

# **Department of Ichthyology and Fisheries Science**

## **Research Report Series No 23**



Photograph by Carl Huchzermeyer ©



**RHODES UNIVERSITY**

Department of Ichthyology and Fisheries Science  
P.O. Box 94, Grahamstown 6140

**October 2011**  
(Version 1)

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**Edited by: CLW Jones & T Moss**

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**Wednesday 5 October 2011**

**08h50-09h00: Welcome by Prof. Tony Booth (Acting Head of Department)**

**Honours seminars (Chair: Dr CLW Jones)**

**09h00-09h20: Devin Ayres (BSc Honours) - The use of effluent-grown algae as an alternative dietary protein source for *Oreochromis mossambicus* (Peters 1852): Effect on reproductive output.**

Supervisor/s: Prof. H Kaiser ([h.kaiser@ru.ac.za](mailto:h.kaiser@ru.ac.za)); Dr CLW Jones ([c.jones@ru.ac.za](mailto:c.jones@ru.ac.za))

Funder/s: Water Research Commission (K5/2008); South African Breweries Ltd (SAB); THRIP; Rhodes University JRC

Many of the studies on the reproductive output or performance of tilapiine fishes involved the effects of dietary protein level on reproduction. Little is known about how algae replacement diets influence reproductive output. Mozambique tilapia, *Oreochromis mossambicus*, are omnivorous and therefore utilise proteins obtained from plants, insects and fish. We hypothesised that algae replacement will not have an effect on reproductive output because algae form part of their natural diet.

The objectives of this research project were to compare fecundity, relative fecundity, gonadosomatic index (GSI), maturity stages and size of eggs in *O. mossambicus* fed on diets containing brewery effluent-grown algae. Diets were assigned as follows: Diet A was the control diet with 0 % of the fish meal portion of the diet substituted with protein from algae; Diets B and C had 40 % and 80% of the fish meal portion of the diet substituted with protein from algae, respectively.

Adult *O. mossambicus* (100 g fish<sup>-1</sup> ± 40 SD) were conditioned in recirculating systems. Males and females were kept separately in two systems and fed 3% of their body weight per day of the experimental diet to which they were assigned. Three isonitrogenous diets (35% protein) were formulated with fishmeal, soya meal and algal meal as protein sources. Algae were collected from South African Breweries Ltd (SAB) in Port Elizabeth. The control diet contained a protein formulation (35% protein), with fishmeal accounting for 53.2 % and soya for 40 % of the protein fraction. The other two diets were prepared with substitution levels of algae for fishmeal on an equivalent protein basis, 40% and 80% of the fishmeal protein were replaced with algae respectively. After 11 weeks of conditioning males and females were removed from the tanks and their GSI, HSI values and numbers and sizes of eggs determined.

There was no significant difference between treatments in average GSI, egg length, egg width, egg count and maturity stages (analysis of variance,  $p >> 0.05$ ). The high cost of constantly supplying fish meal, requires the incorporation of alternative protein sources in fishmeal diets for sustainable growth of the industry. As the algal replacement did not reduce the reproductive output such diets could be used as alternative tilapia feed and substitute commercial fish meal-based diets.

**09h20-09h40: Roy Bealey (BSc Honours) - Developing a precise method for ageing *Oreochromis niloticus* from Lake Manyame, Zimbabwe**

Supervisor: Dr OLF Weyl ([o.weyl@saiab.ac.za](mailto:o.weyl@saiab.ac.za))

Funder/s: The CIB DST-NRF Centre of Excellence for Invasion Biology; National Research Foundation/SAIAB

Nile Tilapia (*Oreochromis niloticus*) have been widely distributed by humans beyond their native range. The species is considered highly invasive and understanding its biology and ecology is a fundamental requirement for its management and potential control. Accurate and precise age estimates are a fundamental requirement for biological studies. As a result, there is a need to determine which hard structures and preparation methods are most appropriate yield the most precise estimates. The aim of this study was therefore to compare the precision gained from otoliths and scales for ageing Nile tilapia from Lake Manyame, Zimbabwe.

Samples were collected from Lake Manyame between December 07 and December 09 using a combination of angling, spearfishing and purchase from commercial gill net fishermen. A sample size of 368 specimens was collected with both sagittal otoliths being removed from each fish and a scale removed from the same region of each fish's body following a measurement of total length to the nearest millimetre. Scales were read as whole structures while all otoliths were sectioned before reading. One of the otoliths from each fish was burnt before sectioning to determine whether this method improved the ease with which growth rings could be interpreted.

Age readings were conducted using a compound microscope under transmitted light and structures were read multiple times to allow intra reader comparisons of precision. Three readers were involved in the structure reading process, each of which had differing levels of experience in obtaining age estimates from hard structures, so that inter reader analysis could be done for each structure and method. Statistical analysis of the results included the use of Average Percent Error (APE), an Index of Practicality and Precision (IPP) and various graphical methods which allow the interpretation of variation between readings and readers. A comparison of results with the Von Bertalanffy growth model will provide an indication of the potential accuracy of age estimates using a length at age relationship.

Average Percent Error (APE) was higher for unburnt otoliths (APE = 0.316) than for burnt otoliths (APE=0.271) indicating greater inter reader precision obtained for readings of burnt otoliths. Analysis of scale readings indicates a lower level of precision than that obtained using otoliths but scales can be used to provide age information without sacrificing the fish. Sectioned, burnt otoliths therefore appear to be the most appropriate structure for ageing this species.

**09h40-10h00: Simon Calderwood (BSc Honours) - The effect of sex sorting on somatic growth and reproductive investment of farmed abalone (*Haliotis midae*)**

Supervisor: Dr CLW. Jones (c.jones@ru.ac.za)

Funder: HIK Abalone Farm (Pty) Ltd; Aquafarm Development (Pty) Ltd; Roman Bay Sea Farm (Pty) Ltd; THRIP

Farmed abalone growth rate and canning yields decline at certain times of the year in sexually mature stock. This may be associated with gonad development and spawning, as the peak spawning months of abalone coincide with this decline. Investment in gonad development reduces somatic growth of the abalone impacting the canning of abalone since the gonad is discarded during processing. An investigation was undertaken to determine if sex separation of cultured abalone improves somatic growth and effects gonad development.

Abalone of a similar age and size were sex sorted and either males, females or unsexsorted abalone were stocked into farm tanks. Each of these treatments were represented in three tanks, stocked at 9.6 kg/tank with 56-64g abalone, and subject to farm production conditions. Abalone growth and gonadosomatic index (GSI) were monitored for 149 days, with a weigh and measure at the start, after 70 and at the end.

Sex sorted abalone had a higher meat mass and higher percentage meat yield compared to non-sex sorted abalone at 70 days, but not at the start or at the end of the trial (multifactor ANOVA:  $p=0.00017$  and  $p=0.00083$  for whole weight and meat yield, respectively). The GSI was estimated and did not differ between treatments ( $p>0.05$ ). However, a significant difference was found in visceral mass and percentage visceral yield (note: viscera included gonad and other tissue combined); non-sex sorted abalone had a higher percentage visceral mass than sex sorted abalone ( $p=0.00007$ ). Sex sorted abalone also had a higher condition factor than non-sex sorted abalone ( $p<0.00001$ ). Both these differences were apparent at 70 days, but not at the start or finish of the trial.

Within the mixed sex treatment, differences were found between males and females at 70 days. Females had a significantly greater visceral mass than males ( $p=0.0016$ ). Similarly, males had a greater percentage meat yield and higher condition factor than females ( $p=0.005$  and  $p=0.000007$  for meat yield and condition factor, respectively).

Sex sorting abalone produced a higher percentage meat yield, higher condition factor and a lower percentage visceral mass than non-sex sorted abalone, over a limited period of the trial only. It appears that sex sorting might improve production over times of year when abalone would normally invest in gonad production. This remains to be verified in future work.

**10h00-10h20: Zyangani Chirambo (BSc Honours) – A preliminary investigation of the species composition and the population structure of selected species in a man-made saltwater impoundment; Marina Martinique, Jeffreys Bay**

Supervisor/s: Mr MTT Davies ([m.davies@ru.ac.za](mailto:m.davies@ru.ac.za)); Dr WM Potts ([w.potts@ru.ac.za](mailto:w.potts@ru.ac.za))

Funder/s: Rhodes University JRC; Home Owners Association-Marina Martinique

Marina Martinique is a 14 ha enclosed and closed saltwater impoundment constructed in 1990. The fishery in the impoundment is currently unmanaged and there is no information on the fish species composition or population structure. Understanding an ecosystem requires studying part or all aspects of that system. This is crucial when designing effective management tools for both public and privately managed water bodies. In order to provide a fisheries management plan for the homeowners association of Marina Martinique, it was necessary to conduct a basic assessment of the fish populations.

The aim of the project was to obtain information required to develop a fishery management plan for the Marina Martinique. The objectives were to:

- Determine the fish species composition;
- Determine the size composition of the important angling species;
- Estimate the population size of selected species; and
- Estimate the relative abundance (CPUE) of the fish species.

Three two-day sampling trips (March, June, September 2011) were conducted. A variety of gears including seine nets, fyke nets and hook and line was used to capture fish in ten different sites. Fish were measured, weighed and either tagged with hallprint dart tags or fin clipped and injected with oxytetracycline.

A total of 1549 fish belonging to 13 species and 8 families were captured. The sparids *Diplodus sargus capensis* (n = 845) and *Sarpa salpa* (n = 520) dominated the catches, while important fishery species such as *Lithognathus lithognathus* (n = 8) and *Pomadasys commersonnii* (n = 7) were rarely captured. The length frequencies of the dominant species (*Diplodus sargus capensis* 73 - 296 mm FL, *Sarpa salpa* 124 - 270 mm FL) revealed several cohorts, but only large specimens of the other species were captured.

A total of 325 fish were tagged and 55 were fin clipped. Of these five (three were Cape stumpnose (*Rhabdosargus holubi*), one blacktail (*D. capensis*) and one zebra seabream (*D. hottentotus*)) were recaptured. One of the Cape stumpnose was recaptured at the original capture site, while 4 moved considerable distances (650 and 800 m). Using the Schnabel mark recapture model, the population size of *Rhabdosargus holubi* was estimated to be approximately 282 (132 LCV - 432 UCV) individuals.

The Marina is presently dominated by smaller less desirable angling species that may be reproducing in, or recruiting into the marina. It appears that the important angling species that were captured in large numbers historically, have been overexploited and restocking programs with a predominantly release fishery may be favoured to optimise angling in the long term. However, with a shortage of fodder species (e.g. Mugilidae), stocking of important predatory fish should be preceded by the stocking of prey species.

**10h20-10h55: Tea break**

## **Honours seminars continued (Chair: Dr CLW Jones)**

### **11h00-11h20: Chenelle de Beer (BSc Honours) - Validating and solving technical problems in the interpretation of individual growth histories using fish otoliths.**

Supervisor: Dr W Potts ([w.potts@ru.ac.za](mailto:w.potts@ru.ac.za))

Fish age and growth is regarded as the most significant of biological variables and is key for stock assessment models as it forms the basis for a variety of calculations including mortality rates and productivity. Otoliths are the preferred bony structures used for estimating population growth. However, a proposed new technique of otolith interpretation (increment measurement) may be a potentially powerful “tool” to provide individual fish growth history information. Although this “tool” would allow the prediction of fish growth rates, its accuracy, precision and validity has not been investigated.

The aim of the study was to validate the technique and assess the technical accuracy of the individual growth increment method. Objectives were as follows:

- Assess the relationship between otolith width and individual fish growth history;
- Investigate the relationship between the mean increment widths and mean population growth;
- Evaluate the technical accuracy, including both precision and readability, in order to develop a protocol for future studies.

Two groups of known age (1 and 2 years) *Argyrosomus japonicus* (n = 47) were obtained from Espadon Marine. Fish were measured, otoliths removed and lengths obtained. Otoliths of young (1-4 years) *Clarias gariepinus* from slow (Ngotwane Dam, n = 19) and fast growing (Darlington Dam, n = 24) populations were taken from pre-existing collections. Otoliths were sectioned (0.4mm) through the centre of the nucleus using standard techniques. *A. japonicus* otoliths were also sectioned approximately 0.6mm from the centre of the nucleus. Sections were mounted onto slides and photographed under transmitted light using a dissection microscope. MotiC software was used to measure the distance from the centre of the nucleus to the edge of the otolith in *A. japonicus* and the width of all growth increments in *C. gariepinus*.

Although there was no significant difference ( $p=0.08$ ) in increment width, between the side and centre sections of *A. japonicus*, interpretation was possible for 46% and 98% of the sections, respectively. There was a positive, strong relationship between the otolith and fish length (Coefficient of determination,  $R^2 = 0.83$ ) and a positive, weak relationship between nucleus to edge measurement and fish length ( $R^2 = 0.26$ ). The Average Percentage Error (APE) was 2.87, indicating a high intra-reader precision.

The mean corresponding otolith increment widths were greater, although not significantly so (t-test with unequal variances,  $p > 0.05$  in all cases) for *C. gariepinus* from the slower growing (Ngotwane Dam) population.

The readability of otoliths using this technique appears to be dependent on the sectioning process. An appropriate sectioning protocol has been developed to improve the readability of otoliths. It appears that increment measurements do have application for interpreting the individual growth histories of *A. japonicus* and increment width measurements for *C. gariepinus* were representative of population growth rates. Pilot studies are, however, essential to determine the suitability of this technique for each study species.

**11h20-11h40: Kerthu Shipoke (BSc Honours) - Effect of diet composition on the gut structure of *Haliotis midae***

Supervisors: Prof. PJ Britz (p.britz@ru.ac.za); Mr J Kemp ([groovypalm@gmail.com](mailto:groovypalm@gmail.com))

Funder/s: Rhodes University JRC; HIK Abalone Farm (Pty) Ltd

The South African abalone aquaculture industry has rapidly developed with production reaching approximately 1000 tons per year since commercial operations commenced in the 1990s and the trend is expected to continue. Abalone farmers feed either seaweed and artificial feed or a combination of both. However, little is known about the South African abalone digestive tract and its digestive process which limits our ability to improve feed formulations. Therefore, an experiment was designed to investigate the cellular structure of the digestive system of the South African abalone and to test whether different diets have an effect on the abalone gut cell structure.

The experiment consisted of three treatments; kelp, Abfeed and combined Abfeed/kelp diet. Farm reared *Haliotis midae* with an average shell length of 52.3 mm were fed for six months on Abfeed and kelp, and four months on combined diet. Three abalone samples with a length of 67-70 mm were selected from each treatment. Tissue samples taken from the crop, stomach and digestive gland regions were prepared using the procedures highlighted in Cross *et al* 2001 for light microscopy and electron microscopy viewing. Histological observations were done using light microscopy, transmission electron microscopy (TEM) and scanning electron microscopy (SEM). Observations of the crop (the first part of the stomach that receives lubricated food from the oesophagus) revealed that the epithelial cells are coated with microvilli. Further observation revealed the occurrence of phagocytes, iron granules, fragmentation spherules and secretory cells in *Haliotis midae* fed all three treatments. Secretory cells appeared to be more prominent on the crop epithelial cells of the kelp fed abalones compared to those fed Abfeed and the combined diet. The crop epithelial cells of the kelp fed abalone were coated with mucus which appeared to be absent in those fed on the Abfeed and combined diet. The stomach, which receives the coarse food particles from the crop, mainly possessed secretory cells, iron granules as well as a thick layer of gastric shield.

The study successfully established a methodology for studying abalone gut wall structure. SEM was suitable for viewing the gut surface. However, TEM proved impractical for viewing the cellular structure as it was difficult to cut sections in the right position, and the high-resolution of the images made it difficult to locate the correct areas to view. Thus light microscopy was used to view gut wall sections stained with 1% toluidine blue in Na<sub>2</sub>CO<sub>3</sub>. The descriptive information obtained from the sections was not sufficient to conclude that there were or were not significant differences in the gut structures of abalone fed with a formulated diet (Ab-feed) and the one fed on fresh macro algae (kelp). Therefore, further investigation is required to study the morphology of the epithelial cells as well as the absorptive cells.

**11h40-12h00: Kerry-Ann van der Walt (BSc Honours) - Species identifications in South Africa's ornamental trade – how accurate are the names?**

Supervisors: Dr E Swartz (ernst.swartz@gmail.com); Dr T Mäkinen (temakine@gmail.com)

Funder: South African Institute for Aquatic Biodiversity (SAIAB)

Many of the freshwater species used for the ornamental trade in South Africa may be imported or sold under the incorrect common or scientific names. The ornamental fish trade in South Africa is currently regulated by a permitted list to prevent unwanted species to establish and become invasive in natural environments. In this study, it was tested whether species in the ornamental fish trade can be identified through DNA barcoding by comparing their DNA sequences to a reference library known as the Barcode of Life Data System (BOLD), and whether they are being sold under accurate names. The aim is to develop tools to help prevent illegal freshwater fish species from entering the country and to assist the trade with quality control.

Two hundred specimens (100 species, two replicates per species) were purchased from aquarium traders. Live specimens were anesthetized with an overdose of clove oil. A sample of muscle tissue was removed and a photograph was taken of each specimen for submission to the BOLD database. The remaining specimens were fixed in formalin and serves as vouchers. Species identifications were based on the common names provided by the traders for which scientific names were found using the Catalog of Fishes (California Academy of Sciences) and FishBase. A total of 190 specimens were sent to the Canadian Centre for DNA Barcoding at the University of Guelph (Canada) for extraction, PCR and sequencing of the mitochondrial COI gene (the standard marker for vertebrates). A total of 93 specimens have been successfully sequenced thus far. These were used to construct a neighbour joining tree based on the Kimura 2-parameter distance model.

Of the 93 specimens, 77 (83%) matched identifications by other researchers in BOLD. Two specimens did not match and were not closely related to any other fishes in BOLD. One specimen is traded under the non-specific “jewel cichlid” that could refer to several different species. Two specimens could not be identified yet, due to colour morph confusion. In addition, the neighbour-joining tree revealed a cluster of Malawi cichlids that could not be identified with confidence (8 specimens), probably due to recent rapid evolution. Only 3% of the specimens are possibly traded under wrong common names that could cause confusion in terms of quality control and/or invasive potential. The results of this study therefore shows that DNA Barcoding can be a useful tool to identify imported freshwater fishes with further development of the BOLD database, except in the case of rapidly evolving groups such as the Malawi cichlids.

## **Freshwater aquaculture and effluent treatment (Chair: R Bennett)**

**12h00-12h20: Rory Scheepers (MSc student) - Treated brewery effluent as a water source for the culture of the ornamental fish *Xiphophorus helleri* (swordtail)**

Supervisor/s: Prof. H Kaiser (h.kaiser@ru.ac.za); Dr CLW Jones (c.jones@ru.ac.za)

Funder/s: South African Breweries Ltd; Water Research Commission (K5/2008); THRIP; Rhodes University JRC

Large industries utilize and discard large volumes of effluent, globally. This “polluted” water is sent to the local municipalities for treatment, at costs to the industry and the environment. The aim of this work is to establish if alternative, environmentally sustainable water treatment methods such as high rate algal ponding and wetlands can be used to treat brewery effluent to a standard suitable for fish culture.

The number of juvenile fish harvested in the treated brewery effluent and municipal water control did not differ significantly between treatments, with means  $631 \pm 11$  and  $660 \pm 26$  juveniles per month ( $p=0.99$ ). There were no significant differences in brood size ( $32.6 \pm 2.2$  and  $34 \pm 4.2$ ) for treated brewery effluent and municipal water systems ( $p=1.0$ ). Similarly, the mean number of broods did not differ significantly with  $5.6 \pm 0.20$  and  $4 \pm 2.11$  and for the effluent and municipal systems ( $p=1.0$ ).

There were no significant difference in standard length and condition factor between juveniles in two treatments at the start of the trial, with means of 15.3 mm and 0.0016 respectively ( $p<0.05$ ). Similarly, there were no significant difference in fish length and weight between treatments after 91 days, with overall means of 44.5 mm and 1.29 g ( $p=0.68$  and  $p=0.85$  for length and weight respectively), and there was difference in mean weight gained ( $p=0.73$ ).

For sexually mature fish, there was no interaction between time, fish gender and water source ( $F_{(1,32)}=0.03$ ,  $p=0.86$ ), water source and gender ( $F_{(1,32)}=1.30$ ,  $p=0.26$ ), time and gender ( $F_{(1,32)}=2.17$ ,  $p=0.15$ ) and there was no interaction between time and water source ( $F_{(1,32)}=0.77$ ,  $p=0.39$ ). Mean weight of fish at the start ( $2.27 \pm 0.50$ g) was significantly different from the mean weight at the end ( $2.93 \pm 0.75$ g) ( $t=-3.276$ ,  $p=0.0002$ ), indicating that mature fish continued to grow in both systems. The mean weight of the males ( $2.16 \pm 0.52$ g) was significantly different to that of the females ( $3.04 \pm 0.59$ g) ( $t=-5.002$ ,  $p=0.00013$ ).

The similarity of fish size, condition factor and reproductive output between swordtails cultured in the different treatments, suggests that treated brewery effluent may be a suitable water source for the culture of swordtails and can be used to culture fish without negative effects, provided mitigating measures are adopted to ensure that fish health is not affected by unscheduled upstream dumping of brewery waste.

In addition, reproductive biology and health was monitored. Comparisons in the gill, liver and kidneys histology are currently being carried out. Egg size, yolk volume, yolk density, and yolk lipid content, and fecundity is also being compared between treatments. Water quality still needs to be analyzed to determine if differences in the levels of phosphate, nitrate, nitrite, ammonia, pH, dissolved oxygen, chemical oxygen demand and water temperature were observed between treatments.



## **12h20-12h40: Lara Crous (MSc student) – Application of constructed wetland technology in the treatment of brewery effluent**

Supervisors: Mr MTT Davies (m.davies@ru.ac.za); Prof. PJ Britz (p.britz@ru.ac.za)

Funder/s: South African Breweries Ltd; Water Research Commission (K5/2008); THRIP; Rhodes University JRC

The use of constructed wetlands (CW) has been established as a method for treating wastewaters and offers a low cost and eco-friendly alternative to conventional treatment systems. Brewery effluent in particular is characterized by a high organic nutrient content and offers the potential for biological treatment. The aim of this study was to assess the use of the CW in the treatment of brewery effluent to achieve an environmental discharge standard.

1. How many meters of CW is required to treat nutrient levels in brewery effluent to discharge standards?
2. Is the CW capable of treating water directly from the AD?

A 60m<sup>2</sup>, four celled planted horizontal subsurface CW was built to treat post HRAP water. Water samples were taken from the inlet and every 15m. Onsite analysis of Chemical oxygen demand (COD), NH<sub>4</sub>, NO<sub>2</sub>, NO<sub>3</sub>, PO<sub>4</sub>, chlorides, pH was conducted on a weekly basis. Multiple regression indicated COD and NH<sub>4</sub> reached discharge standards after 30m (p-values 0.0056 and 0.013 respectively). NO<sub>2</sub> and NO<sub>3</sub> were within the DWAE limit of 6mg/L and 10mg/L from the inlet and also showed a significant decrease after 30m (p-values 0.049 and 0.002 respectively). There was no statistically significant decrease in PO<sub>4</sub> along the CW. For replication purposes, the CW was then used as four individual beds. Samples were taken from the inlet, 3m, 6m, 9m along the bed and at the outlet. Multiple regression showed a significant decrease in COD from 118 mg/L to 86 mg/L after 15m (p-value 0.00). This does not meet the DWAE discharge limit, and so a longer CW is required, indicated by the previous result. NH<sub>4</sub>-N (p-value 0.00), PO<sub>4</sub>-P (p-value 0.001), NO<sub>2</sub>-N (p-value 0.001) and NO<sub>3</sub>-N (p-value 0.008) all reached discharge limits within the first 3m.

Using the four beds, a second experiment was conducted comparing the treatment of post AD water with that of post HRAP water. The CW showed an average COD decrease of 36.4% (153 mg/L to 97 mg/L) in post HRAP influent and a 42% decrease in post AD water (178 mg/L to 103 mg/L). A 99.9% decrease in NH<sub>4</sub>-N was recorded in post AD water (40 mg/L to 0.04 mg/L) and by 87% (0.91 mg/L to 0.11 mg/L) in post HRAP influent. The lower inlet value is a result of the pre-treatment in the HRAP system. The CW is therefore effective in ammonia removal in post AD water to within the DWAE limit of 6mg/L. The CW reduced PO<sub>4</sub>-P by 90% (post AD) and 71% (post HRAP) to within DWAE limit of 15mg/L.

The CW proved effective in reducing NH<sub>4</sub>-N, NO<sub>2</sub>-N, NO<sub>3</sub>-N to their respective discharge limits within 3m. Chemical oxygen demand however requires a longer length of 30m to reach discharge standard. The CW also proved capable of treating effluent directly from the AD.

## **12h20-12h40: Anneke Cilliers (MSc student) – The treatment of brewery effluent with high rate algal ponds**

Supervisors: Prof. PJ Britz (p.britz@ru.ac.za); Dr CLW Jones (c.jones@ru.ac.za)

Funder/s: South African Breweries Ltd; Water Research Commission (K5/2008); THRIP; Rhodes University JRC

The imperative to recycle waste has become a global imperative which is increasingly being written into policy, legislation and corporate governance. In a joint development agreement between SABMiller and Rhodes University, a 10-month “proof-of-concept” phase (May 2009 – February 2010) was implemented, followed by an 11-month “optimisation phase” (March 2010 – January 2011). The application of high rate algal ponds (HRAP) and a constructed wetland in the treatment of brewery effluent was tested with the aim of producing an effluent that met the South African Department of Water and Environmental Affairs’ (DWEA) general limits for discharge into a natural water resource.

The objective of the baseline “proof of concept” phase of the experiment (May 2009 – March 2010) was to monitor the seasonal performance of HRAPs. The hydraulic retention time (HRT) fluctuated between 17.29 d and 18.29 d in the primary facultative pond (PFP) and between 11.16 d and 12.00 d in HRAPs. The pH of the system increased as brewery effluent moved through the pilot plant AD ( $7.57 \pm 0.03$ ) into the PFP ( $8.24 \pm 0.03$ ) and into the HRAPs ( $10.05 \pm 0.03$ ). The mean chemical oxygen demand (COD) concentration increased from  $130.12 \pm 6.94$  mg/L (post-AD) to  $171.21 \pm 7.99$  mg/L (post-HRAP). The presence of algal cells contributed to higher COD values post-HRAP. The mean ammonia ( $\text{NH}_4\text{-N}$ ) concentration was lowered from  $46.59 \pm 2.47$  mg/L  $\text{NH}_4\text{-N}$  (post-pilot plant AD effluent) to  $1.08 \pm 0.12$  mg/L  $\text{NH}_4\text{-N}$  (post-HRAP effluent). The nitrite ( $\text{NO}_2\text{-N}$ ) concentration remained below one mg/L  $\text{NO}_2\text{-N}$  in post-pilot plant AD, post-PFP and post-HRAP effluent. The mean phosphate concentration in decreased from  $29.81 \pm 1.39$  mg/L  $\text{PO}_4\text{-P}$  (post-AD) to  $17.30 \pm 1.16$  mg/L  $\text{PO}_4\text{-P}$  (post-HRAP). The salt concentration increased as effluent moved through the AD, PFP and HRAPs.

The objective of the optimization phase of the experiment (2 March 2010 – 16 January 2011) was to manipulate the HRT to achieve the maximum treatment rate that met the DWEA general limits for discharge into a natural water resource. Nitrogen removal efficiency was used as an indicator of nutrient removal success. The optimal HRT for autumn was 4.30 d at a temperature of 20.53 °C in HRAP A2 (heated) and 18.96 °C in HRAP B2 (ambient). The optimal HRT for summer was 2.74 d at a temperature of 29.90 °C in HRAP A2 (heated) and 26.36 °C in HRAP B2 (ambient). The mean COD concentration was lowered from  $152.33 \pm 4.85$  mg/L (post-AD effluent) to a mean COD concentration of  $95.00 \pm 3.75$  mg/L (HRAP A2) and to  $100.82 \pm 5.93$  mg/L (HRAP B2). The incoming  $\text{NH}_4\text{-N}$  concentration decreased from a mean of  $42.53 \pm 1.38$  mg/L (post-AD effluent) to  $1.70 \pm 0.81$  mg/L (post-HRAP effluent). The main methods for  $\text{NH}_4\text{-N}$  removal were probably through  $\text{NH}_4\text{-N}$  volatilization at high pH in winter and through algal uptake at higher algal productivity and shorter HRTs in autumn and summer. The HRT was the main factor that influenced algal productivity. A shorter HRT caused higher algal productivity. The HRT was determined by season

## **13h00-13h55: Lunch break**

## **Marine Ecology, conservation and related biology (Chair: T Moloi)**

### **14h00-14h20: Tshoanelo Moloi (PhD student) - Molecular systematics and antifreeze biology of sub-Antarctic notothenioid fishes.**

Supervisor/s: Dr M Mwale (m.mwale@saiab.ac.za); Mr O Gon (o.gon@saiab.ac.za); Prof. C-H Cheng (c-cheng@uiuc.edu).

Funders: South African National Antarctic Program/National Research Foundation; United State National Science Foundation-Office of Polar Program grant; South African Institute for Aquatic Biodiversity (SAIAB).

Notothenioidei is a suborder of perciform fish dominating the cold water of the Southern Ocean. These fishes have evolved antifreeze glycol-protein (AFGP) in their blood and body fluids that enable them to survive in cold environments. The suborder consists of eight families which have wide-ranging species distributed north and south of the Antarctic Polar Front. Previous studies split widespread notothenioid species into separate species or subspecies; eg. *Notothenia coriiceps* was divided into *N. neglecta* in the south Atlantic islands and *N. coriiceps* in south Indian Ocean islands. Studies of the antifreeze attributes of sub-Antarctic species also give a diverse picture in different species. Since there have been disagreement on the taxonomic status and lack of information concerning the status of the antifreeze attributes of sub-Antarctic notothenioids, there is a need to use a new approach resolve this situation. This phase of the study aims to characterize the AFGP attributes of sub-Antarctic notothenioid species. The specific objectives include:

1. Characterisation of serum antifreeze activities in terms of: (a) thermal hysteresis using a Cifton Nanoliter osmometer; and (b) total serum osmolality (using a Wescor freezing point osmometer).
2. Molecular analysis of the antifreeze glycoprotein (AFGP) gene using the Southern and Northern blot procedures.

Blood and tissue samples of a number of sub-Antarctic notothenioid species have been collected from the Atlantic Sector (ICEFISH 2004 cruise), Marion Island (2009 and 2011 relief cruise) and Antarctic Peninsula (American collaborator). This includes about 19 species from 10 sub-Antarctic genera: *Bovichthus*, *Cottoperca*, *Dissostichus*, *Gobionotothen*, *Lepidonotothen*, *Notothenia*, *Patagonotothen*, *Parachaenichthys*, *Pseudochaenichthys* and *Champscephalus*. Large molecular weight DNA has been extracted from the samples collected during ICEFISH 2004 and Marion Island 2009 cruise and used for molecular analysis of AFGP. Blood serum was used to determine antifreeze activities. Marion Island 2011 samples are still to be processed. Most sub-Antarctic notothenioids showed low or no AFGP activity in their blood. However, *Notothenia rossii* and *Artedidraco mirus* appeared to have higher AFGP activity in their blood. The DNA of two Patagonotothen species and *D. eleginoides*, showed no detectable hybridization signal. *L. squamifrons* genomic DNA showed very low hybridization signal compared to its sister species *L. larseni* and *L. nudifrons* and also it expresses no functional AFGP in its RNA. This study provides the first report on AFGP attributes of seven sub-Antarctic species.

**14h20-14h40: Pholoshi Maake (PhD student) - Systematics and phylogeography of mormyrid fishes in South Africa**

Supervisors: Dr E Swartz (e.swartz@saiab.ac.za); Mr O Gon (o.gon@saiab.ac.za); Prof. B Kramer (bernd.kramer@biologie.uni-regensburg.de)

Funder: National Research Foundation (SAIAB)

The family Mormyridae comprises of 18 genera and about 200 species of freshwater fishes endemic to tropical Africa. Mormyrids are unusual in having an electric organ that they use for communication and object location. The electric organ signal or discharge (EOD) is species specific and has been used as a taxonomic character in systematic studies of this family. Two genera of mormyrids, each with a single species, have been recognized from South Africa, *Marcusenius pongolensis* (bulldog) and *Petrocephalus wesselsi* (Churchill). Local populations of the bulldog in South Africa exhibited unusual variability in molecular genetics, morphology and EOD, but little is known about the characteristics of Churchill populations. This systematic study aimed to incorporate morphology, genetics and EOD studies to provide a better understanding of the taxonomy, biogeography and phylogenetic relationships of these mormyrid fishes.

During surveys, samples from different river systems or different major branches received higher priority than samples from sites in close proximity. The genetic data, morphology and EODs were analysed using various statistical and phylogenetic packages. Standard DNA isolation methods were used to isolate DNA from muscle tissue preserved in 100% ethanol. Preliminary analyses of mitochondrial DNA cytochrome *b* identified two genetically divergent forms of *Marcusenius* in South Africa. The one form can be linked to the name *M. pongolensis* and is widespread throughout the study area. An undescribed form occurs in the Limpopo River system and the Nseleni/Mhlatuze systems in KwaZulu-Natal. Sequence divergence between the two species was 2% and analysis of the EOD pulses of the two species are congruent with the genetic dataset. The new form is deeper bodied, possibly reflecting adaptation to floodplains and slow moving backwaters. The slim body form of *M. pongolensis* is possibly an adaptation to the faster flowing habitats in which they occur.

The phylogeny of *Marcusenius* and *Petrocephalus* revealed species level differentiation between different river systems. The genetic results confirm morphology and EOD results in identifying a new species of *Petrocephalus* in South Africa that can be distinguished from the widespread *P. catostoma*. *Marcusenius macrolepidotus* is more closely related to the newly discovered *Marcusenius* form than it is to *M. pongolensis* and *M. altisambesi*. The results also indicate that populations from the Congo basin are most likely ancestral to the *Marcusenius* and *Petrocephalus* populations of southern Africa. A separation time of at least 1-5 mya is in agreement with geological data, indicating that the evolution of major river systems have been important in facilitating dispersal and isolation.

**14h40-15h00: Namulawa Victoria Tibenda (PhD student) - Characterizing the digestive system and assessing the nutritional requirements of Nile perch *Lates niloticus* L. 1758**

Supervisors: Prof. PJ Britz (p.britz@ru.ac.za); Dr J Rutaisire (jruta@infocom.co)

Funder: Uganda National Council for Science and Technology (MSI/WAI/1/03/07)

The feasibility of culturing Nile perch (*Lates niloticus*) is being researched as the Nile perch fishery in Lake Victoria has declined, causing a loss of employment and revenue. Knowledge about the morphology of the digestive system and the nutritional requirements of this fish is required to inform the development of appropriate aquaculture feeding technologies. A study was thus undertaken to describe the morphology of the gastro-intestinal tract of Nile perch by means of gross anatomical description, histological, histochemical, scanning and transmission electron microscopy techniques. Morphometric relationships applicable to aquaculture nutrition were derived. Analyses of Nile perch body composition, including fatty acid profiles were performed using standard procedures as a means of inferring the nutritional requirements of Nile perch at different stages of development.

Results revealed that the digestive system of Nile perch is similar to that in other carnivorous fish, with cardiform teeth, stout gill rakers, exceptionally large caecae, a strong striated oesophagus muscularis, gastric glands with numerous oxynto-peptic cells, several absorptive cells in the intestine and a hepato-pancreas. Bacterial cells were observed in the oral cavity, oesophagus and in the stomach. A relatively short gut ( $0.591147 \pm 0.10668$ ) confirmed the carnivorous behaviour of this fish. An equation for the condition factor constant was derived from a data set (weight and length) of 964 samples of Nile perch caught from Lake Victoria, which represented a size range of this species. The mean condition factor was set as 1 by means of constant 41884. The equation for condition factor;  $K = W(g)/L^3(mm) \times 41884$ , is thus recommended to benchmark the condition factor in any future studies of Nile perch. Analysis of the length-mouth gape relationship a directly proportional relationship mouth ( $Y = 0.1302x$ ,  $R^2 = 83.08\%$ ), these results also provided a means of developing appropriate pellet sizes for Nile perch. Proximal analysis showed that the protein content of the body and eggs to be  $66.49 \pm 2.90\%$  and  $39.05 \pm 2.93\%$  respectively, while lipid content in the body was  $6.96 \pm 1.81\%$ , and  $36.56 \pm 0.56\%$  for eggs. Quantities of 16:0, 18:0, 16:1n7, 18:1n9, 18:1n7, 18:3n3, 18:2n6, 20:4n6, 20:5n3, 22:5n3, and 22:6n3 were significant ( $p < 0.005$ ) in the muscle and eggs.

**15h00-15h20: David Kahwa (PhD): Gonad morphology and ultrastructure of the Nile perch *Lates niloticus*: Light scanning and transmission electron microscopy**

Supervisor: Prof. H Kaiser (h.kaiser@ru.ac.za)

Funder: Uganda National Council for Science and Technology, Nile Perch Project

Nile perch, *Lates niloticus*, is a large freshwater predator of commercial importance in the fisheries of Lakes Victoria, Kyoga and Albert, Uganda. However, fisheries production has decreased dramatically in the last decade to warrant the need to seek alternative methods of enhancing its production. The most tangible method is establishment of aquaculture production. Nevertheless, aquaculture management requires information on gametogenesis, gamete type and structure for production planning. Gonad histology provides information on gamete developmental stages. This information is limited for *Lates niloticus*. This study aimed at understanding gonad and gamete type and structure so as to generate technology for the culture of this species.

*Lates niloticus* (n=246) were collected monthly from Lake Victoria Entebbe – Uganda, between May 2008 and April 2009. Gonad tissues were processed for light, scanning and transmission electron microscopy and digital images photographed and analyzed.

The testis of *L. niloticus* is lobular with spermatogonia distributed along the lobule length, hence this is an unrestricted type of testis, typical of teleosts in the order Perciform. The lobule walls were made of myoid cells forming a non-continuous sheet. The lobules were separated by the interstitium with Leydig cells, fibroblast and microphage cells and blood vessels. Leydig cells were commonly found close to blood vessels. Spermatogonia were identified by their large size and presence of electron-dense bodies (nuages) around the nuclear membrane and association with Sertoli cells. Gametogenesis took place in cysts made of a Sertoli cell complex, and each cyst contained isogenic germ cells. Sperm development proceeded through three major phases; spermatocytogenesis, spermatogenesis and spermiogenesis. In *L. niloticus* the latter likely conforms to type II teleost spermiogenesis that is known to produce type II spermatozoa.

Nile perch sperm is simple uniflagellate anacroosomal aquasperm typical of teleosts. It has a round but flattened head with the centrioles perpendicular to one another located outside the *nuclear fossa*. The flagellum is in line with the distal centriole and is placed tangential to the nucleus. There are 5-7 mitochondria localized in a short mid-piece,  $1.68 \pm 0.01 \mu\text{m}$  long. The average length of flagellae was  $29.4 \pm 2.47 \mu\text{m}$  with typical eukaryotic 9+2 doublet of microtubules and an extension forming one lateral wing measuring  $1.08 \pm 0.28 \mu\text{m}$  wide.

The development of Nile perch oocytes was similar to that known from other teleosts, although there was an apparent difference. The absence of yolk vesicle (cortical alveoli) formation was noted. Therefore, we propose modification of classification of development stages in Nile perch.

**15h20-15h35: Tea break**

## **Marine Ecology, conservation and related biology continued (Chair: P Maake)**

**15h40-16h00: Rosanne Thornycroft (MSc student) - Videographic analysis of the coelacanth, *Latimeria chalumnae*, and associated habitats in the iSimangaliso Wetland Park, KwaZulu Natal, South Africa**

Supervisor: Prof. AJ Booth (t.booth@ru.ac.za)

Funder: National Research Foundation

The coelacanth *Latimeria chalumnae* is an iconic, endangered fish. Research within its natural habitat is logistically constrained due to the depths at which coelacanths occur together with strict permitting restrictions. Videography therefore provides alternative “sampling” tool as it is non-invasive and due to modern remote operated vehicle and submersible technology has fewer time and depth constraints.

In order to understanding the coelacanth-specific habitats and quantify its associated fauna submersible video footage obtained in 2003 - 2005 from iSimangaliso Wetland Park’s submarine canyons was analysed.

Canyon habitat was quantified using 50 still frames extracted from video footage collected from each canyon each year. For each still frame, percentage cover was estimated using the software *Coral Point Count with Excel Extensions (CPCe v 3.6)* and species abundance and richness determined. Habitat types were then delineated according to macroinvertebrate and fish abundance patterns using both SHE analysis and Principle Component Analysis (PCA).

Eight different habitats were identified though SHE analysis and PCA. From the PCA, the first two principal components explained ~18% of the variation within the data and the strongest loadings were sponges (0.90) and gorgonians (0.43). When these two components were ordinated three separate invertebrate groups were clearly noticeable; gorgonians, sponges and sea pens. The most distinct habitats when tested with the PCA were found to be sea pens on sandy flats and cave habitats consisting mainly of sponges and bryozoans. Glass sponges were found on chipping boulders in the deep thalweg and were usually alone or accompanied by other forms of encrusting sponges. Sponges were distributed over all habitat types and found in various forms throughout all depths. Gorgonians were present in various forms in three of the habitat types, either accompanied by lollipop-shaped sponges or by a wire corals however there was much overlap between these habitats on the ordination plot.

Future research will attempt to predict coelacanth distribution using Discriminate Function Analysis (DFA) and Logistic regression. While the SHE analysis and PCA provided an exploratory analysis of the data DFA will estimate linear discriminant functions to classify each habitat. Although several habitats are clearly noticeable, the lack of any depth data for the video footage constrains the data analysis.

**16h00-16h20: Reece Wartenberg (MSc student) - Seasonal- and depth-related community dynamics of reef fish assemblages on a large, high latitude coral reef in the Western Indian Ocean**

Supervisor/s: Prof. AJ Booth (t.booth@ru.ac.za)

Funder/s: National Research Foundation

While the relationship between coral-reef fishes and their habitat structure has been rigorously studied the effect of season in structuring reef-fish communities has been overlooked. In South Africa, however, neither of these potential community-level drivers have been investigated. The aim of the study was to determine if possible shifts in ichthyofaunal community structure could be related to trends in season and/or habitat characteristics.

Fish assemblages on a large patch-reef ( $\sim 2 \text{ km}^2$ ), Two-Mile Reef, were surveyed by divers implementing videographic strip-transects. Ninety transects were conducted over two seasons; winter and summer, and three depth ranges; shallow (6 – 14 m), intermediate (14 – 22 m), and deep (22 – 30 m). Assessment of reef characteristics using Principle Component Analysis indicated that depth is implicitly related to habitat complexity and benthic community as depth increases habitat complexity decreases because coral communities shift from dense coral domination to sparse sponge domination, and algal biomass and cover decreases.

A total of 41 families consisting of 209 species and 18172 individuals, dominated by pomacentrids in abundance and labrids in richness, were surveyed. Univariate analyses showed that abundance and richness are significantly higher during summer (ANOVA,  $F_{1,88} = 13.81$ ,  $p < 0.001$ ) and decrease with depth in summer (ANOVA,  $F_{2,42} = 03.31$ ,  $p < 0.046$ ). Multivariate ANOSIM confirms differences between seasons ( $R = 0.056$ ,  $p = 0.003$ ) and between depths ( $R > 0.126$ ,  $p < 0.001$  between all depths) except between seasons at deep depths. Fish assemblages on Two-Mile Reef can therefore be described by five distinct communities; a shallow-winter community, a shallow-summer community, an intermediate-winter community, an intermediate-summer community and a year-round deep community.

Shallow and intermediate communities were pomacentrid dominated while deep communities were dominated by serranids (subfamily Anthiinae). Ordination by multi-dimensional scaling of discriminating species, as determined by the SIMPER routine, confirms this pattern and indicates that high abundances of algal-feeding pomacentrids are observed only at shallow and intermediate sites while high abundances of zooplankton-feeding Anthiinae are observed predominantly at deep sites. An assessment of six functional groups indicated that the diverse invertivores accounted for  $> 40 \%$  of all between-category dissimilarity. The BIO-ENV procedure confirms that, of all combinations of measured factors, observed patterns are ascribed most strongly to depth ( $\rho_s = 0.314$ ,  $p < 0.001$ ).

These results suggest that season and depth, characterised by pseudo-ordinal trends in habitat complexity and benthic communities, are drivers of the coral reef fish communities on Two-Mile Reef – an evidently sensitive system.



**16h20-16h40: Matthew Parkinson (MSc student) - Primary production of phytoplankton and seaweeds in Sodwana Bay, KwaZulu-Natal**

Supervisor/s: Prof. AJ Booth (t.booth@ru.ac.za); Dr S Kaehler (s.kaehler@ru.ac.za)

Funder/s: African Coelacanth Ecosystem Programme; Rhodes University Prestigious Masters Scholarship

The warm waters of northern KwaZulu-Natal are typically clear due to a combination of a low phytoplankton biomass and the absence of large rivers discharging alluvial sediment. Seaweed communities colonise primarily shallow rocky substrata. This study aimed to quantify the rates of primary production of various seaweeds and phytoplankton communities in an attempt to determine the relative importance of localised organic matter inputs into different habitats. Using tracer techniques we estimated the rates of primary production of phytoplankton at various distances from the shore and production rates of representative green, red, brown seaweeds and “turfing” algae.

Samples of seawater were collected in the intertidal zone, over the head of one of the submarine canyons, and 10 km offshore and were incubated in parallel with samples of seaweeds collected from the rocky shore. Each incubation vessel contained a fixed amount of a tracer solution with isotopically enriched %  $\text{NaH}^{13}\text{C}\text{O}_3$  (99.9%). Samples were maintained by overnight mooring in the sea for a period of 24 hours. Seaweeds were removed from incubation vessels and POM samples filtered through precombusted GF/F filter papers. A sample of each seaweed and filter papers, were dried in an oven at  $\sim 50^\circ\text{C}$ . Samples were analysed for their stable carbon isotope signatures. Carbon incorporation (primary production) was then determined by comparing the isotope signatures after incubation with the signatures of samples prior to spiking.

Phytoplankton primary production (per unit of water) was highest in the intertidal zone and decreased offshore. Surface production was higher over the canyon head and 10 km offshore than productivity at 12 m depths. This pattern can be explained by nutrient availability and light levels, with nearshore waters typically possessing higher nutrient concentrations and light attenuation with depth. Phytoplankton productivity was fairly consistent across stations and depths indicating that the potential productivity at the stations sampled are similar, and gradients in production are likely due to differences in phytoplankton abundance.

Average seaweed daily primary production was estimated to be an order of magnitude lower than phytoplankton, when compared per unit of biomass. Chlorophytes exhibited the highest productivity with “turfing algae” the lowest. The higher production of nearshore phytoplankton together with the spatially confined nearshore production of the seaweeds makes inshore waters highly productive relative to offshore waters.

**Thursday 6 October 2011**

**Fisheries and fisheries related biology (Chair: B Ellender)**

**08h40-09h00: James McCafferty (MSc Student) - Competition angling records as indices of fish abundance in South African impoundments**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za); Dr E Swartz (e.swartz@saiab.ac.za)

Funder/s: Water Research Commission (K5/1957/4)

The development and management of inland fisheries is commonly guided by information of potential yield that are derived from empirical models that incorporate morphometric, edaphic and climatic properties of a particular water body. Typically these predictions of productivity are obtained from correlating these morpho-edaphic factors with yield estimates obtained from fisheries monitoring initiatives. Such models can then be applied to predict potential yield and guide decisions on where and how to develop fisheries.

In South Africa inland fisheries are used primarily by recreational anglers and subsistence and commercial fisheries are poorly developed. There is no formal catch monitoring system for any of the fisheries and data on total yield are unavailable. In South Africa the lack of such data hinders decision making, management and development of these potentially valuable resources because areas of high potential cannot be adequately predicted.

While no formal monitoring data are available, recreational anglers are highly organised and keep catch records from angling competitions. These catch records are currently the only fisheries data available for many impoundments and the primary objective of the current study was to: (1) assess catch data from different angling facets as potential estimators of fish abundance; (2) investigate whether catch rates from angling competitions could be correlated with morphological, edaphic and climatic characteristics of impoundments; (3) develop a model to predict areas where high catch rate (and fish abundance) are likely.

For this purpose, catch data were obtained from bass competitions held on 27 impoundments and for bank angling competitions held on 15 impoundments over a period of 10 years. In total >12000 catch records were obtained. To complement these data and to provide fisheries independent basis for comparison, gillnet survey catch records were obtained from 27 impoundments. For analysis, catch data were pooled by impoundment and stepwise multiple linear regression analysis was conducted in order to determine those environmental parameters explaining the most variability in catch per unit effort (CPUE).

Preliminary analyses showed that data from two facets, bass angling and bank angling were potentially useful for assessing catch rate. For bass angling CPUE was significantly correlated with altitude, mean annual rainfall, and conductivity, which explained 48% of the variability. Surface area and total dissolved solids (TDS) were significantly correlated with bank angling CPUE, accounting for 47% of the variability. Experimental gillnet CPUE was significantly correlated with mean depth and dam construction date which together explained 40% of the variability. These results indicate that angler CPUE could be predicated by environmental variables.

**09h00-09h20: Geraldine Taylor (MSc student) - Understanding alien invasive largemouth bass *Micropterus salmoides* in aquatic ecosystems within South Africa**

Supervisor/s: Dr OLF Weyl (O.Weyl@saiab.ac.za); Dr P Cowley (tagfish@gmail.com)

Funder/s: Rhodes University JRC; South Africa Netherlands Research Programme on Alternatives in Development (SANPAD 10/06); National Research Foundation/Southern African Institute of Aquatic Biodiversity (SAIAB)

Although largemouth bass *Micropterus salmoides* is one of the top alien invasive species worldwide, it has been, and continues to be translocated and introduced into still waters for recreational angling purposes. This species was introduced into South Africa in 1928 from its native range in North America. In order to understand largemouth bass in their invaded environments, both for conservation and angling purposes, aspects of their biology including, age and growth, reproduction, feeding, movement, population structure and utilisation are being investigated in three Eastern Cape impoundments (Wriggleswade, Binfield and Mankazana).

Age estimates were obtained from scales and sagittal otoliths. The symmetry of error produced when comparing these two structures for ageing was calculated using the Bowker's test. In Wriggleswade Dam, with many fish older than five years, the error was not symmetrical ( $\chi^2 = 56$ , df = 15, n = 147,  $P < 0.001$ ), and scales tended to underestimate age, with only a 17% otolith vs. scale agreement. However, in Mankazana, where the population comprises only fish younger than five years, error was symmetrical ( $\chi^2 = 1$ , df = 3, n = 144,  $P > 0.25$ ), with a 74% agreement. Sagittal otoliths, decidedly the more accurate and consistent structure for ageing, were therefore used to estimate the age and growth of the two populations sampled. The growth of Wriggleswade Dams' largemouth bass was described by the three parameter von Bertalanffy's growth equation as:

$$L_t = 36.6(1 - e^{-0.52(t-0.00)})$$

Fish from the Mankazana population grew faster and to a larger size, with a growth equation of:

$$L_t = 53.6(1 - e^{-0.32(t-0.66)})$$

Future work will include validation of the periodicity of growth zone formation in both scales and otoliths using Marginal Zone Analysis and mark-recapture of chemically tagged fish using oxytetracycline. Similar assessments of gonadal recrudescence will allow for the definition of the spawning season and age at maturity. Stomach content analyses will be used to determine diet and food webs will be determined using stable isotope analyses. Population utilisation and trends over time will be quantified using catch records from Wriggleswade Dam. Aspects of the movement behaviour will be determined from ten fish surgically tagged with acoustic transmitters and monitored using a passive array of receivers in Wriggleswade since May 2010.

**09h20-09h40: Thethela Bokhutlo (MSc student) - Life history and stock assessment of *Clarias gariepinus* in the Okavango Delta, Botswana**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za); K Mosepele (Harry Oppenheimer Research Institute, University of Botswana)

Funder/s: Government of Botswana, NRF/SAIAB

Large catfishes constitute a large proportion of the gill net fishery catch (23 %). Despite this relatively high abundance, its status has never been fully assessed. For this reason, this study aimed to assess the status of *C. gariepinus* in the Okavango Delta with the particular goal to determine if the species could support a directed commercial fishery.

Investigation of the effects of the flood pulse on abundance of *C. gariepinus* showed that variability in water levels was the major driver of population abundance for this species. Three biological variables: (1) catch per unit effort (CPUE); (2) % catch composition and (3) Shannon's diversity index ( $H'$ ) exhibited a negative and significant relationship with the flood index within a year. Age and growth were determined using sectioned otoliths. Edge analysis showed that growth zone deposition occurred during the drawdown period in summer when water temperature and day length were increasing. The maximum recorded age was 15 years. Growth of this species was best described by the linear models ( $y = 21.50x + 427.39 \text{ mm } L_T; r^2 = 0.35$ ) for females and ( $y = 23.79x + 440.83 \text{ mm } L_T; r^2 = 0.38$ ) for males. Age at 50 % maturity was attained in the first year of life. Spawning occurred throughout the year with an apparent peak between November and April. The mean total annual mortality rate ( $Z$ ) was 0.25 per year, natural mortality rate ( $M$ ) was 0.20 per year and fishing mortality ( $F$ ) was calculated at 0.05 per year.

The life history strategy of *C. gariepinus* was in between the periodic and opportunistic strategies. Therefore management of this species should be aimed at maintaining a healthy adult population and age structure. Per recruit analyses indicate that the current fishing mortality maintains spawner biomass at levels greater than 90 % of pristine levels. Since a large portion of the Okavango Delta is already protected and most other parts are also inaccessible to fishers there is enough spatial refugia for the species and the status quo should be maintained. Under this scenario, there is no need for regulation of the current mesh sizes because the species is harvested after maturity and replenishment of exploited populations will always occur from other parts of the system. Close monitoring will be essential to ensure that re-colonization of overexploited habitats does indeed occur.

**09h40-10h00: Amber Childs (PhD student) - An assessment of estuarine dependency and multiple habitat use of dusky kob *Argyrosomus japonicus*, with implications for fisheries management**

Supervisor: Dr PD Cowley (p.cowley@saiab.ac.za)

Funders: National Research Foundation (SAIAB); Norwegian Research Council (SA/Norway programme for research co-operation); Rhodes University Atlantic Philanthropies Scholarship; DAAD In-Country and In-Region Scholarship.

The dusky kob *Argyrosomus japonicus* (Sciaenidae) is a wide-ranging coastal fishery species. Non-compliance, ineffective management regulations and a lack of law enforcement has led to the collapse of the South African dusky kob stock. Consequently, alternative management strategies, such as area-based management need to be investigated. The aim of this study was to assess the role of estuarine nursery habitats in the life cycle of the dusky kob and then to use this information in the management of this species.

An acoustic array comprising 49 data-logging receivers spanning 300 km of coastline, focusing on the Sundays Estuary and Algoa Bay, was used to monitor the movements of 94 acoustically-tagged *A. japonicus* (237–1130 mm TL) over three years. Additional information from several conventional tag-recapture projects (e.g. ORI's National Tagging Project and SAIAB's Algoa Bay tagging project) was used to determine the migrations and dispersal patterns of the dusky kob. The micro-chemical composition of otoliths from 126 young-of-the-year individuals, collected from seven habitats, was used to identify key nursery habitats for this species.

The results of this multi-method study have shown that estuarine habitats are critical nursery areas for juveniles and important habitats for both sub-adult and adult dusky kob, but their reliance on these nursery habitats creates a bottleneck in their life history. Conventional tagging data showed that dusky kob are resident to specific estuaries and adjacent surf-zones and exhibit low levels of dispersal, with between 77 and 100% of recaptured fish were caught within 10 km from their tagging sites. Similarly, recapture statistics from fish equipped with acoustic transmitters showed that 91% of recaptures were caught within 10 km of their tagging site. The high recapture rate (33%) of acoustically tagged dusky kob further highlights the vulnerability of this species to exploitation in estuarine nursery habitats. Telemetry data also showed that dusky kob exhibited high levels of residency within the estuary (tagged fish spent on average 94% of their time in the estuary) and low levels of connectivity (12% of tagged fish were recorded out of Algoa Bay). Dusky kob were also found to be resident to specific areas in the estuary and that their estuarine movements were significantly correlated to predictable biological rhythmic cycles such as the lunar and tidal phases as well as several other abiotic factors. Otolith micro-chemistry results suggest that turbid estuaries play a critical role in the successful recruitment of dusky kob. The random forest classification analysis showed that juveniles sampled from the more turbid estuaries had a significantly higher PCC (percentage correctly classified) value compared to other less turbid estuaries. This study has highlighted the advantages of using a multi-method approach to provide insights into the ecology and corrective management of this over-exploited fishery species.

**10h00-10h20: Rhett Bennett (PhD student) - Habitat use, movement patterns and stock delineation of an important endemic coastal fishery species, *Lithognathus lithognathus***

Supervisor: Dr PD Cowley (p.cowley@saiab.ac.za)

Funders: Marine and Coastal Management Provincial Research Projects; South African National Biodiversity Institute; South African Institute for Aquatic Biodiversity (SAIAB)

White steenbras *Lithognathus lithognathus* is one of South Africa's most sought after and threatened endemic coastal fishery species. Dependence on estuarine nursery habitats, residency within the inshore marine environment and predictable spawning migrations make white steenbras highly vulnerable to localised overexploitation, and are factors contributing to stock collapse.

This study aimed to determine estuarine space use patterns, estuarine-dependency, coastal movement patterns and genetic stock structure of white steenbras in South Africa, to provide recommendations for corrective management.

Acoustic telemetry studies conducted in the Great Fish, East Kleinemonde, Kariega and Sundays estuaries (Eastern Cape) showed that juvenile white steenbras are highly resident within estuaries up to about three years old (250 – 350 mm FL), with little tendency to undertake excursions to sea. These early juveniles also exhibit distinct diel movement, using the shallow littoral zone at night and deeper channel areas during the day. After having left the estuarine environment after their juvenile nursery phase, white steenbras lose their dependence on estuaries. Acoustic telemetry and conventional dart tagging showed that late juveniles up to 400 mm FL are highly resident within the surf-zone (mean displacement = 2 km, maximum = 25 km). Between 400 and 500 mm FL (roughly 4 – 5 years old), movements tended to increase in distance (mean displacement = 26 km, maximum = 419 km), although movements were generally on a small scale and the majority of individuals remained resident, while larger individuals (>500 mm FL) began to undertake large-scale migrations (mean displacement = 70 km, maximum = 620 km), with only a small proportion (approximately 30%) exhibiting residency. Preliminary analyses of nuclear (microsatellite) and mitochondrial DNA from 321 white steenbras, collected between the Dwesa-Cwebe Marine Protected Area in the east and Langebaan Lagoon in the west, indicated high genetic diversity. Additionally, there was little evidence of spatial variability in genetic stock structure, providing evidence of a single, well-mixed adult population. The multi-method approach adopted during this study provides an improved scientific basis for the management of white steenbras and a platform for research on other estuarine-associated coastal fishery species.

**10h20-10h40: Taryn Murray (MSc student) - Movement behaviour and genetic stock delineation of poenskop *Cymatoceps nasutus***

Supervisors: Dr G Gouws (g.gouws@saiab.ac.za); Dr PD Cowley (p.cowley@saiab.ac.za); Mr BQ Mann (bruce@ori.org.za)

Funders: International Foundation for Science (A/4530-1F); National Research Foundation (IFR20100430040); South African Institute for Aquatic Biodiversity (SAIAB)

The poenskop *Cymatoceps nasutus* is an endemic South African sparid with life history characteristics (e.g. slow-growing, long-lived, sex-changing and late maturing) that make it acutely sensitive to over-exploitation. Catch per unit effort trends reflect a severe and consistent stock decline over the past two decades. Management is compromised by the deficiency of information regarding stock integrity and heterogeneity (i.e. genetic structure and diversity), and movement behaviour (migration and residency patterns) and this formed the basis for the study.

The aims of the study were to a) describe the movement behaviour of poenskop using tag-recapture data and b) determine the genetic stock structure throughout its distributional range. Data on the movement patterns were obtained from the Oceanographic Research Institute's National Tagging Projects and dedicated tagging projects conducted in three geographically separated marine protected areas (De Hoop, Tsitsikamma and Pondoland). These datasets with varying scales of spatial resolution are currently being analysed.

In order to determine the spatial and temporal genetic diversity of the poenskop, fin clips from 370 fish were collected from Stilbaai in the Western Cape to Umtentwini in KwaZulu-Natal. DNA was extracted from samples using commercially available kits. A fragment of the mitochondrial control region was amplified by PCR using newly designed, species-specific primers. Samples were then sequenced at Rhodes University's Sequencing Unit and a commercial sequencing facility (Macrogen). Indices of genetic diversity (using ARLEQUIN), estimates of genetic differentiation among regions, the partitioning of genetic variance within and among regions (AMOVAs), and Bayesian estimates of connectivity and migration have been calculated.

DNA sequence data have been generated for 294 samples from 35 localities. The haplotype diversity ( $H_D$ ) for all samples from all regions was  $0.8743 \pm 0.020$  (range =  $0.795 \pm 0.109$  and  $0.903 \pm 0.031$ ). These analyses suggest that *C. nasutus* has a high genetic diversity with very little spatial structure; however more analyses will confirm this. These results are very similar to those found for other local sparids namely *Chrysoblephus laticeps* (Teske *et al.* 2010) and *Rhabdosargus holubi* (Oosthuizen 2007), which were also found to exist as single, well-mixed stocks throughout their distributional ranges.

In addition, information obtained from available fishery records, published and unpublished survey and research data will be used to evaluate the current and historic trend in catch and *cpue* of poenskop.

**10h40-10h55: Tea Break**

## **Estuarine and freshwater ecology, conservation and related biology (Chair: M Ramoejane)**

**11h00-11h20: Craig Midgley (MSc student) - Changes in the fish assemblage structure of transition-zone estuaries in South Africa – possible effects of climate change?**

Supervisor/s: Dr NC James (n.james@saiab.ac.za); Prof. AK Whitfield (a.whitfield@saiab.ac.za); Dr SJ Lamberth (stephenl@nds.agric.za)

Funder/s: National Research Foundation (SAIAB)

Changes in the occurrence and distribution of organisms, linked to increasing temperatures, have been recorded both locally and globally, which in the long-term will affect the population dynamics of the systems in which they occur. The effects of global climate change on estuarine fish populations occurring in the transition-zones between biogeographical regions are not well researched in South Africa. The estuarine ichthyofauna in the permanently open Mbashe Estuary and the temporarily open/closed Mbhanyana Estuary, situated in the transition zone between the subtropical and warm-temperate biogeographical regions, were sampled in spring 1997 and 2010 using a seine net. It is hypothesized that temperate species at the northern limit of their distribution will decrease in abundance, while tropical species will increase in abundance.

Littoral habitats in the lower, middle and upper reaches of the estuaries were sampled. All fish caught were identified and measured to the nearest millimetre standard length (SL) before being released. If identification was not possible in the field the fish were preserved in 10 % formalin and identified in the laboratory. Physico-chemical parameters such as temperature (°C), salinity, pH and dissolved oxygen (mg/L) were recorded at each site. The ichthyofauna were divided into four groups based on their distribution: tropical, warm-temperate, cool-temperate and widespread. Although no clear trends were observed in either the number of tropical species recorded or the proportion of tropical species in the catch in the permanently open Mbashe Estuary, there was a slight decrease in the abundance of temperate species at the northern limit of their distribution from 1997 to 2010. In the temporarily open/closed Mbhanyana Estuary, which is cut off from the effect of sea temperatures for longer periods, the contribution of tropical species to the total catch of marine species increased from 16% in 1997 to 44% in 2010, while warm-temperate species decreased from 82% to 55% of the catch. . Although this could be linked to climate change more data, collected over a longer time period is required to make any conclusive remarks. This shows how critical long term data is for understanding community changes in estuaries caused by climate change.

The estuarine ichthyofauna in the permanently open Breede Estuary, situated in the transition zone between the cool-temperate and warm-temperate biogeographical regions, was sampled in summer and winter from 2000 to 2010. The Breede Estuary data has not been analysed yet.



**11h40-12h00: Alexis Olds (MSc student) - Introduction pathways, distribution and relative abundance of alien invasive fish species in a South African RAMSAR wetland, Wilderness Lakes**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za); Mr MKS Smith (kyles@sanparks.org)

Funder/s: The CIB DST-NRF Centre of Excellence for Invasion Biology; SANParks; NRF/SAIAB

The Wilderness Lakes System, comprised of three lakes (Island Lake, Langvlei and Rondevlei) and their interconnecting channels, the Touw Estuary and the Serpentine channel, form a component of the Garden Route National Park (SANParks). With the exception of the Touw Estuary, the lakes and interconnecting channels have been designated as a RAMSAR site. As a result of introductions into the catchment area, four alien fishes have been recorded from the system. Three of these, the common carp *Cyprinus carpio*, Mozambique tilapia *Oreochromis mossambicus* and largemouth bass *Micropterus salmoides* are popular angling species while a fourth, the mosquitofish *Gambusia affinis* was most likely introduced for mosquito control. Elsewhere, these alien fishes, listed among the 100 worst invasive organisms worldwide, have been linked to a variety of impacts including competition with native biota, alterations of invertebrate communities, predation, habitat alteration and introduction of fish diseases. Understanding the status and establishment of these fishes in the Garden Route National Park is therefore important.

The overall aim of this research project was to contribute to the understanding of the dynamics of alien invasive fishes in the Wilderness Lakes and to contribute to an effective alien invasive management strategy for SANParks. Specific project objectives included the identification of introduction pathways as well as source populations of alien invasive fish. Assess the spatial, temporal distribution and relative abundance of native and alien fish species throughout the system and finally to investigate various physico-chemical parameters and assess their relation to the abundance and distribution of alien invasive fish.

In this research the lakes system was sampled seasonally using a range of gear types. Each water body was sampled by means of fyke nets, multi-meshed gill nets, 30m and 10m seine nets to determine the abundance and distribution of fish within the lakes. Various physio-chemical parameters were also collected to correlate these data to environmental variables. Interviews with farm owners and anglers provided information on the source and distribution of alien fishes. The data collected was compared with a 1985 study.

Analyses show that the species composition in this system is similar to other temporary open closed estuaries in South Africa in that the fish fauna comprises mostly native estuarine and euryhaline marine species. Alien fishes were however, common and abundant. *Oreochromis mossambicus* comprised 91% of the total fyke net catch and 18% of the total gill net catch spanning the entire system. The littoral zones of the lakes and in the interconnecting channels *G. affinis* contributes 26% to the ichthyofauna. Sampled *C. carpio*, which occurs throughout the system, increased in size over the sampling period from an average of 203mm FL in 2009 to 542mm FL in 2010. Spawning was confirmed by the presence of ripe running fish and the collection of juveniles in early 2011. The establishment success of this species appears to have been limited in their establishment by temperature and salinity.

**12h00-12h20: Rogan Field (MSc Student) - Inter-basin water transfer schemes and hybridisation within the genus *Labeo***

Supervisors: Prof. H Kaiser (h.kaiser@ru.ac.za); Dr OLF Weyl (o.weyl@saiab.ac.za)

Funders: National Research Foundation Competitive Grant (UID 73667)

South Africa is a water-scarce country. Consequently, numerous inter-basin water transfer schemes (IBTs) have been established pumping and channelling water from areas of perceived excess to those with known deficiencies. Although a practical solution, IBTs could pose a serious threat to biodiversity. IBTs allow for the transfer of indigenous and invasive alien species between river basins and previously isolated ecosystems increasing the risks of invasions as well as the potential for hybridisation.

*Labeo capensis* of the Orange River system has been introduced into the previously isolated Fish River system with the proposed pathway of introduction being the Orange-Fish tunnel. Consequently, mixing of *Labeo capensis* and *Labeo umbratus*, the latter from the Fish River, has occurred. Exhibiting similar life history patterns and utilising similar spawning grounds and seasonality the potential for hybridisation between these species has been documented. This project seeks to quantify that risk through the assessment of the development of eggs, larvae and juvenile *L. capensis*, *L. umbratus* and the reciprocal hybrids generated through a captive breeding program.

Wild-caught *L. capensis* and *L. umbratus* of over 25 cm total length will be transported to Rhodes University. They will be held in a custom-built broodstock facility comprised of eight 2200 L Porta-Pools in a closed recirculation system. Spawning of broodstock will be induced through injection of Aquaspawn®. Eggs and milt will be stripped and fertilisation will be by way of the 'dry method'. Egg, larvae and juvenile development will be closely monitored and recoded. Data will include percentage fertilisation, 4, 32, 64 cell divisions, survival to eyed ova, and hatching success. Larval development and juvenile development will be described based on morphometric and meristic data of both species and their reciprocal hybrid offspring.

The results should allow for direct comparisons between the success of fertilisation and egg, larvae and juvenile development between parent stock and reciprocal hybrids and thus contribute to making an assessment of the risk of hybridisation between these species. This study will also provide a description of the hybrid specimens for the identification of wild-caught hybrids. Finally, the results of this project may be used to advise conservation officials to the threat of hybridisation within this genus.

**12h20-12h40: Evans Simasiku (MSc student) - Population Dynamics of *Oreochromis andersonii*, *Oreochromis macrochir* and *Tilapia rendalli* (Pisces: Cichlidae) in the Kavango River floodplain, Namibia**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za); Mr. D. Tweddle (Namibian Nature Foundation, Caprivi Region/ SAIAB)

Funder/s: DAAD, SAIAB, NNF, Ministry of Fisheries and Marine Resources, Namibia

Fishing is important for food security, income and recreation in the Kavango region of Namibia. *Oreochromis andersonii*, *Oreochromis macrochir* and *Tilapia rendalli* are important components in commercial and subsistence fisheries and are popular among recreational anglers. Increased utilisation of fishes, particularly in floodplain habitats have led to a need for the development of management recommendations for these fisheries. This requires information on the biology and ecology of the fishes that are targeted by the harvesting fisheries. An important component of this is understanding the utilization of floodplain habitats by juvenile fishes and their movement between the main river and the floodplains. The objective of this MSc study is therefore to investigate and contribute to understanding of how *O. andersonii*, *O. macrochir* and *T. rendalli* utilise floodplain habitats. The objectives of the MSc are to assess the biology and ecology of juveniles of these species on the floodplain and will include assessments of relative abundance, size structure, growth, diet and food resource use on the floodplain.

Research to date has focussed mainly on the Kavango River floodplain and has concentrated on assessing their relative abundance and size structure. Seine net surveys have shown that a total of 23 species, representing 8 families (*Cichlidae*, *Characidae*, *Poecillidae*, *Cyprinidae*, *Scilbeidae*, *Clariidae*, *Mochokidae*, and *Schilbeidae*) were collected between March and August 2011. Cichlids were the most abundant by number and the three major components of the fish fauna were *Tilapia sparrmanii* (34%), *O. macrochir* (15%) and *Brycinus lateralis* (8%). Seasonal analysis showed that the peak abundance of the *Characidae* and *Cyprinidae* lagged slightly behind that of the *Cichlidae* and *Poecillidae* in the floodplain. Length frequency analysis indicated that *O. andersonii*, and *O. macrochir* juveniles formed a single cohort whereas multiple cohorts of *T. rendalli* were observed. These results imply that *T. rendalli* appear to have a much longer spawning season than *O. andersonii*, and *O. macrochir*. Research next year will focus on feeding ecology and ageing of juvenile fishes on the floodplain.

**12h40-13h00: Gamuchirai Chakona (Mataruse) (MSc Student) - Phylogeography and conservation of a newly discovered *Galaxias* species from the Joubertina area in the Eastern Cape Province of South Africa**

Supervisor: Dr ER Swartz (e.swartz@saiab.ac.za)

Funder: South African Institute for Aquatic Biodiversity (SAIAB)

Most freshwater restricted species tend to display higher levels of genetic structuring than marine species, because both terrestrial and marine environments can be barriers to dispersal between river systems. A newly discovered lineage of *Galaxias zebratus* (hereafter *Galaxias* 'Joubertina') occurs in two currently isolated river systems (the Krom and Gamtoos) in the eastern Cape Floristic Region. This lineage appears to be confined to freshwater, therefore both marine and terrestrial environments are effective barriers to its dispersal. Very little is known about the genetic diversity, phylogenetic relationships and processes that influenced the lineage's present distribution pattern. The aim of the present study is to map the distribution of *Galaxias* 'Joubertina' and investigate the processes that influenced its phylogeographic pattern.

A total of 72 localities (30 from the Krom and 42 from the Gamtoos) were sampled and 180 fish were collected for genetic analysis. The field surveys confirmed that *Galaxias* 'Joubertina' has a highly restricted geographic range and is only known from six small mountain streams. Data from mitochondrial cytochrome *b* and nuclear (S7) sequences revealed that there is no genetic differentiation between the Krom (Krom River system) and Twee (Gamtoos River system) populations. Shallow genetic differentiation (0.3%) was found between the Krom population and the other Gamtoos populations. Analysis of molecular variance (AMOVA) showed that variation was explained mainly by differentiation among populations within river systems (68.9%; 40%) rather than between river systems (-0.47%; 0.01%). Nested Clade Analysis inferred past gradual range expansion followed by fragmentation and restricted gene flow with isolation by distance as possible explanations for the shallow divergences between populations from the two river systems and unique haplotypes from Krom and Twee rivers.

River capture is an unlikely explanation for the geographic distribution of genetic diversity, because there is no evidence of recent drainage rearrangements between Krom and Gamtoos. Similarly, sea level regression is unlikely as this lineage is absent from tributaries lower down in the Krom River system. Rare events such as inter-basin dispersal during pluvial periods seem to be the most plausible explanation for the shallow genetic divergence between populations from the two river systems. However, the lack of differentiation between the Twee and Krom populations suggest ongoing or very recent gene flow between the two river systems. Canals that connect the Krom and Twee could have facilitated dispersal of this lineage between these historically isolated river systems.

**13h00-13h55: Lunch break**

## **Estuarine and freshwater ecology, conservation and related biology continued** **(Chair: W Kadye)**

**14h00-14h20: Mpho Ramoejane (PhD student) - Phylogeography of *Labeo umbratus* and *L. capensis***

Supervisors: Dr ER Swartz (e.swartz@saiab.ac.za); Dr O Weyl (o.weyl@saiab.ac.za)

Funder: National Research Foundation (SAIAB)

The closely related *Labeo umbratus* and *L. capensis* are primary freshwater fish species that are endemic to southern Africa and have been shown to hybridise in Hardap Dam (Namibia) and Darlington Dam (Eastern Cape Province, South Africa). *Labeo umbratus* has a wide distribution across the Orange, Gouritz, Gamtoos, Sundays, Great Fish and Bushmans river systems. They have apparently been translocated to the Keiskamma, Baffalo and Nahoon river systems. *Labeo capensis* only occurs in the Orange River system, but have been translocated to the Great Fish and Sundays river systems through inter-basin water transfer schemes (IBTs). The IBTs also allowed populations of *L. umbratus* to mix. In the present study a mitochondrial marker was used to assess which evolutionary processes were responsible for present genetic diversity patterns and to assess the role of geological and climatic processes in the differentiation of populations. Mitochondrial DNA has the advantage that it does not recombine, and is therefore not affected by hybridisation.

To assess the geographic distribution of genetic diversity, more than 600 base pairs of the mitochondrial cytochrome *b* gene were sequenced for 275 specimens. Phylogenetic trees and networks were constructed to illustrate genetic distances among lineages. Genetic patterns, inferred populations histories and evolutionary process were then compared to palaeoriver systems reconstructed for the Last Glacial Maximum 18 000 years ago. This was done to assess the role of climatic and geological change in shaping the population histories of these two species.

Preliminary results show differentiation between the Orange and southern flowing river systems for *L. umbratus*. There were very little divergence among southern flowing river systems for this species. However, several alleles were restricted to single river systems, indicating a lack of current gene flow. Private alleles in the suspected alien populations suggest that these might be natural populations and that *L. umbratus* might have a much wider natural range than previously thought. There was very little structuring in *L. capensis*, suggesting high levels of gene flow in this species across the Orange River system. It is possible that at least *L. umbratus* was introduced to Hardap dam, because of extensive hybridisation with *L. capensis* in this reservoir and due to their natural absence from the lower Orange below Augrabies waterfall.

**14h20-14h40: Bruce Ellender (PhD student) - Ecological consequences of non-native fish invasion in Eastern Cape headwater streams**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za); Dr E Swartz (e.swartz@saiab.ac.za)

Funder/s: The CIB DST-NRF Centre of Excellence for Invasion Biology; Water Research Commission (K5/1957//4), Rhodes University JRC, SANPAD 10/06

Non-native fish invasion is considered a primary threat to freshwater fishes, particularly in headwater streams which typically have low species diversity but a high degree of endemism. Introduction of non-native fishes is, however, often a matter of risk perception rather than real risk analysis and an introduction can only be considered harmful if it leads to measurable loss of biodiversity or changes in ecosystem functioning. This research therefore attempts to investigate the ecological consequences of non-native fish invasion in the headwaters of two Eastern Cape river systems, the Swartkops and Keiskamma River systems.

The Swartkops River, contains the endangered *Pseudobarbus afer* and the non-native *Micropterus salmoides*, *Micropterus dolomieu* and the catfish *Clarias gariepinus*. Headwater streams of the Keiskamma River system where *Barbus trevelyani* are threatened by *M. salmoides*, *M. dolomieu*, *Oncorhynchus mykiss* and *Salmo trutta*. The research includes: (1) determining suitable non-destructive sampling methods for surveying imperilled fishes; (2) assessments of the distribution and abundance of indigenous fishes in relation to non-native fishes; (3) genetic assessments to determine the size of evolutionary significant units of endangered species to assess whether range restriction from non-native fish predation will impact on the genetic diversity of the indigenous fishes; (4) quantifying the invasive impact of non-native fishes on indigenous fishes in stream ecosystems.

Progress includes (1) two experiments on sampling methods for endangered fishes and distribution surveys are complete. To determine whether underwater video analysis (UWVA) could be used as an alternative to electrofishing for estimating species diversity and abundance in upper catchment streams experiments on the Fernkloof (five sites), Waterkloof (five sites) and Rondegat (19 sites) streams were undertaken. Results indicate that UWVA is a suitable tool for estimating occurrence and relative abundance of some fish species in clear water environments; (2) distribution and abundance surveys have been completed on the upper Swartkops (75 sites) and Keiskamma (75 sites) River systems and this data is currently being analysed. Three dams (Cata, Binfield Park and Sandile) on the upper Keiskamma system have also been surveyed intensively to determine the abundance and diversity of fishes therein (20 net nights per dam). Additionally to determine the effect of temperature on fish distribution 33 temperature loggers have been placed in the Keiskamma River system; (3) genetic sampling is complete with 5 samples per tributary from both the upper Swartkops (30 *Pseudobarbus afer*) and Keiskamma (40 *Barbus trevelyani*) River systems; (4) Invasive impact will be estimated by using the equation  $I = R \times A \times E$  where overall impact,  $I$ , is defined as the product of the range size  $R$  of a species, its average abundance ( $A$ ) across that range, and  $E$ , the effect per individual or per biomass unit of the invader.

**14h40-15h00: Albert Chakona (PhD student) - Paleogeographic events and dispersal as drivers of lineage diversification and biogeographic distribution in three sympatric freshwater fishes from the south-western Cape Floristic Region of South Africa**

Supervisors: Dr ER Swartz (e.swartz@saiab.ac.za); Dr G Gouws (g.gouws@saiab.ac.za); Prof. P Bloomer (p.bloomer@up.ac.za)

Funders: National Research Foundation; Rufford Small Grants Foundation; International Foundation for Science; WWF International Prince Bernhard Scholarship; South African Institute for Aquatic Biodiversity (SAIAB)

The evolutionary history of the Cape Floristic Region (CFR)'s freshwater restricted taxa is not fully understood. This study used phylogenetic analyses of mitochondrial cytochrome *b* sequences to investigate genetic diversity within three broadly co-distributed freshwater fish genera (*Galaxias*, *Pseudobarbus* and *Sandelia*) from the south-western CFR to shed light on the processes that promoted lineage diversification and shaped geographical distribution patterns. A comprehensive data set (480 sequences) from 146 localities across all river systems in the south-western CFR was used. The data was analysed using phylogenetic and allele network methods and divergence times for the clades retrieved were estimated using three independent approaches.

Nine extremely divergent (3.5 – 25.3%) lineages were found within *Galaxias*. Similarly, deep phylogeographic divergence was evident within *Pseudobarbus*, with four distinct (3.8 – 10.0%) lineages identified. *Sandelia* has two deeply divergent (5.5 – 5.9%) lineages, but seven haplogroups with strong geographical congruence were also identified. The Miocene-Pliocene major sea-level transgression and the resultant isolation of populations in upland refugia appears to have driven widespread allopatric divergence in the three genera. Subsequent coalescence of rivers during the Pleistocene major sea-level regression, as well as intermittent drainage connections during wet periods, are proposed to have facilitated post-speciation dispersal of lineages that currently occur across isolated river systems.

The high degree of genetic differentiation recovered from the present and previous studies suggest that freshwater fish diversity within the south-western CFR may be vastly underestimated, and taxonomic revisions may be warranted. Most of the newly discovered lineages have very restricted geographic ranges and probably have small population sizes. Conservation measures in the CFR should first address immediate threats such as invasion by alien fishes, habitat degradation and excessive abstraction of water. Once the populations are secured, the risk of inbreeding due to small population sizes can be assessed, and critical populations can be enlarged to ensure long-term survival.

**15.00-15.20: Carl Huchzermeyer (MSc student) - An assessment of the fish diversity and artisanal fishery of the Bangweulu floodplains, Zambia, with particular reference to the ecological input of terrestrial herbivores, and to conservation area management goals**

Supervisor/s: Dr OLF Weyl (o.weyl@saiab.ac.za), Dr R Bills (r.bills@saiab.ac.za)

Funder/s: NRF/DAAD Bursary; African Parks

The community-owned conservation area, the Bangweulu Wetlands Project, encompasses a large part of the south-eastern swamps and floodplains of the Bangweulu system, on the Congo River catchment, in Zambia. The ecology of the area is dominated by the annual flood cycle, which inundates a huge part of this flat area. The floodplains support a rich artisanal fishery, and are home to an estimated 75 000 black lechwe antelopes (*Kobus lechwe smithemani*), particularly on plains around the Lukulu river estuary, also the main study area.

The aim of the study is to get a broad understanding of the fish diversity and ecology, and the utilisation of the fish resource by residents living inside the conservation area. Field work in 2011 has focussed on data collection, recording seasonal changes in distribution and resource use patterns. Taxonomic work, laboratory work on biological samples and data analysis will be completed in 2012.

Fishing activities commence as the floods recede in March, and carry on until flooding occurs in December. Gears used on the floodplain are reed basket traps and mosquito-net funnels set in earthen 'fish-weirs', small mesh-size multifilament gillnets and longlines. During the dry season, seine nets are used to catch fish in channels, mostly to the exclusion of other gears. Fish stranded in pools and shallow areas without channels are caught by hand and with spears, often after the application of a fish poison. Catch composition and CPUE data is being collected for various gears.

The fish fauna consists of very typical floodplain groups, and 27 species have been collected during this study. Additional species occur in the streams flowing into the swamps, where there is little fishing. Cyprinids, clariids, mormyrids and cichlids make up the majority of fish occurring in the area.

The bulk of fish caught are dried for sale. Oily fish such as *Clarias*, *Marcusenius* and large cichlids are sundried and then smoked, while smaller species are sundried. There is a limited amount of fresh fish trade, mostly in cichlids.

Biological parameters of five important fishery species are being collected every month. These are *Marcusenius macrolepidotus*, *Serranochromis 'robustus'*, *Clarias ngamensis*, *Barbus trimaculatus* and *Tilapia sparrmanii*. Parameters include length, weight, maturity and gonadal staging, otolith collection and preservation of stomach contents. Parasites are noted and preserved for later identification of key groups.

The research may extend into quantifying the role that the herds of black lechwe antelopes play in nutrient cycling. This has been initiated and dung per unit area is being measured and analysis for nitrogen and phosphorous content, and stable isotope work is planned to compare nutrient flows inside and outside areas where lechwe occur.

**15h20-15h35: Tea break**



## **Fisheries and fisheries related biology continued (Chair: V Tibenda)**

**15h40-16h00: Denham Parker (MSc Student) - The biology and life history of the largespot pompano, *Trachinotus botla* (Pisces: Carangidae), off the KwaZulu-Natal coast, South Africa**

Supervisor: Prof. AJ Booth (t.booth@ru.ac.za)

Funder/s: National Research Foundation (F10002).

*Trachinotus botla* is one of approximately fifty carangid species distributed off southern Africa. In northern KwaZulu-Natal it comprises an important component of both recreational and subsistence linefisher's catches. Over the past two decades it has been exposed to an increase in fishing effort. The effects that the increase in exploitation will have on the *T. botla* population is not fully understood as there is currently no information pertaining to its general biology or life history characteristics. Further investigation into the biology of the species is therefore necessary.

A total of 369 fish have been collected with a size range of 119–495 mm FL. Results of this study show that *T. botla* is a fast growing, short lived and early maturing species. The species has a protracted spawning season ranging from November to February with histological examination of the gonads suggesting it is an asynchronous iteroparous species. Mean monthly surf zone water temperature was significantly correlated to gonadosomatic index (GSI) and the periodic regression predicted a 12 month cyclic pattern of reproductive activity. The sex ratio was slightly female-biased (1 male: 1.3 females), however this bias only increased with very large fish.

*Trachinotus botla* is an opportunistic predator, feeding on a wide variety of prey items. Teleostei (especially engraulids) are important dietary items of fish of all sizes. Small fish also fed on copepods and terrestrial insects, while large fish fed mainly on mussels and crabs. Overall, the opportunistic utilisation of “superabundant” prey items appears to characterise of the species feeding habits. An ontogenetic dietary shift was observed in *T. botla*, with probable links to a shift in habitat utilisation with size. Seasonal variation in diet within size classes was minimal. Copepods were the only prey taxon to show notable differences being significantly more abundant in the stomachs of fish sampled during winter.

A parasitic tongue-replacing isopod, *Cymothoa* sp. has been found in the buccal cavity of 44% of the fish sampled. A further 5% of the fish had significant tongue damage as a result of previous parasite infection. Larger females are found on the tongue of the host and the smaller male on the gill rakers of one gill chamber. *Cymothoa* sp. was noted to almost occupy the entire oral cavity. Condition factor of infected fish did not differ significantly between infected and uninfected fish (t-test;  $p > 0.05$ ). Stable isotope ratios of carbon and nitrogen were not found to be significantly different ( $p > 0.05$ ) between infected and uninfected fish suggesting the buccal obstruction does not limit the feeding ability of the fish.

**16h00-16h20: Devin Isemonger (MSc Student) - Modelling the genetic and distributional response of the shared fisheries species, *Polysteganus praeorbitalis*, to climate change in the Western Indian Ocean**

Supervisors: Dr M Mwale (m.mwale@saiab.ac.za); Dr N James (n.james@saiab.ac.za)

Funders: South African Institute for Aquatic Biodiversity (SAIAB); Western Indian Ocean Marine Science Association; Rhodes University

Climate change has resulted in recorded shifts in both the longitudinal and depth ranges of marine fish species through related changes in sea surface temperatures, ocean chemistry and primary production. These shifts are likely to affect the abundance of certain species and the community structure and functioning of marine ecosystems in which they occur, and are likely to result in sociological and economic impacts for communities which harvest living resources. In the face of environmental change, levels of genetic diversity may determine, in part, the ability of species to adapt to new conditions through contemporary evolution, thereby reducing negative effects on abundance and distribution. The subject of this study, *Polysteganus praeorbitalis*, is an important fisheries species that ranges from Beira in Mozambique to Algoa Bay in South Africa. Targeted by several different fisheries within this range, *P. praeorbitalis* has been severely overexploited, leading to a 90% decline in CPUE in South Africa since 1941. It's status as an overexploited, trans-boundary stock endemic to the Western Indian Ocean makes *P. praeorbitalis* a good species on which to model the effects of climate change in conjunction with overfishing, providing information which may be useful in the management of this and other similar species.

The aim of this project is to predict the effects of climate change in the Western Indian Ocean on the distribution of *P. praeorbitalis*. An assessment of the genetic diversity and stock structure of this species along its range will also be conducted in order to predict the ability of the species to adapt to a range of climate change scenarios and to evaluate the current status of the stock in terms of genetic diversity.

The current distribution of the species will be modelled using species presence data and historical environmental data (such as sea surface temperature, salinity and depth) using appropriate statistical techniques and programmes, such as generalised additive models, BIOMOD and MAXENT. The distribution model will then be projected by applying it to another set of future environmental layers. An analysis of the genetic diversity of *P. praeorbitalis* will be carried out on DNA extracted and amplified using standard procedures and kits, and sequenced by MacroGen.

Sample collection is still on-going with roughly 100 samples collected thus far and sampling complete for the northern Transkei and southern KwaZulu-Natal (KZN) regions. Further samples are still required for the Mozambique, northern KZN, southern Transkei and Eastern Cape regions. Extraction and sequencing is complete for the samples that have been collected and analysis of this the data will begin shortly.

**16h20-16h40: Murray Duncan (MSc Student) - The genetic assessment of slinger (*Chrysoblephus puniceus*) stocks in the South Western Indian Ocean and management implications**

Supervisors: Dr M Mwale (m.mwale@saiab.ac.za); Dr S Fennessy (seanf@ori.org.za)

Funders: Western Indian Ocean Marine Science Association; Southwest Indian Ocean Fisheries Project;  
Deutscher Akademischer Austausch Dienst

The slinger, *Chrysoblephus puniceus*, a seabream of the family Sparidae, is an important, endemic, linefish species in the South West Indian Ocean, contributing the most in terms percent contribution to the commercial linefishery catch in Kwa-Zulu Natal (South Africa) and Southern Mozambique. Previous stock assessments have indicated that *C. puniceus* stocks are over exploited in both South Africa and Mozambique. These assessments are due for updating but some key questions need to be answered before another stock assessment can be conducted. There is presently very limited information on the migration and recruitment of this reef species despite its fisheries importance. General fisheries theory might predict that larvae would flow from the north to the south, with a return migration of mature fish from the south to the north. A first step would therefore be to establish the levels of genetic connectivity among populations of *C. puniceus* in South Africa and Mozambique.

The project has three primary objectives:

1. to provide a genetic assessment of the structure and variability of *C. puniceus* throughout its distribution range in the South West Indian Ocean. The levels of genetic divergence indicated by mtDNA data and microsatellite data will also be compared to examine the utility of these two types of markers for detecting different levels of divergence;
2. to determine spatial and temporal patterns of genetic connectivity of *C. puniceus* among areas within the SWIO subtropical region; and
3. to demonstrate how the specific knowledge of genetic stock assessments and connectivity can complement and be integrated into marine biodiversity conservation planning and the management of exploited resources.

Tissue Samples were collected throughout the distributional range of *C. puniceus* for genetic analyses. Ten microsatellite loci and one mitochondrial (d-loop) marker will be used to amplify and sequence genetic data to analyse the genetic structure of the species. 281 samples from the entire species distribution have been extracted, amplified with specific control region primers and sequenced. This sequence data has been aligned and is being analysed. 10 microsatellite markers still need to be amplified.

**16h40-17h00: Wilbert Kadye (PhD student) - Impact of non-native sharptooth catfish *Clarias gariepinus* in the Great Fish and Sundays Rivers**

Supervisor: Prof. A Booth (t.booth@ru.ac.za)

Funders: National Research Foundation (FA2007043000021); Rufford Small Grants Foundation

Invasive species are a major concern as they have the potential to alter community structure and food web relationships within the invaded habitats. An integrated approach to compare feeding patterns food webs within the invaded habitats is crucial in understanding the role of the invader and its potential impact. The African sharptooth catfish *Clarias gariepinus* was introduced into the Eastern Cape Province in South Africa where it poses threat to indigenous species and other biota. Stomach content analysis and stable isotope ratios of carbon and nitrogen were used to quantify food sources and to compare the spatial variation in the community and catfish niches among habitats within invaded systems.

Stomach content analysis indicated a catholic diet dominated by fish. The estimates from isotope mixing model that showed a discernible increase in indigenous fish prey and decrease in aquatic invertebrates prey with catfish size classes, corroborated with those observed from stomach content analysis. The communities and catfish centroid locations, based on the scatter of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , indicated statistical differences among localities and habitat type, suggesting that each locality had a distinct community. Analyses of the measures of dispersion however showed no evidence of differences for the comparisons between communities, suggesting that there were no differences in the clustering among individuals despite the difference in the composition of different food webs. By comparison, there was evidence of differences in the path trajectories for the comparisons of the isotopic niches of the catfish populations in different localities. This suggests that the catfish exhibited plasticity in its niche depending on resource availability within the invaded localities.

In order to detect causal patterns associated with the catfish, a Multiple Before-After Control-Impact (MBACI) experimental design was used to examine macroinvertebrate communities within two rivers that were differentially impacted by catfish. Comparisons of the impact within the two rivers suggest contrasting response patterns. Within a river that had high ambient density of catfish, there was a significant increase in mean diversity and mean richness in the Control treatment that excluded catfish. This suggests that the macroinvertebrates responded more to catfish exclusion, which inferred community flux within catfish exclosures in localities with a high density of the invasive predator. No evidence of differences was found for mean dry mass suggesting that the macroinvertebrate community was dominated by taxa that were predation resistant. By contrast, within a river with no catfish, the patterns of macroinvertebrate communities were more consistent until disruption due to a press-type disturbance caused by the introduction of catfish in the Impact treatment, providing evidence of the catfish impact. These patterns were supported by multivariate analysis.

**Friday 7 October 2011**

**Humphrey Greenwood Memorial Lecture**

**08h00-08h20: Welcome and introduction by Acting Head of Department, Prof. Tony Booth**

**08h20-09h00: Guest speaker - Dr Malcolm Smale - Marine apex predators: advances in understanding the ecology of white and tiger sharks**

## **Marine ecology, conservation and related biology continued (Chair: A Chakona)**

**09h00-09h20: Sisanda Mayekiso (MSc student) - Population genetic structure and biogeography of three wrasse species (Labridae) within the Western Indian Ocean**

Supervisors: Dr G Gouws (g.gouws@saiab.ac.za); Dr M Mwale (m.mwale@saiab.ac.za)

Funder: DST-African Coelacanth Ecosystem Programme (ACEP); South African Institute for Aquatic Biodiversity (SAIAB)

The Western Indian Ocean (WIO) possesses a diverse marine fauna and its regional components shows substantial endemism. However, an understanding of the origins and relationships of these regional faunas, and the processes that have led to their establishment is lacking. The WIO is an exceptional model for studying factors and processes for shaping species diversity and intraspecific variation over a large geographic scale, because there are known biogeographic breaks and oceanographic features that influence the genetic structure of marine species. A molecular genetic approach was used to provide insight into patterns of contemporary and historical connectivity, isolation of the regions, and the origins and relationships of the regional WIO faunas. This has potential implication for accurate interpretation of the origins for the WIO regional faunas.

The objectives of this study were to firstly examine genetic connectivity and regional differentiation for each species and the processes that has led to these establishments. Secondly, to observe genetic connectivity, migration and gene flow of each species among the different biogeographical regions of the WIO, and lastly to explore the factors that might have influenced the distribution of each target species in the WIO.

Three reef associated species of the family Labridae namely (*T. lunare* and *T.hebraicum*) and *Cheilio inermis* were selected for this study. These species are good study models because they are widespread, abundant in the WIO. Specimens were collected from numerous WIO localities. PCR and fluorescent dye terminator sequencing were used to amplify and sequence two mitochondrial genes (cytochrome *b* and ATPase 6) and one nuclear gene (first intron of the nuclear *S7* ribosomal protein gene). Statistical analyses were done in a phylogenetic and biogeographic framework. Traditional population genetic analyses were used to calculate haplotype diversity and sequence divergences within species and among regions of the WIO. Population structure was analysed using AMOVA. Phylogenetic relationships were examined using parsimony and maximum likelihood procedures.

The results of the present study were broadly comparable to a pilot study that examined different gene regions (16S and COI) with less extensive sampling. *Thalassoma hebraicum* revealed low levels of regional differentiation within the WIO. On the other hand, high connectivity and panmixia was found for *T. lunare*. Low mtDNA differentiation was characteristic within species. Other studies have also found high gene flow within the WIO and low genetic structuring.

**09h20-09h40: Jerraleigh Kruger (MSc student) - A morphological comparison of *Spondyliosoma* species and *Lithognathus mormyrus* from Angola and South Africa.**

Supervisors: Dr WM Potts (w.potts@ru.ac.za); Dr G Gouws (g.gouws@saiab.ac.za)

Funders: National Research Foundation – South African Biosystematics Initiative (74457); The South African Institute for Aquatic Biodiversity (SAIAB)

The Benguela Current and its associated upwelling cells formed approximately 2 million years ago. The cold upwelling cell between Angola and South Africa has been responsible for the genetic isolation of various southern African fish species with warm temperate distributions. In isolation, morphological change can occur at different rates between different taxa, which are variably referred to as robust or plastic species. This study aims to test if the genetic isolation caused by the upwelling cell has been expressed phenotypically in three species that are considered morphologically plastic. *Lithognathus mormyrus* has a disjunct distribution from the Mediterranean along the west coast of Africa to South Africa. *Spondyliosoma* has a similar distribution with *S. emarginatum* occurring to the south and *S. cantharus* to the north of the cold upwelling cell.

A total of 90 specimens of *S. cantharus* were collected from Angola and 107 specimens of *S. emarginatum* were collected from South Africa. Seventy three and 74 specimens of *L. mormyrus* were collected from southern Angola and South Africa respectively. Twenty three morphological measurements and six meristic counts were taken per individual. The data were log-transformed and Principal Component (PCA) and Discriminant Function (DFA) Analyses were performed comparing *S. cantharus* (Angola) and *S. emarginatum* (South Africa), and South African and Angolan populations of *Lithognathus mormyrus*.

The PCA showed no meristic differentiation between the *L. mormyrus* populations with counts either being identical or having overlapping ranges. Morphometric measurements compared using PCA showed insignificant variance between the populations of *L. mormyrus*. The meristic features of *S. emarginatum* and *S. cantharus* were strongly separated (99.2% variance) in the PCA by the lateral line scale count, which ranged from 80-92 and 65-68, respectively. Despite this there was a low degree of variation in the morphometric characters. The DFA agreed with the separation of *S. emarginatum* and *S. cantharus*, with the strongest differentiation being provided by the lateral line scales. *Lithognathus mormyrus* separates into two groups according to the DFA, but due to similarities between the populations the DFA analysis could not determine diagnostic characters.

There was a high degree of morphological variation within the *Spondyliosoma* and *Lithognathus mormyrus* populations. These preliminary analyses suggest that phenotypic expression has occurred due to genetic isolation and it is also possible that speciation has occurred in both groups. Given that these *Spondyliosoma* species and *Lithognathus mormyrus* populations were isolated at the same time these results suggest that morphological change does not occur at the same rate in these closely related genera. However, because of the high levels of variation, more analysis has to be done to test if any of the characters are diagnostic between the two populations and variation within populations must be compared to between populations.

**09h40-10h00: Moqebelo Morallana (MSc student) - Phylogenetic relationships within the genus *Lutjanus* and the placement of the Western Indian Ocean snappers in context to the wider Indo-Pacific.**

Supervisors: Dr G Gouws (g.gouws@saiab.ac.za); Dr M Mwale (m.mwale@saiab.ac.za)

Funders: DST-African Coelocanth Ecosystem Programme, South African Institute for Aquatic Biodiversity (SAIAB)

Lutjanids (snappers) are reef and bottom-associated fish occurring in tropical and sub-tropical marine waters throughout the eastern Pacific, Indo-West Pacific, and western and eastern Atlantic. The genus *Lutjanus* has 65 species worldwide with 15 species of these species found in the Western Indian Ocean. Two molecular phylogenies have been published for the genus, but not all species were represented in both studies and these have had very limited geographic coverage, focussing on the South China Sea (SCS) and West-Pacific (WP), respectively. Thus, the placement of Western Indian Ocean (WIO) snappers is still unknown. Other studies have also shown limited differentiation in certain species (e.g., *L. fulviflamma* and *L. kasmira*) at smaller geographic scales, suggesting that differentiation among WIO representatives and conspecifics from the IWP requires investigation.

The aim of the study is to (1) determine the placement of the Western Indian Ocean snappers in the context of the wider Indo-Pacific, and (2) to examine the differentiation among the WIO representatives and IP representatives of the same species. Moreover, the current study includes additional species not included in the published studies, expanding the data set.

Samples were obtained from National Fish Collection at the South African Institute for Aquatic Biodiversity, spanning the wider WIO. Additional species were obtained from museums elsewhere. DNA sequences from the two published studies were downloaded from GenBank for comparison, and COI sequences were obtained from the BOLD database. For newly included specimens, DNA sequence data were generated from two mitochondrial genes (16S and COII). These data sets were combined with published data to determine the phylogenetic relationships within the genus *Lutjanus*. Maximum parsimony, maximum likelihood and neighbor-joining were used for each data set on its own and combined.

COI data consisted of 60 taxa with 630 base pairs (bp) long sequences, COII had 34 taxa with 739 bp long sequences, and 16S had 52 taxa with 559 bp long sequences. The 16S results clustered the newly-included *L. malabaricus* and *L. gulcheri* close to each other. *Lutjanus lemniscatus*, *L. lutjanus* and *L. madras* were sister taxa to a “yellow-lined and black-spotted” species complex. Other new species clustered within the IP representatives. The COII results grouped *L. bengalensis*, *L. kasmira*, *L. notatus* and *L. quinquelineatus* as one cluster. The COI data grouped same species together. In summary, same species representatives from the WIO grouped together with their IP counterparts. The phylogeny of the 16S and COII were similar to published phylogenies in terms of groupings found and additional taxa did not alter these groupings. These results are consistent with morphology-based identifications, with strong correlation between molecular and morphological characteristics.



**10h00-10h20: Christine Coppinger (MSc student) - Assessing genetic diversity of catface rockcod (*Epinephelus andersoni*) in the subtropical Western Indian Ocean and modeling the effects of climate change on their distribution**

Supervisors: Dr N James (n.james@saiab.ac.za); Dr M Mwale (m.mwale@saiab.ac.za)

Funders: Western Indian Ocean Marine Science Association, National Research Foundation

*Epinephelus andersoni* is endemic to the south-east coast of Africa, with a distribution range extending from Quissico in Mozambique to Kynsna in South Africa. It is an important recreational and commercial fishery species, particularly in Kwa-Zulu Natal and has been under intense fishing pressure. Due to its complicated life history, this species is particularly vulnerable to over-exploitation. There has been very little research done on this species and no proper fishery assessment on which to base fishing regulations. More research on this species is needed, particularly at a time of potentially significant climatic changes, which could put further pressure on this endemic species. This project aims to determine the genetic diversity of *E. andersoni*, to model the current distribution of this species and to project the distribution model by applying it to future climatic layers.

The genetics of this species will provide information on levels of connectivity and migration patterns which are unknown. Analysing the present and historical patterns of genetic diversity and structuring will help to determine potential barriers to their movement and whether the species may be more vulnerable to change than assumed. This combined with niche modelling will allow us to estimate and predict how *E. andersoni* may react to future climate change. This study will provide important information for the management of this species.

Thus far 203 genetic samples of *E. andersoni* have been collected from the entire range of its distribution and DNA has been extracted from 193 of these using standard analytical procedures and kits. Polymerase chain reactions (PCRs) have been done using four sets of primers: L15242 and H16458; CNCR1-F and CNCR1-R; L15803 and H677; L16560 and H677 to amplify the control region of mitochondrial DNA and the most successful primer pair so far has been using universal primers L16560 and H677. DNA sequences of 22 individuals of approximately 300 base pairs in length have been successfully done. Sequence analysis of the control region data is underway and further reactions will be done to amplify the first intron of the S7 ribosomal nuclear gene. Both gene regions will be aligned and analysed using several genetic statistical software packages.

Niche modelling techniques will be used to determine range shifts of *E. andersoni* under future climate change scenarios. Species distribution data will be collected from various sources including fishbase; environmental data such as NOAA\_OI\_SST\_V2 data will be provided by the NOAA/OAR/ESRL PSD, Boulder, Colorado, USA, from their Web site at <http://www.esrl.noaa.gov/psd/>. The linear trend with SST spatially for each month of the year will be estimated from 30 years of data, and then what the SST would be in 30 or 50 years will be calculated using the trend of the last 30 years.

**10h20-10h35: Tea break**

## **Mariculture (Chair: D Kahwa)**

**10h40-11h00: Nani Rossetti (MSc student) - The development of a “least-cost” diet for dusky kob, *Argyrosomus japonicus* (Pisces: Sciaenidae)**

Supervisors: Dr CLW Jones (c.jones@ru.ac.za); Prof. P Britz (p.britz@ru.ac.za)

Funders: THRIP; Marifeed (Pty) Ltd

Fish meal and fish oil are commonly used as ingredients in finfish feed and, with declining fisheries resources worldwide, such ingredients are becoming less available and more expensive. Therefore it is essential to explore the utilization of alternative ingredients. The aim of this study is to investigate the use of vegetable oil as a sustainable and competitive alternative to fish oil in marine finfish feed.

Six diets (18 % total lipid and 46 % protein) were fed to juvenile kob. Each with a graded decrease in fish oil contribution to total lipid, with a corresponding increase in vegetable oil contribution of 1, 14, 28, 42, 56 and 70 %.

There were no significant differences in fish length and weight among treatments after 84 days, with overall means ( $\pm$  standard error) of  $239.9 \pm 13.26$  g (Kruskal-Wallis:  $H=9.9$ ;  $p=0.08$ ) and  $252.3 \pm 04.3$  mm (ANOVA:  $F=3.14$ ;  $p=0.07$ ), respectively. However, there was a significant trend of a decrease in specific growth rate (SGR) (range:  $0.2 \pm 0.01$  to  $0.3 \pm 0.02$ ) with an increase in vegetable oil replacement ( $r^2=0.35$ ,  $p=0.019$ ). Similarly, condition factor (CF) decreased from  $1.59 \pm 0.03$  to  $1.53 \pm 0.02$  with an increase in vegetable oil level ( $r^2=0.32$ ,  $p=0.027$ ). There were no differences in red blood cell count, haematocrit and haemoglobin concentration, with overall means of  $2.76 \pm 0.1$  ( $X^2=0.03$ ,  $p=0.99$ ),  $0.27 \pm 0.04$  % (Kruskal-Wallis:  $H=3.8$ ,  $p=0.6$ ) and  $6.9 \pm 0.8$  g dL<sup>-1</sup> (ANOVA:  $F=0.4$ ,  $p=0.8$ ), respectively. Visceral fat index (VFI) increased significantly from  $1.28 \pm 0.2$  to  $2.24 \pm 0.1$  (ANOVA:  $F=5.24$ ,  $p=0.009$ ;  $r^2=0.54$ ,  $p=0.0005$ ) and hepatosomatic index (HSI) also increased from  $0.83 \pm 0.07$  to  $1.80 \pm 0.12$  (ANOVA:  $F=9.9$ ,  $p=0.0006$ ;  $r^2=0.5$ ,  $p=0.001$ ) with an increase in vegetable oil replacement.

Fish fed diets with 42 to 70 % substitution showed greater number of lipid vacuoles in the liver, which were also larger in size, and hepatocytes nuclei were displaced to the cell periphery. Linoleic acid accumulated in the liver, increasing from 5.48 to 42.05 % in fish fed the control diet and those fed the diet with 70 % substitution. There were significant differences in percentage of EPA in the liver, ranging from 13.63 to 1.97 % for 1 to 70 % vegetable diets, respectively (Kruskal-Wallis:  $H=50.33$ ,  $p<0.01$ ). Similarly, liver DHA range from 14.34 to 3.28 % for the 1 to 70 % treatments (ANOVA:  $F=97.37$ ,  $p<0.01$ ). The n-3/n-6 ratio was significantly reduced with inclusion of vegetable oil in the diets (Kruskal-Wallis:  $H=51.55$ ,  $p<0.01$ ).

The trend of decreasing growth rate with increasing oil replacement towards the end of the trial corresponds with increases in VFI, HSI and lipid vacuoles in the liver. This suggests that dusky kob is less able to metabolise soybean oil at increased substitution levels (with a cut-off at around 28 %), which would account for the poorer growth at higher levels. The accumulation of fatty acids with C<sub>18</sub> carbons in the liver suggests that kob are unable to synthesis EPA and DHA from these fatty acids.

Espadon Marine (Pty) Ltd is acknowledged for making fish available for this research.

**11h00-11h20: Maryke Musson (MSc student) - Finding the Aquaculture ‘Sweet Spot’ where research input improves business output with regards to the commercialisation of dusky kob, *Argyrosomus japonicus***

Supervisor: Prof. H Kaiser (h.kaiser@ru.ac.za)

Funders: Marine and Coastal Management Frontier Fund; Espadon Marine (Pty) Ltd

*Argyrosomus japonicus*, commonly also known as dusky kob, kabeljou or mullet, is one of the best-known angling fishes around the South African coast. Dusky kob carries high potential for large-scale commercial production in land-based systems and cage culture. The application of research should play an important role in optimising commercial production of kob, and to attract investors and confidence in marine fin fish farming in South Africa.

Research projects have determined optimal growth criteria for dusky kob, and such results can create ‘paper fish’, i.e., business models for funding applications. Scientific research should be merged with on-farm accounting management to develop farming practices that deliver production results and support the establishment of a viable business. Recent and current research outputs to increase production efficiency include:

Temperature: Best growth was achieved at 25.3° C; optimal FCR at 21.4°C and temperature preference ranged from 25-26.4 °C. Commercial production shows good growth, optimal FCR and most cost effective oxygen utilization at a temperature range of 19-21°C.

Egg and larval selection to improve batch quality: Egg and larval quality could predict larval growth and survival. The selection of high-quality eggs and larvae reduces the production cycle period and fish mortality.

Defining weaning requirements: Dusky kob larvae can be weaned onto specialized artificial diets from 10 days post hatch. This reduced the dependency on live cultures during larval rearing.

Defining feed nutritional requirements: Diets to achieve best growth in dusky kob have been optimized to ensure raw material sustainability and cost-efficiency.

Effect of light intensity on production efficiency: Increased growth and feeding at partially shaded light levels. Commercial producers may now consider adequate light levels for improved tank maintenance as well as photoperiod management for optimal feeding.

To ensure the growth of an aquaculture industry in South Africa, industry requires the research support from academic institutions. Research should contribute to financial modelling, as the commercial aim of any business would be to get to market in the shortest possible time within the most cost-effective parameters. Appropriate research objectives will assist in turning ‘paper fish’ into a marketable product while optimizing the “cost to production ratio”.

**11h20-11h40: Kerry Pieterse (MSc student) - Fish haematology – investigating the use of an immunostimulant in fish culture of dusky kob, *Argyrosomus japonicus***

Supervisors: Dr H Kaiser (h.kaiser@ru.ac.za); Dr W Vermeulen (wav@wrm.co.za)

Funders: Welbedagt Research and Manufacturing; Rhodes University JRC

Disease outbreaks are encountered in the rapidly developing aquaculture industry. This is largely due to the stressful environment that the fish are exposed to under intensive aquaculture conditions, which compromises the immune system of the animal. Prevention and treatment of diseases and pathogens is usually via vaccines, antibiotics or chemotherapeutants. These methods have limited success with the emergence of drug-resistant bacteria, ineffectiveness, high costs and unwanted environmental impacts. As prevention is better than treatment, the use of immunostimulants to enhance specific and non-specific immunity offers a promising alternative to antibiotics and vaccines. The use of immunostimulants has been found to improve the specific and non-specific immune defence of aquatic animals, to provide increased resistance to pathogens and to improve growth rates and survival. The first part of this study will evaluate the efficiency of Fish Assist® as an immunostimulant for dusky kob, *Argyrosomus japonicus*. In addition, the effect of high light intensity as a stressor to dusky kob will be studied by observing immune response markers and differential blood cell counts.

The research thus far has been based on an exploratory approach. Observing and analyzing blood leucocytes, haematocrit testing and various histological assessments. The experimental study will be undertaken once the immunostimulant has been incorporated into a diet.

The experiment will be conducted by administering the immunostimulant to the fish either as part of the artificial diet or as capsules. In both the experimental and control groups there will be the inclusion of a stressor (e.g., light intensity or temperature fluctuations) on some of the treatments. At the end of the trial fish will be collected for blood samples to test haematocrit, leucocytes and study haematology. Growth and survival will also be determined.

**11h40-12h00: Nicholas Riddin (MSc student) - Gonad development and growth in farmed South African abalone *Haliotis midae***

Supervisors: Dr CLW Jones (c.jones@ru.ac.za); Prof. H. Kaiser (h.kaiser@ru.ac.za)

Funders: National Research Foundation; Aquafarm Development (Pty) Ltd; HIK Abalone (Pty) Ltd; Marifeed (Pty) Ltd; Roman Bay Sea Farm (Pty) Ltd

The economic viability of commercial abalone farming depends on the system used for grow-out, and, due to the long culture period and slow growth of abalone, the profit can be marginal in land-based systems. Thus, maximizing growth rate and minimizing grow-out period I expected to improve the profit margin. According to farm records, abalone on farms appear to be growing slower during certain periods of the year. This is affecting the abalone in the weight range of 50-70 g / abalone the most, with winter growth rates being 10% lower than those in summer. This reduction in growth rate seems to coincide with enlarged gonads in the farmed abalone. It should be tested whether ingested energy is being spent on gonad development instead of somatic growth. The aim of this research is to understand the environmental or dietary causes for the above-mentioned observation, and to find a method of controlling it with an economically viable farm-scale solution.

Individual abalone ( $53.32 \pm 6.27$  g / abalone and  $62.76 \pm 2.37$  mm) were measured monthly over the course a year to determine growth. Mean monthly length and weight gains did not differ significantly between the two farms, but showed seasonal variation ( $F = 4.19$ ,  $p = 0.0001$ ). Similarly, seasonal trends in percentage viscera as a proportion of body size were noted ( $F = 5.61$ ,  $p = 0.00001$ ) which may suggest distinct spawning periods. These data will be correlated to environmental variables to determine whether any of these factors influence abalone growth rate or gonad development on the farms. Abalone from Aquafarm Development (Pty) Ltd. had significantly larger gonads than those from HIK Abalone (Pty) Ltd. ( $F = 17.83$ ,  $p = 0.00003$ ), while the meat yield did not differ significantly between the two farms ( $F = 0.007$ ,  $p = 0.93580$ ).

The last component of this study aims to determine the effect of dietary energy level on gonad growth and development. Maturing ( $48.76 \pm 3.59$  g) abalone were fed one of seven experimental diets with a range of protein to energy ratios and protein for 262 days. Growth and gonad development of animals will be compared between treatments. Experimental diets compared the use of fishmeal and soya as protein sources, with commercially used protein and energy levels, as well as diets with reduced energy. Cannery cook-out yields will be compared to determine whether the treatments have any effect on cook-out weight loss. An economic model will be designed to determine the economic impact of a farm-scale diet change.

**12h00-12h20: Gareth Nicholson (MSc student) - Investigating the effects of stocking density on the health, growth and production of South African abalone (*Haliotis midae*)**

Supervisors: Dr CLW Jones (c.jones@ru.ac.za); Prof. H Kaiser (h.kaiser@ru.ac.za)

Funders: Marifeed (Pty) Ltd; Aquafarm Development (Pty) Ltd; HIK Abalone Farm (Pty) Ltd; Roman Bay Sea Farm (Pty) Ltd; THRIP

The profitability of abalone farms is heavily influenced by their production per unit of grow-out space. Production is a function of abalone growth and stocking density per unit of grow-out space. With farms having physically expanded to the maximum and with increasing costs of production (electricity, maintenance etc.) the only realistic way for an abalone farm to increase its production is through optimizing stocking densities. There is no documented research with regard to stocking density and its affect on the performance of *H. Midae* and optimal stocking densities for this species have not yet been determined. Preliminary on-farm research suggests that stocking densities may be increased until a threshold is reached at which growth rates and animal health may be reduced.

The primary aim of this study is to develop a better understanding of optimal stocking densities for different size abalone, by determining the effect of stocking density on the growth, health and production of *H. midae*, when water quality is not a confounding factor. The study will also attempt to determine what causes a reduction in growth at high stocking densities and try to establish measures to counter this reduction.

Experiments are currently being carried out on the Aquafarm Development and HIK Abalone farms in Hermanus. The effects of four different stocking densities are being tested on three different size classes of abalone. Each size class constitutes an individual experiment with each density for the size class representing a treatment. The tested stocking densities are 16 %, 20 %, 22 % and 24 % of the available surface area in each basket and each treatment will be replicated four times. The examined size classes are 15-35 g, 45-65 g, and 75-95 g. Experimental trials will run over a period of eight months with measurements being made at four month intervals. Growth and abalone performance will be calculated in terms of weight gain, SGR, FCR, PER, percent biomass gain and condition factor. Ten abalone replicate<sup>-1</sup> will be sacrificed at each measurement interval in the 45 – 65 g size class and sent to the farms health assessor for detailed health analyses. Indices such as canning yields, drip loss and condition factor will be taken at the end of the trial and used as other indicators of health.

During the eight month trial period, abalone behaviour will be observed in an attempt to establish what could be causing a decrease in growth at higher stocking densities. This information will be used for the second part of the study in which further experiments will be set up to test whether any of these suspected causes are in fact resulting in decreased levels of growth.

**12h20-12h40: Justin Kemp (PhD student) – Nutritional physiology of South African abalone (*Haliotis midae*)**

Supervisor/s: Prof. PJ Britz (p.britz@ru.ac.za)

Funder/s: Marifeed (Pty) Ltd; National Research Foundation; Rhodes University Henderson Fund; Rhodes University JRC

Abalone feed production levels have grown from approximately 300 tons in 2005 to 1070 tons in 2010 as more and more farms have switched from harvested seaweed (kelp *Ecklonia maxima*) to formulated feeds. Commercial farms use approximately 1.3kg of Abfeed® to produce 1kg of abalone, which suggests that the 1070 tons of Abfeed® produced in 2010 accounted for 823 tons, or 77%, of total abalone production. These figures highlight that by far the majority of abalone in South Africa are now produced using formulated feeds, and as such the provision of high quality formulated feeds is imperative to the continued success of the industry. Furthermore, given that the kelp resource is already fully subscribed in the regions where abalone are farmed, any future growth in the industry will rely on formulated feeds, either fed exclusively or under a mixed-diet regime in conjunction with seaweeds. Despite the availability of a locally produced abalone feed, and significant research on nutritional formulations, including the reduction of the dietary fishmeal and protein content, we know surprisingly little about the molluscan digestive process, particularly the utilisation of seaweed. Abalone are herbivores in their natural environment, feeding on a variety of green, brown and red seaweeds, and have been shown to grow particularly well on a mixed seaweed diet under culture conditions.

A growth trial is currently underway assessing the role of fresh seaweed supplementation into the diet of animals fed a formulated feed. Five graded supplementation levels of a “mixed bag” of fresh seaweed composed of ulva, gracilaria and kelp are being tested under controlled optimum conditions at the Port Alfred Marine Research Laboratory. Production, health, metabolic, histological and potentially microflora indicators will be assessed.

This trial will also be an essential part of the extended acclimation period for metabolic studies investigating the effect of diet on the metabolic costs of digestion. Furthermore, given the highly refined nature of the starch component of formulated feeds currently used in the industry, the possibility of hyperglycemia in abalone will be investigated. The protocol for this trial will be presented in detail.

## **Fisheries and fisheries related biology continued (Chair: D Kahwa)**

**12h40-13h00: Nicola Downey (PhD student) - A first description of the offshore reef environment off the Eastern Cape coast, South Africa, with reference to deep spawning chokka squid (*Loligo reynaudi* D'Orbigny, 1839)**

Supervisor/s: Prof. W Sauer (w.sauer@ru.ac.za); Dr M Roberts (squid@metroweb.co.za)

Funder/s: National Research Foundation ( KFD2008062700004); South African Squid Management Industrial Association

The South African chokka squid, previously thought to be a shallow water inshore spawner, has also been found to spawn at depths of 71 to 270 m. Inshore spawning has been studied in detail. Mid-shelf spawning however, is still very much an unknown phenomenon. This project aims to determine where (substrate and general morphology of mid-shelf spawning grounds) and why (environmental conditions) squid spawn offshore, further our knowledge of mid-shelf spawning, assess the importance of mid-shelf spawning (recruitment) and determine whether mid-shelf spawning is a separate process from that which occurs inshore or is an offshore continuum.

Department of Agriculture, Forestry and Fisheries demersal trawl data was used to determine the contribution of mid-shelf spawned eggs to total egg biomass. Filament and acoustic telemetry tagging studies were used to investigate squid movement between the mid-shelf and inshore spawning grounds and movement on the mid-shelf spawning grounds, respectively. Temperature and turbidity data were collected to compare inshore and mid-shelf spawning environments. A benthic survey, during which echosounder and CTD data was collected and an underwater camera deployed, allowed for mapping of bathymetry, substrate types and morphology of the mid-shelf spawning grounds.

Mid-shelf spawned eggs were found to contribute an estimated 18% to total squid egg biomass. An analysis of environmental data showed fewer turbidity events to occur on the mid-shelf compared to inshore. Bottom temperatures on the mid-shelf were generally colder and more stable than inshore. On occasion, due to downwelling, the mid-shelf bottom temperatures were slightly warmer than bottom temperatures inshore. Substrate types and morphology differ substantially between the two spawning environments, with the mid-shelf consisting mainly of rocky reef. The movement of squid between these two areas indicates the existence of a single population as opposed to an inshore and offshore population.



## **Non-presenting students**

### **Siyabonga Maliza (Masters student) - Development of probiotic diets for the South African abalone (*Haliotis midae*) industry**

Supervisors: Prof. H Kaiser (h.kaiser@ru.ac.za); Dr CLW Jones (c.jones@ru.ac.za); Prof. PJ Britz (p.britz@ru.ac.za)

Funders: Department of Agriculture Forestry and Fisheries; National Research Foundation

In farm-cultured abalone stress can lead to immunosuppression and increases the susceptibility to bacterial, viral and parasitic disease, often followed by mortality. Thus, handling and poor water quality can reduce farm production efficiency. Probiotics in aquaculture have been effective in a wide range of species in enhancing immunity, survival, improving feed utilisation and growth. Three putative probionts identified as a result of *in vitro screening* were beneficial to laboratory-reared abalone.

The aim of this study was to produce an abalone feed that contains a suite of probionts that promote abalone growth and health under the stress of farming conditions. Objectives were to a) compare growth, survival and physiological responses of abalone fed Abfeed®S 34 and probiotics (i.e., the probiotic diet) to abalone fed Abfeed®S 34 as a control treatment in a factorial design with handling stress as the second independent variable.

Growth experiments were conducted at HIK Abalone Farm (Pty Ltd) and Roman Bay Sea Farm (Pty) Ltd after which haemocyte and phagocytosis counts were done. (1) To determine the effect of probiotic diet on abalone growth and health under presumed stressful conditions, abalone at HIK Abalone Farm were fed either the test or the control diet and subjected to normal farm handling or to additional handling stress.

At HIK there was no significant interaction between diet and handling on average length and weight gain month<sup>-1</sup> after 4 and 8 months ( $p > 0.05$ ). Average length (stressed =  $73.9 \pm 0.52$ , unstressed =  $75.8 \pm 0.57$  mm) and weight gain (mean: stressed =  $68.5 \pm 1.20$ , unstressed =  $74.3 \pm 1.86$ ) increased significantly in unstressed animals after 8 months ( $p < 0.02$ ). After 8 months phagocytosis count was significantly different between dietary treatments with values of 74 and 53.5 counts per sample for the probionts and control treatment, respectively ( $p < 0.02$ ). There was no effect of stressor application ( $p = 0.14$ ) and no interaction between dietary treatment and stressor application for this variable ( $p = 0.61$ ). There was no difference in feed conversion ratio between treatments with values ranging from 2.9 to 3.8:1.

At Roman Bay Sea farm, average length ( $68.183 \pm 1.460$ ) and weight ( $53.560 \pm 4.272$ ) of animal fed probiotic diet significantly improved over time ( $p < 0.00001$ ), compared to those fed the control with length ( $67.856 \pm 1.516$ ) and weight ( $52.450 \pm 3.616$ ) after 8 months. There was no significant difference in haemocyte counts between animals fed either probiotic or control diet after 4 and 8 months ( $p > 0.05$ ).

The non-specific immune response in abalone fed the probiotic diet after four months, given as phagocytosis count, differed between dietary treatments, while after eight months there was no difference at HIK Abalone Farm. There was no difference in average haemocyte counts between animals fed either probiotic or control diet after 4 and 8 months on both farms. Abfeed® S34 enriched with a mixture of probionts showed a significant improvement in growth and survival of farmed abalone. Probiotics showed no significant improvement on health parameters examined in this study after eight months.

**Siviwe Babane (MSc student) - Effect of size variation on aggressive and feeding behaviour of juvenile dusky kob *Argyrosomus japonicus***

Supervisor/s: Prof. H Kaiser (h.kaiser@ru.ac.za)

Funder/s: Marine and Coastal Management (Department of Environmental Affairs and Tourism); Espadon Marine, East London

Dusky kob *Argyrosomus japonicus* belong to the family Sciaenidae which includes carnivorous fish that are normally found in temperate regions in shallow coastal and estuarine waters. Dusky kob juveniles are, therefore, aggressive and this behaviour likely contributes to the high mortality being reported for this species. Cannibalism in dusky kob occurs from day 18 in intensive culture and becomes less problematic when fish have reached 80 mm total length. Size grading is commonly used to improve growth of small fish, reduce cannibalism, decrease size variability and improve feed uptake. The main aim of this technique is to improve weight gain by all individuals and to increase survival rate. Many studies have been conducted on the effect of size-grading in other fish species, however, there is a paucity of information on the effects of size-grading in rearing dusky kob. Thus, a study focusing on the effect of size variation and the time at which fish were observed on juvenile dusky kob aggressive and feeding behaviour was conducted to reduce cannibalism.

The aim of this work was to better understand factors determining cannibalism in juvenile dusky kob. The objective of this study was to determine the effects of size variation and observation time on behaviour of this species under intensive rearing conditions.

Following results from the previous work which showed that size variation influenced aggressive and feeding behaviour of dusky kob using repeated measures ANOVA, a trial was undertaken with very small juveniles to study the influence of the aggressor's size on its behaviour. Dusky kob juveniles were donated from the hatchery stock of Espadon Marine in East London. Coefficient of variation (CV) was used to determine the size variation in each treatment. The treatment, in this case, referred to the four groups of non focal fish (smaller fish) with different values of size variation. Each focal fish (largest fish) was exposed to four treatments and the sequence of observations was randomised. Thirty-minute behaviour observations were conducted to address aspects of aggressive and feeding behaviour. The largest fish was observed before and after feeding to record its aggression. Preliminary analyses suggested that the time at which fish were observed affected aggression, with higher mean frequencies recorded in observations conducted 12 hours after feeding in the evening than observations made before feeding in the morning. On overall, positive correlations between aggressive behaviour observed/30 min. fish<sup>-1</sup> and size variation (CV) were found through the use of least squares regression analysis. Thus, aggression seemed to increase with increasing size variation. Further studies are recommended to assess how size variation and aggressive behaviour are correlated to each other and to confirm the present findings.

**Matthew Naylor (MSc student) - Water quality and abalone, *Haliotis midae* L. production in a serial-use raceway system**

Supervisors: Prof. H Kaiser (h.kaiser@ru.ac.za); Dr CLW Jones (c.jones@ru.ac.za)

Funders: National Research Foundation; HIK Abalone Farm (Pty) Ltd; Aquafarm Development (Pty) Ltd; Roman Bay Sea Farm (Pty) Ltd

The concentrations at which specific water quality variables affect abalone growth are not well documented. In production systems water quality parameters are altered simultaneously by the metabolic activity of the culture animals.

Trial 1 described the changes in water quality between tanks in a serial-use raceway in relation to accumulated biomass and water flow. Three serial-use raceways each with seven tanks in series were used. A sample of 50 abalone per tank were individually weighed and measured at the start and end of the trial. Growth reductions (ANOVA;  $F_{6, 14} = 13.92$ ,  $p < 0.0001$ ) and increased FCR values (Kruskal-Wallis test;  $H_{(6, 21)} = 16.35$ ,  $p = 0.012$ ) were observed for abalone in the lower tank positions. pH and dissolved oxygen were negatively correlated with tank position. FAN and nitrite concentration were positively correlated with tank position. It is estimated that a flow index of  $7.2 - 9.0 \text{ L h}^{-1} \text{ kg}^{-1}$  will cause significant growth reductions in *H. midae*.

Trial 2 determined the effect of dietary protein level on TAN and FAN concentrations in a serial-use raceway. Three iso-energetic diets containing 33%, 26% and 22% protein were used. Temperature, pH, TAN and FAN were measured over 17 days. TAN and FAN concentrations were correlated with dietary protein ( $t = 6.63$ ,  $p < 0.0001$  and  $t = 6.41$ ,  $p < 0.0001$ , respectively) and flow index ( $FI = \text{L h}^{-1} \text{ kg}^{-1}$ ) ( $t = 5.42$ ,  $p < 0.0001$  and  $t = 3.9$ ,  $p < 0.0002$ , respectively). Mean FAN concentrations in tanks fed the 22% and 26% protein diets, were 66% and 41% lower than in tanks fed the 33% protein diet.

Trial 3 determine the effect of FAN concentration on *H. midae* growth. Two iso-energetic diets, containing 33% and 26% protein were used to create a high FAN and low FAN treatment respectively. No significant differences were observed between treatments (ANOVA;  $F_{1, 14} = 0.47$ ,  $p = 0.51$ ), however growth was significantly reduced with increasing tanks in series (ANOVA;  $F_{2, 14} = 85.61$ ,  $p < 0.0001$ ). Supplementation with pure oxygen to  $\pm 100\%$  saturation did not significantly improve growth (T-test;  $t = -2.61$ ,  $df = 4$ ,  $p = 0.06$ ) in abalone exposed to high FAN and low pH.

Trial 4 determined the effect of dosing with sodium hydroxide on water pH and abalone growth in a serial-use system. Increased pH resulted in higher weight gain ( $F_{1, 12} = 4.51$ ;  $p = 0.055$ ) and length gain ( $F_{1, 12} = 4.56$ ;  $p = 0.054$ ) at an  $\alpha$ -error level of  $< 5.5\%$ . FCR ( $F_{1, 12} = 2.93$ ;  $p = 0.11$ ) and condition factor ( $F_{1, 12} = 1.98$ ;  $p = 0.18$ ) were not influenced by dosing with NaOH. Weight gain (Wg) and length gain (Lg) were significantly correlated to water pH (Wg:  $F_{1, 16} = 16.4$ ,  $p = 0.0009$ ,  $r^2 = 0.51$ ; Lg:  $F_{1, 16} = 16.2$ ,  $p = 0.0009$ ,  $r^2 = 0.50$ ) and multiple regression models of pH, dissolved oxygen and FAN suggested that pH was the best predictor of growth.

**Enrico Gennari (PhD student) - Thermal physio-ecology of the white shark *Carcharodon carcharias***

Supervisors: Dr PD Cowley (p.cowley@saiab.ac.za); Mr R. Johnson (r.johnson@oceans-initiative.com)

Funders: Oceans Research; South African Institute for Aquatic Biodiversity (SAIAB); Padi Aware

The white shark is one of few elasmobranch species capable of maintaining parts of its body warmer than the surrounding environment. This ability enhances among other things, its neural activity, vision, swimming stamina and decreases digestion time, making it an effective apex predator. Very little is known about the thermal physiology of white sharks in the wild, since studying these large, highly mobile and potentially dangerous predators, often in rough sea conditions, is challenging.

This project will make use of acoustic telemetry methods to determine whether white sharks are not only able to maintain a higher body temperature but also thermoregulate depending on the ambient conditions. An understanding of their thermal physiology will be acquired by investigating habitat use and movement patterns in relation to the natural environment in Mossel Bay, South Africa.

A new prototype acoustic transmitter was designed and manufactured to monitor the muscle temperature of the tagged sharks. At the same time, a tag (with temperature and depth sensors) was externally attached and a transmitter (with a temperature sensor) was fed (in a piece of bait) to each of the studied sharks. This provided, for the first time, a means to assess the complete thermal profile of a white shark from ambient water temperature through the warm white muscle to the warmest body core temperature of the stomach.

To date four white sharks (TL= 2, 2.5, 3, 3.5 m) have been triple tagged with an accumulative tracking effort of 520 hours. This included a continuous tracking session of 106 hours on one individual. The intensive tracking protocol has produced large amount of data (>5 million rows) that were filtered using a protocol created in R. In order to examine white shark habitat use patterns in Mossel Bay a three dimensional map of the bay was created. The movement patterns of tagged sharks were described in three dimensions by combining data from positional fixes (X and Y co-ordinates) taken with a GPS on board the tracking boat and the swimming depth sensor (Z co-ordinate). Behavioural patterns (e.g. hunting, resting and moving) were described using three dimensional rates of movement. Tracking data suggests that white sharks make use visual, auditory (never reported before) and magnetic clues to identify man-made structures and seafloor characteristics to navigate. Four core areas (50% kernel) were identified in Mossel Bay.

Preliminary analysis of the physiological data indicates that white sharks are able to raise body core temperatures and white muscle temperatures by up to 10 °C and 7 °C above ambient water temperature, respectively. Stomach temperature seems to be independent of the environmental conditions and ontogenetically constant. In order to understand the effects of body size, additional tracking of small (<1.5 m) and large (>4 m) white sharks is scheduled for 2011 and 2012.

**Frikkie van der Vyver (MSc student) - Investigating area-disaggregated models for management of the chokka squid *Loligo reynaudii* resource.**

Supervisors: Prof. W Sauer (w.sauer@ru.ac.za); Ms J Glazer (JeanG@nda.agric.za)

Funders: SASMIA

Genetic and morphometric results from previous *L. reynaudii* population studies revealed a potentially complicated stock structure, with possible boundaries occurring somewhere between Tsitsikamma and the Western Agulhas Bank (Shaw et al. 2010, Stonier unpublished). Clearer evidence of isolated stocks may require a change in the current resource assessment given that it is an area-aggregated model. The study thus aims to incorporate data on separated stocks into the existing mathematical models to investigate management of the resource on an area-disaggregated basis. This may potentially allow for spatial management of the resource, perhaps separating the fisheries into distinct geographical zones.

Specific objectives are to:

- test for a genetic breakpoint among squid populations between Tsitsikamma and the Western Agulhas Bank;
- contribute to validation of catch and effort in the jig fishery; and
- evaluate the feasibility of assessment of the resource on an area-disaggregated basis.

Genetic material and morphological samples are being collected on the West and southern Cape coast onboard commercial jig and research trawl vessels. Molecular genetic typing of individuals will be conducted on an AB3010 DNA sequencer using a suite of *Loligo*-specific microsatellite DNA marker loci, including *L. reynaudii*-specific marker loci developed during a previous project (Naud & Shaw 2008). Tests for genetic separation of stocks will be conducted using both population frequency based ( $F_{ST}$ ) and individual-based (STRUCTURE) analysis. Initial hard and soft part morphological character data analysis will be done using a Principle Component Analysis. All PCA's will be performed using the Unscrambler (Version 9.8) statistics package. Discriminant function analysis and MDS plots will also be conducted using Primer Package. A base case using the existing model on an area-aggregated basis will be developed by using the best jig, trawl and survey input data determined through validation. Using this validated catch, CPUE, as well as new genetic and morphological information, the current model will be assessed on an area-disaggregated basis. Based on a number of alternative scenarios, different sets of possible boundaries for the jig and trawl fisheries will be created to separate stocks. Data from these various scenarios will be used in a Bayesian model to generate results for the different area-disaggregated scenarios. Results may then be analysed and a number of hypothesis developed to determine which scenario yields the best plausible alternative to the current area-aggregated assessment of the resource.

**Emmanuel Mbaru (MSc student) - An assessment of the Kenyan coastal fishery between 2001 and 2009, with an evaluation of angler perceptions towards the development of an offshore FAD fishery**

Supervisor/s: Dr PD Cowley (p.cowley@saiab.ac.za); Dr L Dagorn (laurent.dagorn@ird.fr); Dr M Soria (marc.soria@ird.fr)

Funder/s: South West Indian Ocean Fisheries Project (SWIOFP)

Due to the overexploitation and degradation of near shore coastal areas, Fish Aggregating Devices (FADs) were introduced in the early 1980s to many parts of the Western Indian Ocean as ‘miracle solutions’ for making populations of large pelagic fishes available to small scale fisheries. Prior to the development of an offshore FAD fishery in Kenya, a retrospective analysis of the marine fishery in terms of catch, effort and participation was performed from 2001 to 2009. More recently, a socio-economic assessment of fishers and non-fishers as well as attitudes and perceptions towards the development of a FAD fishery was conducted.

Data on fishing gear, catch composition, CPUE and total catch were collected from 22 213 angler outings at four prominent landing sites. The fishery was characterized as being artisanal with low capital investment. Over 90% of fishing vessels were non-motorized with dugout canoes (dhows) being the dominant vessel used (77%). Dominant gear types included basket traps, baited hooks (both handlines and longlines) followed by nets (gillnets, trap nets, seine nets) and spear guns. Although 57 species belonging to 19 families were recorded, the catch was dominated by four species. CPUE values were low and rarely exceeded 1.5 kilograms per angler per day. Overall, beach seines and gillnets yielded higher CPUE values, while anglers operating motorized boats landed the highest mean catches.

During the socio-economic survey, 55% of all the respondents confirmed their willingness to engage in offshore FAD fishing activities and most (about 85%) were familiar with FAD associated species. To some extent, typical FAD fishing techniques were practiced in all the areas surveyed, particularly at Watamu. Interviewees from many of the sites displayed similar and low resilience values between 0.21 and 0.23, particularly Kilifi and Malindi. There was considerable variation in the expenditures of respondents, and as expected was highest in communities proximate to urban areas.

This study represents the first assessment of marine fisheries in Kenya and although it is unknown to what extent the introduction of FADs will change exploitation strategies, the baseline data collected will assist with the management of fishery resources.

**Alexander Winkler (MSc student): Report not available**

**Michelle Soekoe (Phd student) – Report not available**

**Fabien Forget (Msc student) – Report not available**

**JD Filmalter (MSc student) – Report not available**

**Georgina Robinson (PhD student) – Report not available**

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