be significant in aquaculture (Williams and Jones 1994; Ogawa 1996). Most digeneans have complex life cycles involving two or more host species that are difficult to complete in culture systems. Digeneans that inhabit the digestive tract are generally considered less pathogenic and have received comparatively little attention in the literature (Williams and Jones 1994). Nonetheless, *Stephanostomum bicoronatum* could cause some pathology to *A. japonicus* (Table 48) considering Grau *et al.* (1999) observed mucosal and submucosal lesions in *Seriola dumerili* infected with a similar species, *S. pristis*. Considering the low likelihood and negligible/low consequences, *S. bicoronatum* and *Pleorchis sciaenae* pose a negligible risk to sea-cage aquaculture of *A. japonicus* (Table 48).

The monogenean *Calceostoma glandulosum* has been reported previously from farmed *A. japonicus* in South Australia (Hayward *et al.* 2007b) and were found to infect wild fish in South Australia in this study (Chapter 2, Table 12). There is no record of a *Calceostoma*

species causing mortality or disease in cultured fishes and Hayward *et al.* (2007b) found no evidence of gross pathology associated with low infection intensities on farmed *A. japonicus* in South Australia. It is interesting to note that other monogenean species are considered a high risk. This suggests that the biology of some species of monogeneans, presumably their attachment methods, fecundity, egg dispersal and/or larval behaviour, renders them more benign than others.

Farmed fish may become infected with nematodes if they are fed live foods containing infective parasite stages, or if infected intermediate hosts are able to be consumed by caged fish. Anisakid nematodes often cause the development of lesions (Dezfuli *et al.* 2000) and are potentially harmful to consumer health. Anisakiasis is a parasitic disease of the human gastrointestinal tract, caused by ingestion of live marine nematode larvae. *Anisakis simplex* and *Pseudoterranova decipiens* are nematode species known to be associated with this disease (Ugenti *et al.* 2004), and *Contracaecum* spp. and *Hysterothylacium* spp. are also potentially infective to consumers (Williams and Jones 1994; Table 48). Infection can occur when raw or undercooked fish that contain live parasites is consumed.

Philometrid nematodes have been detected in wild mulloway in Western Australia (Bryn Farmer, personal communication) and South Australia (Chapter 2, Table 12), but have not been reported from farmed fish. Philometrid nematodes parasitise male and female fish, with infection usually associated with spawning individuals (Hesp *et al.* 2002). The testes of *Chrysophrys auratus* (as *Pagrus auratus*) infected with *Philometra lateolabracis* undergo extensive atrophy (Hine and Anderson 1981), while heavy infections of philometrid species in the ovaries of *Mugil cephalus* led to necrosis of the oocytes. Indeed, invasion of the gonads of sexually mature *Pomatomus saltatrix* by *Philometra saltatrix* reduced the reproductive potential of this species (Hesp *et al.* 2002). Consequently, philometrid nematodes may be a problem for wild mulloway captured and maintained for brood stock, but are of less concern for sea-caged fish produced for the commercial market (Table 47).

7.5.2 Parasite risks to sea-cage aquaculture of Lates calcarifer

High risk

The monogeneans *Neobenedenia melleni* and *Benedenia epinepheli* are notorious, allegedly generalist pathogens of tropical and subtropical fishes in aquaria and aquaculture worldwide.

Both species attach to the skin of their host and graze on skin cells, continuously laying eggs into the water which hatch into ciliated larvae that directly re-infect fish. High infection intensities on fish lead to secondary infections by bacteria, ultimately resulting in emaciation and death. In 2000 an outbreak of *N. melleni* resulted in the loss of 200,000 farmed barramundi (50 t) in Hinchinbrook, Queensland (Deveney *et al.* 2001). Outbreaks of *B. epinepheli* have been recorded in aquaculture on at least six species of fish and on 18 species in aquaria (see Whittington *et al.* 2001b for review).

Whittington (2004; 2005) has hypothesised that *Neobenedenia* '*melleni*' and *N.* '*girellae*', the two species widely attributed globally to pathogenic infections in aquaria and aquaculture, may each be a complex of several species. Furthermore, the source of *Neobenedenia* outbreaks in sea-caged fish is mostly unknown (Whittington and Chisholm 2008) and this applies to the *N.* '*melleni*' infection of barramundi in Hinchinbrook (Deveney *et al.* 2001). Moreover, *B. epinepheli* is only recorded in the wild from two host species (Whittington *et al.* 2001b). These reports emphasise the importance of broader studies to determine specific parasite identities and their host range because it may influence husbandry practices and farm location.

Moderate risk

At least 10 diplectanid monogenean species (including *Diplectanum* spp., *Laticola* spp. and *Pseudorhabdosynochus* spp.) infect sea-caged *Lates calcarifer* (Chapter 2, Table 10). Infected fish exhibit a darkened body, rub against the net, pale gills, lethargy, loss of appetite and excess mucus production (Leong *et al.* 2006). Leong and Wong (1990) recorded that a large proportion of diseased *L. calcarifer* were infected with *Laticola latesi* (as *Pseudorhabdosynochus latesi*) and *Diplectanum* sp. Until more is known about the specifics of parasite attachment, any associated pathology and fecundity, these diplectanid genera are considered moderate risks.

Aporocotylids may be problematic in aquaculture because their intermediate invertebrate host may inhabit areas close to farmed fish, such as on cage structures or nearby sediment, and infection of the definitive host by emerging cercariae is direct. *Cruoricola lates* infect farmed *L. calcarifer* in Malaysia, Thailand and Australia (Herbert *et al.* 1994) and *Parasanguinicola vastispina* infects cultured fish in Malaysia (Herbert and Shaharom-Harrison 1995) (Chapter 2, Table 10). Although these genera have no known pathology,

aporocotylids are associated with mortalities of farmed amberjacks, *S. dumerili*, in the Spanish Mediterranean (Crespo *et al.* 1992) and Japan (Ogawa and Fukudome 1994) and farmed *Thunnus maccoyii* in Australia (Hayward *et al.* 2010).

Caligus epidemicus presents a predominant threat to aquaculture in Australian and Asian waters due to its distribution and low host specificity (Ho *et al.* 2000; Hutson *et al.* 2007b). This species is also known to cause mass mortality of mullet (Mugilidae) and porgies (Sparidae) in Australia (Hewitt 1971) and Taiwan (Lin 1996) and is known from sea-caged *L. calcarifer* in Malaysia (Venmathi Maran *et al.* 2009). In view of the parasites' direct life cycle and potential for mortality and disease outbreaks, *C. epidemicus* presents a moderate risk for sea-cage aquaculture of *L. calcarifer* (Table 49).

Lernanthropus spp. infections are emerging in sea-cage barramundi industries in Queensland (Chapter 2, Table 10) and Western Australia (Hutson, unpublished data). Prior to this study, *L. latis* was known from wild *Lates calcarifer* from Celebes, India, Sri Lanka and Thailand (see Ho and Kim 2004b) and farmed *L. calcarifer* in the Northern Territory, Australia (Kuo and Humphrey 2008). *Lernanthropus latis* presents an extreme likelihood of establishment and proliferation in sea-cage aquaculture (Table 50). The consequences for culture may include disease problems (lacerated tissue, erosion and necrosis of secondary gill lamellae) and mortality (e.g. Manera and Dezfuli 2003; Henry *et al.* 2009). According to Kuo and Humphrey (2008) this species has not yet been associated with significant fish mortality events even though their presence is usually associated with poor fish health. Vinoth *et al.* (2010) document *L. kroyeri* from *Lates calcarifer* sampled in India, but did not accession any specimens for further study. Considering *Lernanthropus latis* has been documented previously from the same host in India (Tripathi 1957) and that *L. kroyeri* has only been previously recorded from *Dicentrarchus labrax*, validation of this record would be necessary prior to inclusion in these risk analyses.

Cymothoa indica (Isopoda) is known to infect hatchery reared *Lates calcarifer* larvae in India (Rajkumar *et al.* 2005). The parasite is believed to have been introduced to hatchery fish through wild zooplankton used as feed. Infections in the branchial and anterodorsal regions resulted in skin lesions and were associated with lowered growth rates and mortality (Rajkumar *et al.* 2005).

Low risk

Farmed *L. calcarifer* sampled from Port Hinchinbrook, Queensland, exhibited high burdens of metacercarial cysts in the gills (Chapter 2, Table 10). Assessment of risk is challenging with limited information available on species identification or potential to cause pathology. In view of the intensity of the infection and gross damage observed, further investigation is warranted.

Negligible risk

Myxozoa are now recognised as relatives of cnidarians and are believed to have a two-host life cycle involving fish and invertebrates (Moran *et al.* 1999). An undescribed species has been documented from the gall-bladder of farmed *L. calcarifer* (Table 50). *Myxobolus spirosulcatus* (see Maeno *et al.* 1995) is also known from the gall-bladder of farmed *Seriola quinqueradiata* in Japan (Yokoyama and Fukuda 2001), but there have been no apparent pathological changes or mortality associated with infection. Assessment of the risk of the unidentified myxozoan is challenging with limited information available on species identification or potential to cause pathology.

We found no literature concerning potential pathology of larval tetraphyllideans and the trypanorhynch *Nybelinia indica* documented from the digestive tract of L. calcarifer (Chapter I, Table 50).

Non-aporocotylid digenean parasites can be easily managed in farms by feeding fish with uninfected items. Some farmed fish may still become infected by trematodes, by feeding opportunistically on wild species harbouring infective stages moving through the netting, but it is unlikely that these parasite species will proliferate in the farmed population. Given the paucity of information available in the literature concerning the relative pathogenicity of trematodes that infect the digestive tract, they are considered to present negligible risk for *L. calcarifer* sea-cage farming (Table 50).

7.6 Husbandry practices to manage metazoan parasite infections

Accurate identification of parasites in wild organisms and in aquaculture is critical as knowledge of the specific biology of species can help to facilitate selection of the most

appropriate management methods (e.g. location of farm; fallowing of sites; sea-cage net changes; use of antifoulants; cage position with regard to current direction; use of therapeutic chemicals; timing of treatment to break life cycles). Parasite identification is also significant in biosecurity as some pathogenic species in aquaculture are known to have inadvertently translocated from one region to another, leading to the spread of disease. In some cases, parasites may establish in hatcheries or nursery facilities (e.g. *Cymothoa indica* in *L. calcarifer*) and subsequent stress associated with transportation of juveniles to sea-cages may facilitate parasite proliferation. Consequently husbandry must begin with brood stock and fingerlings on land based facilities.

Parasitic crustaceans (Copepoda and Isopoda), monogeneans and aporocotylid trematodes present the greatest likelihood of establishment and proliferation in sea-cage aquaculture because invasive stages directly infect their host. Suggested husbandry practices to manage outbreaks of these parasite groups are discussed.

7.6.1 Monogenean and crustacean infections

Current management of isopods, copepods and monogenean infections in Australia's sea-cage aquaculture industry relies on bathing cages of fish in fresh water or hydrogen peroxide solution (Rach *et al.* 2000; Mansell *et al.* 2005). Bath treatments of sea-caged fish are labour-intensive, time-consuming, weather dependent, costly and stressful to fish (fish cannot be fed on days prior and post treatment). Although the industry has developed considerable expertise in bath treatments, mortalities may still occur due to difficulties in calculating bath solution, physical damage to fish from crowding, or lack of oxygen (Williams *et al.* 2007). Ectoparasites can also be treated with approved, prescribed veterinary medications. However, prolonged use of chemical treatments may lead to the development of parasite resistance and reduced efficacy. Seven years of continued use of emamectin benzoate to manage *Caligus* infections in sea-caged *S. salar* in Chile has resulted in reduced efficacy (Bravo 2010). Alternatively, Chambers & Ernst (2005) demonstrated that hydrographic conditions should be taken into account to minimise infection within and between sea-cage farms by dispersing eggs and infective larvae.

Although there are no registered chemotherapeutants in Australia, ectoparasite species could be managed in the event of an outbreak using prescription or permit medication. Applications to seek exemptions from the need for registration for a number of chemicals used by the

aquaculture industry can be submitted to the National Registration Authority. A formal exemption means that the aquaculture industry can legally use a particular drug or chemical for certain specified uses without the need to get it formally registered or permitted. The following chemicals have been granted exemptions from the need for registration as a result of this process: calcium carbonate, calcium hydroxide, calcium oxide, calcium/magnesium carbonate, calcium sulphate, zeolite, aluminium sulphate, ferric chloride, inorganic fertilizers, and organic fertilizers. A Minor Use Permit (MUP) is a short term permit given for approximately 12 months on the basis that not enough information or resources are available to sustain a full application for registration and the quantities used are relatively small.

Cycles of reinfection can be prevented if treatments are coordinated strategically to break the life cycle (Ernst *et al.* 2005). A similar result can be achieved by fallowing farm sites (Bron *et al.* 1993). In order to effectively break parasite life cycles, fundamental biological information (e.g. species determination, epidemiology, fecundity, fate and embryonation time of eggs, time to reach sexual maturity, adult longevity) is required for a breadth of environmental conditions. Controlled experimental infections in the laboratory that examine the effect of temperature and salinity on 1) adult parasite survival; 2) adult parasite fecundity; 3) parasite egg hatching success; 4) parasite larval survival and 5) host infection rates will enable predictive models that assess parasite populations, infection dynamics and the timing and efficiency of control measures. Models determine effects of specific management strategies for specific environmental conditions (i.e. temperature and salinity) in order to break parasite life cycles.

Knowledge acquired from these models can be synthesised to create informed workable management practices to control parasites in different environmental scenarios and different aquaculture systems. This can include husbandry and intervention recommendations for brood stock and fish in grow-out. In addition, knowledge of what parasite species are locally abundant and/or the seasonality of wild host association with farm sites could be incorporated into a more complex model that accounts for reinfection from external sources. Minimising the number of escaped, infected fish that remain in the vicinity of the farm will also reduce reinfection of treated fish.

7.6.2 Aporocotylid infections

Blood flukes of fishes (Aporocotylidae) are an emerging problem in marine fish aquaculture worldwide. Recent studies of Australian marine fishes have revealed an unexpected, extremely diverse blood fluke fauna (Chapters 1, 2; Nolan and Cribb 2006a; Nolan and Cribb 2006b). Blood flukes are an important group for studies of diversity and host-parasite relationships because they infect some of the most commercially valued farmed marine fishes in the world and can be highly pathogenic to their intermediate and definitive hosts. Asexual stages castrate their intermediate host (Koie 1982) and eggs laid by adult parasites may cause asphyxiation and mortality in cultured fish (Ogawa and Fukudome 1994).

Control of blood fluke infections may only be achieved in semi-open aquaculture systems by separating intermediate and definitive hosts, as elimination of susceptible intermediate hosts in open water is impractical and cost-prohibitive (Bullard and Overstreet 2002). We are aware that some farms in Japan use orally delivered praziquantel to treat *Seriola* spp. infected with adult blood flukes, however, to our knowledge, the effectiveness of this treatment has not been quantified.

Identifying the intermediate host species for aporocotylid species may help to determine suitable sea-cage sites away from potential infection sources as the industry expands (Hutson and Whittington 2006; Chapter 2). The intermediate host species for nearly all marine aporocotylids are currently unknown. However, sequence data from adult aporocotylids in fish may be matched to parasite stages recovered from intermediate hosts. Unpublished sequence data from this study (Chapter 2) was provided to other researchers to facilitate the identification of intermediate blood fluke stages recovered in terebellid polychaetes in Port Lincoln, South Australia (personal communication, T. Cribb). Continued generation of adult and larval sequence data for a number of informative genes or gene fragments will facilitate our growing understanding that the role intermediate invertebrate hosts play in the environment and whether it is feasible to relocate sea-cages to areas where viable intermediate hosts are absent.

7.7 Conclusion

Parasites present major economic and environmental concerns for aquaculture and fisheries. In Australasia wild fish are widely introduced to farms for brood stock and may be reservoirs

of parasite infection in open and closed systems. The likelihood of parasite establishment and proliferation in aquaculture can be estimated by assessing and examining exposure of farmed fish to infective parasite stages. Parasites with single host life cycles and/or free swimming infective stages are most likely to establish and proliferate in aquaculture because they may reproduce rapidly and can directly infect their hosts. Amplification of parasite species in sea-cage culture may also present risks for aggregating conspecifics. The species most likely to threaten sustainability of the *A. japonicus* sea-cage aquaculture industry is *Benedenia sciaenae*, while *Neobenedenia melleni* and *B. epinepheli* present the highest risk for sea-cage culture of *L. calcarifer* sea-cage culture. Current ectoparasite control methods provide only short term solutions and substantially increase production costs. An understanding of wild sources and/or reservoirs of high risk parasite species at sea-cage farm locations (e.g. Catalano & Hutson 2010) and knowledge of their biology and life stages will help determine and refine proactive management strategies.

8 MARINEPARASITES.COM

8.1 Abstract

The website MarineParasites.com was developed to enable increased accessibility to information on parasites of marine fishes. More than 3,525 unique visitors from 114 countries have viewed the website to date. Positive feedback has been received from representatives of the target audience including recreational fishers, scientists and members of the aquaculture industry. The website has facilitated further communication and extension through national television and has made a major contribution to the availability of knowledge about parasites infecting recreational, commercial and farmed finfish in Australia.

8.2 Introduction

The Australian public and, indeed, the public globally, are poorly informed about parasite infections in wild fish species. When infections are noted, hysteria in the media may result as there is no factual information presented simply about marine finfish parasites that is readily accessible to the public. One of the most important objectives of this research project was to develop a comprehensive website that provides basic information on parasite appearance and biology to enable lay people to identify and understand the different types of parasites in fish. MarineParasites.com presents information on some of the most commonly cultured and recreationally captured fishes in southern Australian waters, as well as barramundi from tropical waters.

8.3 Materials and Methods

MarineParasites.com was designed by the PI together with Mr Simon Dibb of Elysium Design, to ensure a professional, user friendly layout. The site is now managed by the PI using Dreamweaver RCS4.

Six key menu items were created: Home, Parasites, Fish hosts, Research, Staff and Acknowledgements which include:

1) Background information on marine parasites and parasite diversity

2) Ten colour posters with detailed information on parasites of important fish species
3) Recent research news
4) Over 100 images of parasites, hosts species, laboratory and field work
5) Two media articles and a link to a televised feature about marine parasites that aired in September 2010 on Channel 10's *Scope*
6) A list of staff and collaborators
7) Acknowledgements of grants, scholarships, travel and commercial support

The website is image rich in an attempt to capture and engage audience interest (Figure 23). Where feasible, we have attempted to provide photographs of parasites attached to their hosts to assist identification. Most images of parasites *in situ* reflect how anglers would encounter marine parasites on their catch. For those parties interested in identifying parasites to species level, we recommend they contact us or an appropriate state government organisation. For those who contact us, we will provide appropriate publications that include drawings and, in some cases, keys to species that have resulted from this project.

8.4 Results and Discussion

There have been 2, 3,525 absolute unique visitors to the website from 114 countries. The top twelve visiting countries were: Australia, United States, United Kingdom, Chile, Canada, Malaysia, Philippines, Japan, India, New Zealand, Spain and Brazil. The website has been an excellent communication vehicle and the PI is contacted regularly by members of the public with questions and positive feedback. MarineParasites.com acts as a vital first stop for interested lay people and scientists. The PI will continue to maintain and regularly update the website. Ten colour posters (easily downloaded as pdf documents) highlight specific fish-parasite case studies (Figure 24, 25). In addition to the posters, more than 100 images can be viewed in the photo gallery.

Figure 23. A screen shot of the home page of MarineParasites.com. This attention-grabbing image of a tongue biter isopod, *Ceratothoa* sp. in the mouth of a trevally (*Pseudocaranx georgianus*), has been instrumental in attracting and engaging visitors to the website.

Figure 24. Colour poster of problematic parasites for mulloway aquaculture. This poster is one of ten colour posters available through MarineParasites.com which examine the most commonly encountered parasites that infect recreational, commercial and cultured fish species.

Figure 25. Colour poster of problematic parasites for barramundi aquaculture. This poster is one of ten colour posters available through MarineParasites.com which examine the most commonly encountered parasites that infect recreational, commercial and cultured fish species.

9 BENEFITS

This parasitological survey of selected important Australian fish species discovered a plethora of new parasite records and parasite species. This information has intrinsic value for understanding Australia's natural biodiversity (Chapters 2 and 4), geographic population structure of fisheries (Chapter 3) and for identifying threats to captive fishes (Chapters 5-7). Risk analyses provide a disciplined and consistent approach of the relative level of risk associated with individual parasite species. Our risk assessments can be used as a model to assess threats to sea-caged and wild fish species. Parasite species determined to pose the greatest threat to the sustainability and profitability of the mulloway and barramundi industries were identified. Appropriate management measures have been suggested and may now be implemented to help control infections or outbreaks (Chapter 7). The results of this research have been disseminated through a website designed to be easy to navigate and interpret for lay people and scientists (Chapter 8).

To illustrate the direct and specific benefits of this research, we provide details for a *selection* of specific outcomes this research has had for the aquaculture industry, aquatic animal health service, the public, the scientific community and the media.

Aquaculture industry

Bluewater Barramundi. Information provided to personnel on parasites found infecting their sea-caged fish and recommendations for parasite management.

Clean Seas. Recommendation for parasite management was provided in response to a query from Health Manager Tom Bayley in regards to an isopod infection of farmed yellowtail kingfish and biosecurity of grow-out sites.

Clearwater Mulloway. Information provided to personnel on parasites found infecting their sea-caged fish and recommendations for parasite management.

Marine Produce Australia. Preliminary parasite identifications and recommendation for parasite management were provided for Marine Produce Australia for sea-cage aquaculture of barramundi.

Darwin Aquaculture Centre. Assisted in the identification of a problematic free-living platyhelminth in clam culture for Evan Needham, Senior Aquaculture Technician

Aquatic animal health services

ADL-Diagnostics – Chile. Frequent exchange with Dr Ricardo Ildefonso, veterinarian in Chile and assistance with *Paradeontacylix* identification in jacks (*Seriola* spp.).

Batavia Coast Marine Institute. A request was granted to Dr Colin Johnson, Research and Development Officer, Batavia Coast Marine Institute, Geraldton, Western Australia for information on likely parasite issues for yellowtail kingfish (*Seriola lalandi*) sea-cage trials.

Berrimah Veterinary Laboratories. Identifications of parasite species were made for Dr Kitman Dyrting Veterinary Officer/Pathologist, Department of Resources, Darwin.

Digsfish Services. Information on parasite assemblages of southern garfish (*Hyporhamphus melanochir*) was provided to Dr Ben Diggles for incorporation in a risk analysis for bait.

Members of the public

Responses were provided for all electronic mail sent to the PI from members of the public in relation to marineparasites.com. This usually involved addressing specific questions on parasites that recreational fishers had found on and in their catch.

Identifications were provided for parasites infecting southern garfish (*Hyporhamphus melanochir*) and King George whiting (*Sillaginodes punctatus*) from photos and specimens supplied by Mr Alan Slater of Whyalla.

A selection of specimens, images and videos about marine parasites collected during our project are displayed permanently in the Biodiversity Gallery at the South Australian Museum (opened: February 2010). In addition to MarineParasites.com, this museum gallery is another way to inform the public about marine parasite biodiversity and their biology. In 2009, the SA Museum received >800,000 visitors. Joint Investigator (JI), Ian Whittington, has engaged with the public on several occasions in the Biodiversity Gallery about parasites. He has also trained about 25 volunteer guides.

Members of the scientific community

For a list of all presentations arising, see Appendix 4

Centro de Ciencias do Mar, France. A request from Dr Christophe Haond was granted for specimens of marine *Argulus* for his work on osmoregulation.

Flinders University. Seriola spp. tissue from over 150 individuals sampled by the PI were provided for an Honours research project (Penny Miller, Flinders University). The PI also contributed to writing a manuscript that developed from this research (see Appendix 3).

Institute of Parasitology, Academy of Sciences of the Czech Republic. New cestodes were provided to Dr Roman Kuchta for description: Kuchta, R., Scholz, T. and Justine, J.L. 2009. Two new species of *Bothriocephalus* Rudolphi, 1808 (Cestoda: Bothriocephalidae) from marine fish off Australia and New Caledonia. *Systematic Parasitology* **73,** 229-238.

CSIRO. Unpublished data was provided to Catherine Seytre, Marine Ecologist, CSIRO Marine & Atmospheric Research for a quantitative review of fish movements.

Murdoch University. Parasite images were provided to Michael Klunzinger, Fish Health Unit, Murdoch University, Western Australia, for seminars delivered to veterinary students.

New South Wales Department of Primary Industries. Aided Dr John Stewart, Research Scientist with the identification of nematode worms infecting garfish.

Department of Primary Industries, Victoria. Aided with identification of a parasite of recreational fishes as per a request made by Troy Duthie, Recreational Fisheries Officer, Warrnambool.

Australian Museums. The majority of parasite specimens and also some host specimens and fish tissue samples obtained in this study were accessioned to collections (Australian Biological Tissue Collection, Helminths, Ichthyology and Marine Invertebrates) at the South Australian Museum. Specimens were also accessioned to Museum of Tropical Queensland, Museum Victoria and Natural History Museum, London.

University of Adelaide. Parasite and some host material collected was provided to Lizzie Perkins and was sequenced as part of an independent study on parasite phylogeny. This has resulted so far in two published papers and one PhD thesis (see Appendix 3).

Teaching. The PI and the JI contribute teaching to undergraduate courses at James Cook University and the University of Adelaide, respectively. Tomorrow's professional marine biologists are today's students and Hutson and Whittington each use their experiences, discoveries and publications from projects as examples in their teaching.

Training: Three Honours students, Sarah Catalano and Emma Brock (University of Adelaide) and Penny Miller (Flinders University) have benefited directly from research training during the course of this project.

Members of the media

For a list of all media arising, see Appendix 5

The Age. Comment was provided to Ian McIlwraith, business section, The Age, on parasites infecting yellowtail kingfish (*Seriola lalandi*).

Network Ten. Following contact made by Network Ten, the PI participated in filming a marine parasitology segment for the children's science programme, *Scope*, broadcast 25 September 2010.

Whyalla News. Comment was provided to Seema Sharma - Whyalla News on parasites infecting local fishes.

10 FURTHER DEVELOPMENT

Results from this project have been widely disseminated to the Australian public, fishing industry and aquaculture industry through direct communication, the website (MarineParasites.com) and conference presentations. Presentations were delivered at the 7th International Symposium on Fish Parasites (ISFP), the 10th International Congress for Parasitology (ICOPA), Australasian Aquaculture and Australian Society for Parasitology conferences (Appendix 4). This has resulted in a better understanding of parasite diversity, identification and potentially problematic species. In combination with other studies on marine parasitology, the research could be used to attempt an industry-wide approach to parasite biosecurity and animal husbandry but we are far from a comprehensive picture. The results can be applied commercially by aquaculture companies if they choose to adopt any of the suggested strategies (Chapter 7).

We plan to meet one of our extension objectives '*Dissemination of information to industry in a workshop*' in 2011. It is our intention to run this workshop at James Cook University in Cairns on the 10th of July, 2011. This is timely considering the workshop will immediately follow the First Australasian Scientific Conference on Aquatic Animal Health (5th – 8th July) and is prior to The Australian Society for Parasitology annual conference (11th – 13th July), both of which will be held in Cairns. Consequently the workshop will raise broader interest for industry and other stakeholders, aquatic animal health professionals and students.

11 PLANNED OUTCOMES

Parasite species identification and knowledge of parasite biology is critical for effective parasite management in aquaculture. New knowledge of the parasite species infecting farmed fish will facilitate the development of appropriate parasite treatments and husbandry practices in the Australian sea-cage aquaculture industry (Chapter 7). In addition, identification of naturally occurring parasite species in Australia and an appreciation of what may be endemic and what may be shared with neighbours throughout the Indo-Pacific and more broadly is vital for rapid identification of invasive fish parasites and diseases (Chapter 2 and 6). We redescribed three parasites species, describe one new species and document numerous species previously not known to occur in Australian waters. We have identified a plethora of species that will be described or redescribed and published in scientific journals because material and sequence data will continue to be analysed following the official completion of this research project. Specimens have been accessioned to museums and will continue to prove valuable to other taxonomists and systematists worldwide.

MarineParasites.com (see Chapter 8) delivers information to recreational fishers and the general public about parasites in fish. Initially, identification keys were to be posted on the website, but given that specialised optical equipment and terminology is required for definitive identifications, we deemed it more appropriate to provide informative photographs of frequently encountered parasites on fish. Detailed identification keys are provided for blood flukes in *Paradeontacylix* (see Chapter 5). Given that it is often difficult to identify parasite species in this group definitively, we encourage industry to email photographs and send specimens for identification. Furthermore, this course of action contributes significantly to our ongoing studies on parasites of finfish.

Aporocotylid blood flukes threaten the sustainability of *Seriola* aquaculture in Australia. Sequences for aporocotylid blood flukes from this study will be provided to GenBank™ when Chapter 5 is developed for scientific publication. This will enable DNA-based molecular techniques for the identification of blood fluke species in farmed *Seriola* hosts. Blood fluke adults are notoriously difficult to collect from the circulatory system of their fish hosts and the parasites die and degenerate rapidly following host death. There is also increasing evidence that some blood fluke species may exhibit site specificity (Chapter 5; Sho Shirakashi, pers. comm.). Consequently, current routine sampling of only a select number of organs or parts of

the circulatory system may not necessarily reveal an infection when present. Blood fluke eggs that lodge in gill lamellae of *Seriola* spp. can now be isolated from the gills of an infected fish and matched to the sequences we have generated. Matching of these sequences to life stages in intermediate hosts may reveal blood fluke life cycles. Unpublished sequence data from this project were provided to other Australian researchers (University of Queensland, SARDI Aquatic Sciences and the University of Tasmania) for this specific purpose. These investigators found intermediate blood fluke stages in invertebrates and required a 'bank' of adult aporocotylid DNA sequences from fish hosts in order to have any success in obtaining a match. In future, this baseline work may be used to provide diagnostic testing capabilities to the aquaculture industry and may also enable positive identification of candidate intermediate hosts and contribute to life cycle knowledge. Such studies are important to determine the specificity of intermediate host species to infective stages to inform sea-cage location away from potential infection sources.

We gratefully recognise the contribution that this project has had for the career development of one early career researcher and two junior scientists. This project was central to initiating the career of the PI, who successfully secured an academic teaching and research position at James Cook University during the period of the project. It has also initiated the career of three Honours students: Emma Brock BSc Hons (completed in November 2010), Sarah Catalano BSc Hons, now a PhD candidate in molecular genetics and parasitology at the University of Adelaide and Penny Miller Bsc Hons, now a PhD candidate with the Australian Seafood Cooperative Research Centre, Deakin University.

This project developed a financial and scientific partnership between the FRDC and ABRS for the first time. The mission and goals of these two funding agencies differ. FRDC maintains an applied focus to the fishing industry whereas ABRS provides grants for taxonomic research and to build the taxonomic workforce of Australia. In this co-funded project, we have succeeded in balancing these different objectives. It has also involved national collaboration with James Cook University, the South Australian Museum and the University of Adelaide. International collaboration has been enhanced with the Natural History Museum, London and the University of Valencia.

12 CONCLUSIONS

This research project met ten original objectives (See 1.3 Objectives, pg 19).

Discovery and documentation of parasite fauna of wild and farmed fish should be incorporated into any ongoing sampling programs for effective parasite management and risk assessment (Chapters 2, 4-7). Effective mitigation of parasite species infecting fishes in sea-cage farms can only be achieved through reliable parasite identification (Chapters 2-5), knowledge of their biology (Chapter 6) and assessment of appropriate management methods (Chapter 7). Recognition of parasite species that may decrease profitability through reduced marketability, morbidity and/or mortality of stocks is crucial (Chapters 5-7). Parasites also enable further insight into the geographic population structure of fish stocks (Chapter 6) which is critical for fisheries management. This research delivered on its objective to identify parasites of potential threat to the sustainability of the Australian sea-cage aquaculture industry (Chapters 2, 5-7). Husbandry practices were identified that will enable development of the most appropriate management strategies to avoid outbreaks in Australian aquaculture (Chapter 7). Knowledge gained from this project is accessible to the wider public through the development of a professional, user friendly website (Chapter 8).

This research has generated biological material (including new and poorly known parasite species) that could involve several more years of further taxonomic work to produce several more publications - far beyond the means of this three year research project. Our research has emphasised the diversity of parasites that occur in the marine ecosystem and how scant our knowledge is. We plan to continue to work on this material and will continue to publish the results (Appendix 5). This identifies and underscores the need to continue to fund similar projects and continue the partnership with FRDC and ABRS, in order to generate the information required to manage wild fisheries sustainably, ensure the welfare of farmed fishes and also to train marine parasitologists of the future.

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14 APPENDICES

14.1 Appendix 1: Intellectual property

There are no intellectual property issues associated with this project.

14.2 Appendix 2: Staff list

Principal Investigator: Kate S. Hutson[1,2]

Joint Investigator: Ian D. Whittington[1,3,4]

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14.3 Appendix 3: Publications

1. Catalano, S.R., Hutson, K.S., Ratcliff, R.M. & Whittington, I.D. 2011. The value of parasite and host identification for arripid fish. *Marine and Freshwater Research* **62**, 72-82.

2. Hutson, K.S., Brock, E.L. & Steer, M.A. 2011. Spatial variation in parasite abundance: evidence of geographic population structuring in *Hyporhamphus melanochir*. *Journal of Fish Biology* **78**, 166-182.

3. Catalano, S.R., Hutson, K.S., Ratcliff, R. & Whittington, I.D. 2010. Redescription of two species of microcotylid monogeneans from three arripid hosts in southern Australian waters. *Systematic Parasitology* **76**, 211-222.

4. Catalano, S.R. & Hutson K.S. 2010. Harmful parasitic crustaceans infecting wild arripids: a potential threat to southern Australian finfish aquaculture. *Aquaculture* **303**, 101-104.

5. Perkins E.M., Donnellan S.C., Bertozzi T. & Whittington I.D. 2010. Closing the mitochondrial circle on paraphyly of the Monogenea (Platyhelminthes) infers evolution of diet in parasitic flatworms. *International Journal for Parasitology* 40: 1237-1245. doi: 10.1016/j.ijpara.2010.02.017

6. Hutson, K.S. 2009. Marine Parasites. URL: www.marineparasites.com

7. Kuchta, R., Scholz, T. & Justine, J.L. 2009. Two new species of *Bothriocephalus* Rudolphi, 1808 (Cestoda: Bothriocephalidae) from marine fish off Australia and New Caledonia. *Systematic Parasitology* **73,** 229-238.

8. Perkins E.M., Donnellan S.C., Bertozzi T., Chisholm L.A. & Whittington I.D. 2009. Looks can deceive: molecular phylogeny of a family of flatworm ectoparasites (Monogenea: Capsalidae) does not reflect current morphological classification. *Molecular Phylogenetics & Evolution* 52: 705-714. doi: 10.1016/j.ympev.2009.05.008.

9. Repulles-Albelda A., Montero F.E., Holzer A.S., Ogawa K., Hutson K.S. & Raga J.A. 2008. Speciation of *Paradeontacylix* spp. (Sanguinicolidae) in *Seriola dumerili*. Two new species of the genus *Paradeontacylix* from the Mediterranean. *Parasitology International* **57**, 405-414.

Up and coming...

10. Miller, P., Fitch, A., Gardner, M., Hutson, K.S., & Mair, G. (submitted, 28th Feb 2011). Genetic population structure of Yellowtail Kingfish (*Seriola lalandi*) in temperate Australasian waters inferred from microsatellite markers. (*Aquaculture*).

In preparation...

11. Hutson, K.S., Perkins, E.M., Holzer, A.S. & Whittington, I.D. (in prep). *Paradeontacylix* n. sp. (Digenea: Aporocotylidae) infects three *Seriola* spp. (Perciformes: Carangidae) in three oceans.

12. Hutson, K.S. & Boxshall, G.A. (in prep). Redescription of an atypical species of Trebiidae, *Kabataia ostorhinchi* Kazatchenko, Korotaeva & Kurochkin, 1972 from *Oplegnathus woodwardi* (Waite) (Perciformes: Oplegnathidae) off Port MacDonnell, South Australia.

13. Hutson, K.S., Bott, N.J., Hayward, C.J., Bray, R.A, Whittington, I.D. (in prep). Three new aporocotylids (*Cardicola* spp.) from southern Australia with a redescription of *Cardicola whitteni*.

14. Hutson, K.S., Perkins, E., Boxshall, G. and Whittington I.D. (in prep). Ectoparasites (Monogenea and Copepoda) infecting fishes off Port MacDonnell, southern Australia.

15. Hutson, K.S. and Whittington, I.D. (in prep). Parasite risk assessment for sea-cage aquaculture of mulloway (*Argyrosomus japonicus*) and barramundi (*Lates calcarifer*).

Theses

16. Perkins E.M. 2010. Family ties: molecular phylogenetics, evolution and radiation of flatworm parasites (Monogenea: Capsalidae). PhD thesis, School of Earth & Environmental Sciences, The University of Adelaide, 198 pp.

17. Catalano, S.R. 2009. Parasite assemblages of the Arripidae in southern Australian waters. School of Earth and Environmental Sciences, The University of Adelaide, Honours thesis. 69 pp.

18. Brock, E.L. (2010). Metazoan parasite diversity of King George whiting (*Sillaginodes punctatus*): examination of host ontogenic and spatial variation. School of Earth and Environmental Sciences, The University of Adelaide, Honours thesis, 52 pp.

14.4 Appendix 4: Presentations

14.4.1 International conference presentations

1. Hutson, K.S., Catalano, S.R. & Whittington, I.D. 2010. Aquaculture in hot water: emerging parasitic threats to the Australasian finfish industry. 10th International Congress for Parasitology, 16-20th August, Melbourne, Australia.

2. Catalano, S.R., Hutson, K.S. & Whittington, I.D. 2010. Back to basics: rigorous host and parasite identification in commercially and recreationally important arripid fish. 10th International Congress for Parasitology, 16-20th August, Melbourne, Australia.

3. Brock, E.L., Whittington, I.D., Hutson, K.S. & Steer, M.A. 2010. Metazoan parasite diversity of King George whiting (*Sillaginodes punctatus*): examination of host ontogenic and spatial variation. 10th International Congress for Parasitology, 16-20th August, Melbourne, Australia.

4. Hutson, K.S. & Whittington, I.D. 2007. Blood flukes infecting wild yellowtail kingfish (*Seriola lalandi*) and Samson fish (*S. hippos*): implications for *Seriola* aquaculture in Australasia. *Parassitologia* **49**, p 61 (presented at the 7th International Symposium on Fish Parasites, 24-28th September, Voterbo, Italy).

5. Repulles-Albelda, A., Montero, F.E., Holzer, A., Ogawa, K., Hutson K.S., Raga, J.A. 2007. Speciation of *Paradeontacylix* spp. (Trematoda, Sanguinicolidae) from *Seriola dumerili*. Phylogeny of the *Paradeontacylix* genus. *Parassitologia* **49**, p 324 (presented at the 7th International Symposium on Fish Parasites, 24-28th September, Voterbo, Italy)..

14.4.2 National conference presentations

1. Hutson, K.S., Catalano, S.R. & Whittington, I.D. (2010). Attract, aggregate and accumulate: wild fishes as potential parasite reservoirs for sea-caged stocks. Australasian Aquaculture 25th May, Hobart.

2. Hutson, K.S. and Whittington I.D. 2009. Where the wild things are: predicting which parasites from wild fish may be problematic in sea-cage aquaculture. FRDC Aquatic Animal Health Subprogram (AAHS) meeting, Cairns, July 2009.

3. Brock, E.L, Hutson, K.S. and Steer, M.A. 2009. Parasite assemblages as indicators of population structure of southern garfish (*Hyporhamphus melanochir*). FRDC Aquatic Animal Health Subprogram (AAHS) meeting, Cairns, July 2009.

4. Hutson, K.S, Brock, E.L, and Steer, M.A. 2009. Parasite assemblages as potential population indicators for southern garfish *Hyporhamphus melanochir*. Australian Society for Parasitology, Sydney, July 2009.

5. Catalano, S.C., Hutson, K.S. and Whittington, I.D. 2009. Picky parasites: patterns of infection in three important endemic fishes (Arripis spp.) Australian Society for Parasitology, Sydney, July 2009.

6. Hutson, K.S. and Whittington I.D. 2008. Potentially pathogenic parasites for finfish sea-cage aquaculture. Australian Society for Fish Biology, Bondi Beach, New South Wales, 15 September 2008.

7. Hutson, K.S. and Whittington I.D. 2008. Potentially pathogenic parasites for finfish sea-cage aquaculture. Australasian Aquaculture, Brisbane, 5th August, 2008.

8. Hutson, K.S. and Whittington I.D. 2008. Risk assessment for parasites in South Australian sea-cage aquaculture. Australian Society for Parasitology, Adelaide, 7th July 2008.

9. Hutson, K.S. and Whittington I.D. 2008. Potentially pathogenic parasite for finfish sea-cage aquaculture' Australian Society for Parasitology and the ARC/NHMRC network for parasitology, Glenelg, South Australia, 7th July 2008.

10. Hutson, K.S. & Whittington, I.D. 2007. 'Why little guys matter! A new species of blood fluke infecting wild yellowtail kingfish (*Seriola lalandi*) in southern Australia'.

ARC/NHMRC Network for Parasitology and Australian Society for Parasitology Annual Conference, Canberra, 8-11th July, 2007

14.5 Appendix 5: Public outreach

1. Hutson K.S. 2010. 'Marine Parasitology' *Scope*, Channel Ten. 25 September 2010.

2. Biodiversity Gallery, South Australian Museum, 2010. Contribution of specimens, photographs and video footage to the 'Biodiversity Gallery' a new, permanent exhibit at the South Australian Museum.

3. Whittington I.D. 2010. Discussions with members of the public over the opening weekend of the 'Biodiversity Gallery', South Australian Museum, 13-14th February 2010.

4. Hutson K.S & Whittington I.D. 2009. 'How to be a parasitologist' Junior Field Naturalists Society, Adelaide, 28th May 2009.

5. Hutson K.S. & Hutson G.D. 2009. 'In search of Charles Darwin' The South Australian Museum and Royal Society of South Australia, 12th March 2009.

6. Hutson, K.S. 2008. 'Problematic parasites for sea-cage aquaculture of yellowtail kingfish (*Seriola lalandi*) and mulloway (*Argyrosomus japonicus*).' New South Wales Fisheries, February 14th 2008, Port Stephens, New South Wales, Australia.

7. Hutson, K.S. 2008. 'Potentially problematic parasites.' Recfishing Research Steering Committee Meeting 7, Adelaide, 27-28th March 2008, Adelaide, Australia.

8. Hutson K.S. 2008. 'Parasites of recreational and aquaculture fishes' South Australian Research and Development Institute, 22nd May 2008, Adelaide, Australia.

9. National Science Week. 2008. Faces of Science: Dr Kate Hutson, University of Adelaide, The South Australian Museum.

10. Peddie, C. 2008. The Advertiser Review. Gone Fishing: Dr Kate Hutson (marine biologist). Saturday, 9th August 2008.