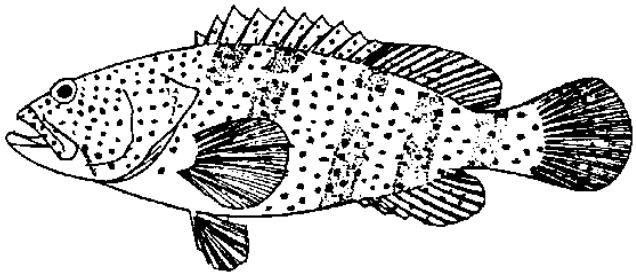
ACIAR Project FIS/97/73

Improved hatchery and grow-out technology for grouper aquaculture in the Asia-Pacific region

Annual Report: July 1999 – June 2000



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Executive Summary

Purpose and context of the project

Aquaculture of high value finfish species, such as groupers, is an industry of increasing importance throughout the Asia-Pacific region, including Australia. The development of large and affluent markets for live reef fish, particularly in Hong Kong and southern China, has increased pressure on wildstock resources. In many areas the demand for live reef fish, and the profitability of this trade, has encouraged overfishing and the use of destructive fishing practices, such as the use of sodium cyanide to 'stun' reef fish for capture by divers. Aquaculture of high value reef fish species can potentially supply product to the live reef fish markets, as well as other regional and domestic markets. The development of aquaculture technology for these species will not only support an economically beneficial aquaculture sector, but will also contribute to reducing pressure on wild stocks. Currently, the major bottlenecks to increased aquaculture production of groupers are the generally poor, and highly variable, survival in larviculture, and the limited sources of trash fish for grow-out. The ACIAR project addresess these issues by collaborating with research and development organisations in Indonesia and the Philippines to carry out priority grouper research to improve larviculture and to develop cost-effective grow-out diets of low fish content. An additional objective of the project is to support, through the Network of Aquaculture Centres in Asia-Pacific (NACA), more effective dissemination of research results arising from the project activities, and to promote greater collaboration and information exchange among centres in Asia involved in grouper aquaculture research and development. This objective is being addressed through an interactive grouper web page and an electronic newsletter for dissemination of information

Names of collaborating researchers and institutions

- Dr Mike Rimmer, Department of Primary Industries, Agency for Food and Fibre Sciences – Fisheries and Aquaculture, Northern Fisheries Centre, Cairns, Queensland, Australia
- Dr Kevin Williams, CSIRO Division of Marine Research, Cleveland, Queensland, Australia
- Mr Joebert Toledo, South-east Asian Fisheries Development Centre, Aquaculture Department, Iloilo, the Philippines
- Dr Ketut Sugama, Research Station for Coastal Fisheries, Gondol, Bali, Indonesia
- Dr Taufik Ahmad, Research Institute for Coastal Fisheries, Maros, Sulawesi, Indonesia
- Dr Michael Phillips, Network of Aquaculture Centres in Asia-Pacific, Bangkok, Thailand

Results / expected results

Larval rearing

Research on pre-feeding larvae at SEAFDEC has demonstrated that survival of *E. coioides* eggs and larvae is improved by incubating them at relatively low densities (200–400 per litre) and with low levels of aeration (100 ml / min). These results, combined with the results of trials with other environmental variables, provide optimal incubation conditions for grouper larvae which improve larval survival.

Larval rearing trials at GRSCF with *Cromileptes altivelis* using a combination of SS- and S-strain rotifer, brine shrimp, and artificial larval diets, gave an average survival of 29.4% (range 23.6–53.7%) following the exclusion of VNN-positive broodstock. *C. altivelis* larvae reached 29.2 mm ± 0.21 mm after 50 days.

Grow-out diet development

Research at RICF Maros has provided a wealth of information on potential ingredients for grouper grow-out feeds, and detailed chemical analyses of these ingredients.

The apparent digestibility of key feed ingredients has been determined with *E. coioides* in a series of experiments at SEAFDEC. The data are being used to develop practical diets for *E. coioides*. Promising results have been found using Australian meat and bone meal, Protamino Aqua (processed meat solubles), white cowpea or ipil-ipil meals as partial replacements of fishmeal. Replacement of up to 80% of the fish-meal with Protamino Aqua was found to have no adverse effect on growth and survival of *E. coioides*. At Gondol, the growth rate of *C. altivelis* was found to decline when soybean meal was isonitrogenously substituted for fishmeal at rates of 20% or more.

The dietary protein requirement of *C. altivelis* was found to decrease from ~54 to 45% as fish size increased from initial weights of ~5 and 17 g in studies at Gondol and Maros, respectively.

Asia-Pacific Grouper Network

Membership of, and interest in, the Asia Pacific Grouper Network continues to grow. Two regional workshops have been held in conjunction with the APEC Collaborative Grouper R&D Network project, at which the results of the ACIAR grouper project have been detailed.

The electronic grouper newsletter, developed to facilitate information exchange within the network, has been extremely popular and now has over 160 subscribers. A web site for the ACIAR Grouper Project has been added to the NACA Grouper web site.

Likely direction of future research

Larval rearing

Future work will continue to investigate the digestive physiology of grouper larvae, including development of the digestive tract and ontogeny of enzymes. Larval rearing methods will continue to be refined to improve larval survival and growth. The impacts of these improvements will be evaluated using the economic models developed for this project.

Ketut Suwirya (RSCF Gondol) will apply for a John Allwright Fellowships Program for Agriculture Research scholarship to undertake a PhD degree at James Cook University. Mr Suwirya's PhD research will involve the development of artificial feeds to replace live feeds for larval rearing of marine finfish, particularly groupers.

Grow-out diet development

The recent demonstration at SEAFDEC of the suitability of high quality meat meal as a total replacement of fishmeal in diets for *E. coioides* under laboratory aquarium conditions at Tigbauan is to be validated under sea cage conditions at Igang Station. This will involve a 4-month experiment in which fishmeal will be partially (50%) or completely replaced by Australian-sourced meat meal in pelleted diets and compared with a control treatment of feeding trash fish. If the results of this sea cage experiment confirm the earlier laboratory findings, consideration will be given to carrying out on-farm demonstration studies. These studies would be useful both to show the potential of pelleted diets to be used as an alternative to trash fish for grouper grow-out and also to generate reliable economic and production data for subsequent economic modeling of the alternative feeding systems (trash fish versus pelleted feed). Continued work will be done in the laboratory to determine the apparent digestibility of other potential feed ingredients and to examine these as partial or complete replacements for fishmeal.

At RICF Maros, further digestibility studies are planned to be carried out throughout the latter part of 2000 at the Barru floating cage site. This work will entail the determination of apparent digestibility of 6 to 7 local ingredients using *C. altivelis* as the target grouper species. Other planned studies involve a number of growth assays with *C. altivelis* to examine the interactive effects of varying dietary concentrations of protein and energy (lipid) and to assess the suitability of local ingredients as partial replacements of fishmeal in pelleted diets and in comparison with the feeding of trash fish.

To enable grow-out diet development studies to commence in Australia this calendar year and as a contingency against grouper fingerlings not being available from NFC because of protracted broodstock spawning problems, a shipment of *C. altivelis* fry is being arranged to be brought from RSCF Gondol and held under quarantine at CSIRO Cleveland. The shipment of about 600 fish is being planned for early October 2000. These fish will be used in comparative slaughter growth assay experiments to measure nutrient retention and growth productivity responses to the feeding of nutrient dense diets. The experiments will entail a factorial arrangement of various dietary concentrations of protein and energy (lipid) and compared against a commercially-manufactured high-energy barramundi diet. If *E. fuscoguttatus* fry become available from NFC, similar comparative slaughter growth assay experiments will be carried out with this species. This will enable the nutritional requirements of *E. fuscoguttatus* to be compared to those of *C. altivelis*. Hopefully, nutritional responses of both of these grouper species will be similar, and also not that dissimilar to barramundi. If this supposition is found to be correct, grow-out diet development of groupers can be fast-tracked, both in Australia and elsewhere, by reference to the much more extensive nutritional knowledge known for barramundi.

Asia-Pacific Grouper Network

The activities of the Asia-Pacific Grouper Network will be continued, particularly in conjunction with the APEC Collaborative Grouper R&D Network project. Regional workshops will continue to be held at regular intervals, and this series will incorporate the ACIAR end-of-project workshop. NACA will continue to coordinate the overall grouper R&D program, based on the outline developed in this project.

The Electronic Grouper Newsletter will be continued, since this is an increasingly popular mechanism for information dissemination. The ACIAR project web site and the NACA grouper web site will be expanded.

APEC has committed to support additional small research topics of relevance to the ACIAR project, including the development of the grouper virus research project, and additional work aimed improving research collaboration and extending the results to farmers and project seeking to improve coastal livelihoods through aquaculture.

Key to abbreviations and acronyms

AAHRI	Aquatic Animal Health Research Institute (Bangkok, Thailand)
AIAT	Assessment Institute for Agricultural Technology
ACIAR	Australian Centre for International Agricultural Research
AFFA	Agriculture, Forestry and Fisheries Australia
AFFS – F&A	Agency for Food and Fibre Sciences – Fisheries and Aquaculture (DPI)
AIMS	Australian Institute for Marine Science
APD	apparent protein digestibility
APEC	Asia-Pacific Economic Cooperation
APGN	Asia-Pacific Grouper Network
ARA	arachidonic acid (20:4n-6)
ARC	Australian Research Council
AusAID	Australian Agency for International Development
BOBP	Bay of Bengal Program
CARD	Capacity-Building for Agriculture and Rural Development
CRD	completely randomised design
CRIFI	Central Research Institute for Fisheries, Indonesia
CSIRO	Commonwealth Scientific and Industrial Research Organisation
DFID	Department for International Development (United Kingdom)
DHA	docosahexaenoic acid (22:6n-3)
DPI	Department of Primary Industries (Queensland)
EPA	eicosapentaenoic acid (20:5n-3)
FAO	Food and Agriculture Organisation of the United Nations
FWG	Fisheries Working Group (APEC)
GC	gas chromatograph
HUFA	highly unsaturated fatty acids
JCU	James Cook University of North Queensland
NACA	Network of Aquaculture Centres in Asia-Pacific
NFC	Northern Fisheries Centre (Cairns, Queensland, Australia)
PSRC	Port Stephens Research Centre (NSW Fisheries)
PUFA	polyunsaturated fatty acids
R&D	research and development
RICF	Research Institute for Coastal Fisheries (Maros, Sulawesi, Indonesia)
RSCF	Research Station for Coastal Fisheries (Gondol, Bali, Indonesia)
S- / SS-	small / super-small strain rotifer
SEAFDEC AQD	South-east Asian Fisheries Development Centre, Aquaculture Department (Tigbauan, Philippines)
TNC	The Nature Conservancy
TVP	Technology Verification Program (SEAFDEC)
UoF	University of Fisheries (Nha Trang, Vietnam)

Progress of Research Work

Project Objectives

The overall objective of the ACIAR project is to increase grouper production in the Asia-Pacific area by developing improved hatchery and grow-out technology. The project has three major components:

1. Larval rearing of groupers

The objective of this component of the research is to improve growth and survival of groupers during the hatchery phase.

The research is concentrating on developing a better understanding of the capacity of grouper larvae to digest various live prey organisms, and the nutritional composition that must be provided by live prey. This information is being used to assess the suitability of different live prey organisms at different stages of the larval rearing process, and to develop improved nutritional profiles for live prey organisms. Direct enhancement of larval nutrition, using artificial diets, is also being examined. These results will be integrated with other studies on environmental factors affecting grouper larvae to develop an improved methodology for larval rearing of groupers.

2. Diet development for on-growing of grouper

The objective of this component is to develop compounded feeds for grouper grow-out that have low environmental impact, have a low content of fishery resource, and are as cost-effective for the on-growing of grouper as the alternative of using trash fish.

This is being addressed in a structured way, acquiring nutritional information on feeds available for diet manufacture, characterising the requirements of groupers for key nutrients and demonstrating the cost effectiveness of the compounded feeds. The research plan recognises that grow-out nutrition work in Australia can only be done subsequent to the successful larval rearing of the fry but this constraint does not apply for the overseas collaborators where collection of fry from the wild is permitted.

3. Support for the Grouper Aquaculture Research and Development Program

The objective of this component is to 'value add' existing grouper aquaculture R&D efforts in the Asia-Pacific region by improving communication and promoting collaborative research between regional laboratories and agencies.

NACA, in cooperation with participating institutions, has prepared a cooperative grouper aquaculture research and development program based on the recommendations and specific research detailed in the proceedings of the Grouper Aquaculture Workshop held in Bangkok in April 1998, and more recent workshops held in Hat Yai (Thailand) and Medan (Indonesia). The program will be circulated to respective institutions to seek institutional support and commitment. NACA, in cooperation with participating institutions, will continue to seek funding support for specific projects under the Grouper Aquaculture Research and Development Program, with particular emphasis on the development of collaborative research and development projects.

NACA is facilitating enhanced communication amongst grouper aquaculture researchers by pursuing reports of research findings from participating institutions, and compiling and publishing this information in regional aquaculture magazines, and on the NACA grouper web site.

Research

Adherence to timetable / staff engaged

The commencement of the project was delayed in regard to the signing of project documentation with overseas agencies. Consequently, there were some minor delays in commencing some project activities. This, plus the prolonged DPI recruitment procedure, caused a delay in recruiting the ACIAR project biologist to NFC.

Two additional staff have been employed under the ACIAR project at Northern Fisheries Centre, Cairns, and NACA, Bangkok.

Dr Shannon McBride has been employed by the Queensland Department of Primary Industries at the Northern Fisheries Centre since November 1999 to assist with research under the ACIAR project. Her duties include the development of improved larviculture techniques for grouper, and a detailed assessment of the ontogeny of the digestive enzymes in grouper larvae. This work is being carried out in close collaboration with DPI-funded researchers at NFC. A copy of the recruitment advertisement for this position is appended (Appendix 1).

Mr Sih-Yang Sim has been employed (full-time, but part-time funded by the ACIAR project) at the Network of Aquaculture Centres in Asia-Pacific secretariat offices in Bangkok. Mr Sim's duties involve coordination of the Asia-Pacific Grouper Network, including compilation and distribution of the electronic newsletter, provision of material for inclusion in the 'Grouper News' section of regional aquaculture magazines, and maintenance of membership lists of network participants.

Methodology and Principal Experiments / Analyses

1 Project administration

1.1 Project meetings

The initial project meeting was held at the Research Station for Coastal Fisheries, Gondol, Bali, Indonesia, from 12–14 July 1999. The project meeting was attended by representatives from all participating research institutions, and also by Mr Ronald Rakiman, ACIAR Country Manager, Indonesia. A summary of the meeting discussion is included in Mike Rimmer's trip report for this project trip, which is summarised in Appendix 7.

The next project meeting will be held in Cairns, Queensland, Australia on 24–25 July 2000.

1.2 Training

There has been considerable discussion of training needs during the project visits and at the initial project meeting at Gondol. The ACIAR project in its original version had only one training exercise listed: one person from each laboratory was scheduled to visit Australia for training in analyses of small samples for fatty acids, lipid classes and vitamins. This training has been carried out only with staff from Gondol: Mr

Ketut Suwirya visited CSIRO's Division of Marine Research Laboratories at Cleveland, Queensland, from 4–24 June 2000 and trained with Dr Kevin Williams and Ms Maggie Barclay on analysis techniques for fatty acids, utilising *Artemia* samples from the larviculture research component of the project at NFC. Mr Suwirya also visited DPI's Bribie Island Aquaculture Research Centre and a *Penaeus japonicus* farm on the Logan River during his stay at Cleveland.

Training for staff at SEAFDEC has been postponed until later in the project. It was decided that SEAFDEC staff have adequate training in the chemical analysis techniques relevant to the project, and that the training budget would more usefully be utilised later in the project. This is now likely to involve cross-training in enzyme analysis techniques, with Gerry Quinitio visiting NFC Cairns to work with Shannon McBride on fluormetric analysis techniques, and Shannon visiting SEAFDEC AQD to work with Gerry and Perla Eusebio on spectrophotometric techniques.

Following discussion with staff at RICF Maros, it was decided that the nutrition team's training needs would be best met by providing training in assessing the digestibility of feed ingredients, and that this could be met by Dr Geoff Allan's team at New South Wales Fisheries Port Stephens Research Centre. This training will now be carried out in August-September 2000, when Dr Allan is running digestibility trials as part of on-going research projects.

RICF Maros also proposed some additional training, which can be carried out in Indonesia or at other laboratories in the region. Reni Yulianingshih will attend Gajah Mada University (Yogyakarta, Indonesia) for training in chemical analysis techniques for nutrition studies. Taufik Ahmad has requested that Asda Laining be trained in nutrition research, preferably in Australia.

The agreed training activities are summarised below.

Centre	**Aspects**	**Staff**	**Place**
SEAFDEC	Digestive enzyme assessment	Gerry Quinitio Shannon McBride	NFC Cairns SEAFDEC AQD
RICF Maros	Digestibility of feed ingredients	Rachmansyah	PSRC, Port Stephens [Aug-Sep 2000]
	Chemical analyses	Reni Yulianingshih	Gajah Mada University, Yogyakarta [Sept 2000]
	Grow-out nutrition / chemical analysis	Asda Laining	(to be identified)
RSCF Gondol	Nutritional analyses	Ketut Suwirja	CSIRO, Cleveland [*completed*]

1.3 Calibration exercise

There was good agreement between most laboratories for routine proximate analyses. Exceptions were for ash analyses where values for squid meal were highly variable between laboratories. Laboratories 9904 and 9905 tended to be give higher ash values for fish, squid and soybean meals than the other laboratories. In the case of laboratory 9904, this could be attributed to the low incinerating temperature used (450 vs 550 ^{o}C) which was the standard protocol in this laboratory for ashing live feeds samples. There was no obvious reason for the apparent variable ash results for laboratory 9905.

Only three laboratories reported gross energy analyses and one of these (9904) derived these empirically using energy conversion factors based on determined lipid, protein and carbohydrate contents. The other two laboratories used bomb calorimetry and recorded very similar results other than for squid and krill meals.

All laboratories used a chloroform:methanol extraction procedure for total lipid analyses. Good agreement was found between laboratories and across all feed samples.

Phospholipid analysis was carried out by four laboratories. Results were highly variable, both between laboratories and between feed samples. For example, three laboratories recorded only trace amounts of phospholipid in milk powder while the other laboratory found an exceedingly high concentration. Clearly, if phospholipid analyses are needed for the project, analytical procedures for phospholipid analysis will need to be critically reviewed and standardised procedures adopted by the laboratories.

Three laboratories reported fatty acid analyses. Agreement between the three laboratories was excellent across all feed samples.

The results of the calibration exercise have been reported to participating laboratories and is appended (Appendix 2). Dr Williams has discussed the findings of the calibration exercise with the respective laboratories at SEAFDEC, Gondol and Maros during recent visits.

1.4 End-of-project workshop

Date and location to be determined.

2 Larval rearing

2.1 Pre-feeding larvae / environmental factors

This component of the research has focused on determining optimal conditions for grouper larvae during the incubation and early larval rearing stages. Research at SEAFDEC has demonstrated that survival of *E. coioides* eggs and larvae is improved by incubating them at relatively low densities (200–400 per litre) and with low levels of aeration (100 ml / min). Embryonic survival is highest at salinities of 32 to 40 ppt; whereas with hatched larvae survival was highest at 16 ppt. These results were validated for practical hatchery use by examining the feeding frequency of grouper larvae under identical experimental conditions. A more detailed description of these experiments is appended (Appendix 3). Image analyses of the different morphometric traits of the grouper eggs and larvae associated with the different treatments are still being conducted.

These experiments have been carried out in static hatching tanks. Similar experiments will be repeated in flow-through tanks at NFC later in the project using *E. fuscoguttatus* eggs and larvae.

At NFC, the first spawning of the flowery cod *Epinephelus fuscoguttatus* occurred in September/October 1999. Fertilisation rates were high (94 – 98%) with a mean oocyte diameter of 936 μm. Fertilised eggs were stocked into 300, 500 and 900-litre larval rearing tanks (clearwater, recirculation system) at densities of 400 or 300 larvae/litre. Some rotifers but few copepods were available to support a larval rearing

run. Consequently, larval survival was poor. By day 6 post-hatching there was close to 100% mortality. Larvae were attracted to the water surface and a large number of mortalities between days 4 and 6 were due to larvae becoming caught at the water surface. On day 3, a small number of larvae were transferred into two 75ml pots with high densities of either copepod nauplii or rotifers. After two hours one larva had ingested rotifers and all larvae had ingested copepod nauplii. More thorough assessment of prey preference of this species will be undertaken this season with emphasis on eliminating mortalities due to water surface tension.

At Gondol, research has primarily concentrated on the problem of early larval mortality due to death by surface tension. *Cromileptes altivelis* larvae at 0–5 days after hatching are relatively weak swimmers and move slowly. They are easily trapped at the water surface by surface tension. When larvae receive a stress like the trapping, they secrete sticky mucus that will accelerate the trapping of other larvae. Finally, a significant number of larvae die in a short time.

The addition of oil to the water surface of larval rearing tanks improved the survival rate of *C. altivelis* larvae. The survival rate of larvae in tanks without oil was significantly lower ($P<0.05$) than those treatments to which oil was added. The highest survival rate was achieved with oil added at 0.3 ml/m^2. A separate experiment demonstrated that addition of oil till day-9 post hatching gave the best survival. This result correlates with the morphological development of the larvae: from day-9 the larvae begin to develop elongated dorsal and pelvic fin spines. At this stage the larvae become more active and floating death decreases. Full details of these experiments are appended (Appendix 3).

2.2 Larval nutrition

2.2.1 Nutritional composition of live feeds

This component of the research aims to improve larval survival by providing live prey of better nutritional value for larval rearing. In particular, fatty acid and vitamin composition of live prey organisms will be examined.

DPI and CSIRO have completed an experiment to develop nutritional enhancement procedures for *Artemia* to modify their HUFA composition. *Artemia* were supplemented with AlgaMac 2000, AlgaMac 3010, DC DHA Selco and A1 Super Selco for 6, 12 and 24 hours. Frozen triplicate samples were stored at -80^{o}C. Samples were analysed by CSIRO, in conjunction with Ketut Suwirya's training visit to the Division of Marine Research Laboratories at Cleveland, Queensland.

This experiment demonstrated that to produce *Artemia* with a reproducible, predictable high level of HUFA, rich in DHA and a high DHA:EPA ratio (2.2:1) then the enrichment diet Algamac 3010 is the best of those tested. The enrichment diet DC DHA Selco produced *Artemia* with a predictable high level of HUFA and a high DHA level but with a DHA:EPA ratio approximately half that of the Algamac diet (1.2:1). Full results of this work are appended (Appendix 3).

At SEAFDEC, the microalgae *Tetraselmis tetraheli, Nannochloropsis oculata, Isochrysis galbana, Chaetoceros calcitrans,* and *Nannochlorum* spp. were mass-produced in 1-ton tanks, concentrated and were analyzed for total lipid. The rotifer *Brachionus* was cultured in 1-tonne tanks fed with *Tetraselmis tetraheli, Chaetoceros*

calcitrans or *Nannochlorum* sp. for four days, then collected for lipid analysis. *Brachionus* from SEAFDEC's Fish Hatchery fed with *Nannochlorum* sp., baker's yeast for 24 hours, or enriched with DHA Protein Selco for 12 hours were also collected for lipid analysis.

Newly hatched *Artemia* sp. nauplii (Sea Dragon and San Francisco Bay brands) were analysed for total lipid content. Enrichment of *Artemia* nauplii with n-3 HUFA using commercial products such as DHA Selco, Algamac products, and AquaGrow products at different dosages and duration, and the decline in nutritional value after enrichment will be done once SEAFDEC's new GC is operational.

Future work will include studies on chain transfer of neutral and polar lipid class composition and fatty acids from eggs through first feeding and on their patterns of conservation and loss during starvation and feeding at different larval stages. Thereafter, *Brachionus plicatilis, Artemia* sp., nauplii and copepodites of *Acartia* and *Pseudodiaptomus*, and neonates of *Diaphanosoma celebensis* fed on various microalgae alone (eg. *Nannochlorum* sp., *Tetraselmis* sp., *Nannochloropsis* sp., *Isochrysis* sp., *Nitzchia* sp., *Chaetoceros* sp., *Thalassiosira* sp.) or supplemented with enrichment media (eg. Selco products, Algamac 2000/3010, Frippak booster/Rotiboost, omega-3 oil) will be analysed for lipid class and fatty acid composition. Information on the PUFA composition of natural food organisms will make it possible to choose a blend of food organisms to select a wide range of dietary levels and ratios of DHA, EPA and ARA. Series of feeding experiments will then be conducted to examine the effect of natural diets containing different lipid class and fatty acid composition with special attention to the influence of DHA, EPA and ARA levels and ratios on grouper larvae.

2.2.2 Nutritional requirements of grouper larvae

This component has only recently commenced. To determine the patterns of conservation and loss of neutral and polar lipid class composition and fatty acid in grouper larvae, samples of eggs and larvae at different developmental stages are being collected for lipid analysis. Larval and oil globule depletion will be measured using an image analyzer.

At SEAFDEC, fatty acid methyl esters of collected samples from *E. coioides* will be prepared once the new gas chromatograph (Shimadzu GC-17AAFW) has become fully operational. At NFC, comparative samples will be collected for *E. fuscoguttatus* when this species begins spawning later this year, and analysed by CSIRO.

2.2.3 Natural and artificial diets

Much of this work is ongoing, and is integrated with the larval rearing research. RSCF Gondol in particular has had good success in rearing larvae of *C. altivelis* using commercial larval artificial diets in conjunction with live prey.

We intend to expand this work through post-graduate study by Ketut Suwirya (RSCF Gondol) who will apply for a John Allwright Fellowships Program for Agriculture Research scholarship to undertake a PhD degree at James Cook University. Mr Suwirya's PhD research will involve the development of artificial feeds to replace live feeds for larval rearing of marine finfish, particularly groupers. Discussions have been held with Dr Peter Appleford and Dr Paul Southgate (Aquaculture Department, James Cook University) regarding potential collaborative projects, and the proposed research will be further discussed at the next project meeting in Cairns.

2.3 Development of the digestive tract and enzymes

This component of the research aims to add substantially to our knowledge of the ability of fish larvae to utilise various prey types. It complements earlier work on the physical constraints (in particular, small mouth size) of grouper larvae at first feed which limit their ability to ingest many prey types.

Larvae of grouper *E. coioides* will be reared at SEAFDEC using semi-intensive culture methods. Larval samples will be collected at days 0, 2, 4, 6, 8, 10, 12, 14, 16, 20, 25, 30 35, 40, 45, and 60 (day of hatching = day 0). Samples will be processed for histomorphology, histochemistry, and digestive enzyme assay. Comparative samples will be taken at NFC with *E. fuscoguttatus* and *C. altivelis*. Where possible, samples will be analysed by both laboratories to allow a comparison of results.

2.3.1 Histology

In the first part of the study, the development of the digestive tract and associated organs will be observed by light microscopy. The development of the digestive tract in early, transparent larval stages is presently being documented at SEAFDEC using an image analyser.

2.3.2 Digestive enzymes - qualitative

Different digestive enzymes will be localized. Image analysis system will be used to observe larval stages that are still transparent. The activity of the localized digestive enzymes will be measured in the second part of the study. Larval samples will be collected and processed for histomorphology and histochemistry as soon as alpha-napthyl dissodium phosphate is available.

2.3.3 Digestive enzymes - quantitative

Work to date in this component of the research has focussed on technique development. Research at SEAFDEC and at NFC is proceeding along parallel lines, using slightly different analysis techniques. While SEAFDEC researchers are using established photometric procedures, NFC researchers are developing fluorimetric analysis techniques to measure digestive enzyme levels in fish larvae. Details of initial technique development work done at NFC are appended (Appendix 3).

2.4 Verification – larval rearing

2.4.1 Intensive larval rearing

At SEAFDEC, newly hatched grouper (*E. coioides*) larvae are reared initially in 10-tonne circular tanks for 24 days following the protocol of Duray *et al*. (1997). Larvae will be fed rotifers (enriched or supplemented with artificial diet) and *Artemia* under green water culture system. Thyroxin will then be added to the rearing water at 0.01 ppm on Day 21 to 25 following the procedure of de Jesus *et al*. (1998) to accelerate metamorphosis. Larvae will be harvested on Day 26 and transferred to 20-tonne tanks and further reared for three weeks. They will be fed on-grown *Artemia* of increasing sizes and slowly weaned over to minced fish. Completely metamorphosed larvae will be further reared to about 50 mm in cages within the tanks for another three weeks.

To verify intensive seed production techniques for grouper, four batches of grouper eggs were directly stocked in 5-tonne circular larval rearing tanks. Percent viable larvae at hatching ranged from 30% to 86%. The initial stocking densities varied

from 2 to 36 larvae/litre. Live food was enriched with vitamin-C-fortified DHA Selco. Survival rates on Day 21 ranged from 11 to 56%. Day 21 larvae were treated with thyroxin either by immersion (0.001 ppm) or by bioencapsulation in *Artemia* (0.5 ppm) for 5 consecutive days. Average survival after thyroxin treatment was 53% by immersion and 59% by bioencapsulation. Larval survival from hatching to Day 35 ranged from 5.4 to 29.8%. Survival rate at harvest (Day 55; ≥ 50 mm size) was 3%.

At RSCF Gondol, the newly hatched *Cromileptes altivelis* larvae measured 1.51–1.55 mm. At the night of day 2, usually 2 days after hatching, the mouth and anus were formed. A pair of pectoral fins appeared almost simultaneously. Mouth width at the time of mouth opening was about 140-150µm. This suggest the use of SS rotifer (100-140µm lorica length) will enable the larvae to ingest most of the food given. The larvae consumed all the reserves (yolk egg) two days after mouth opening.

At day 7, the larvae reached 3 mm TL, at which stage they typically engulf air for their swim bladder inflation. Failure of adequate swim bladder inflation results in death or malformation of the vertebral column.

At day 10–11 the spines start to develop and by day 14 when the larvae reached 5–5.5 mm TL, premaxillar teeth and dentary teeth appeared on jaws. Digestive tract has developed and coiled. Gill rakers start to develop.

At day 21 around 7–7.4 mm TL, the long spines have reached their maximum size and gradually decreased in size thereafter. Metamorphosis typically commences at day 35 and is completed in most larvae by day 40 at which time they have reached of 23–25 mm TL.

The rearing tanks were stocked with approximately 5–10 larvae/litre. Briefly, SS rotifer were fed at day 3–7, S-rotifer day 7–30 kept at density 5–7 ind/L, *Artemia* day 21–40 at density 0.2–0.5/ml, microdiet ('Love Larvae') No2, (140–410µm) day 17–33, No 3 (350–580µm) day 30–40 and No 4 (480–800µm) day 35–42 and dry pellet made by RSCF Gondol from day 42 and thereafter. In 5 trials, the juveniles reached a mean total length of 29.2 mm ± 0.21 mm after 50 days of rearing with the survival rate ranging from 23.6 to 53.7 % with an average of 29.4 %.

2.4.2 Semi-intensive larval rearing

The objectives of this component are:

1 To develop culture techniques for copepods in earthen ponds and tanks.
2 To develop / verify semi-intensive seed production techniques for grouper in earthen ponds and tanks
3 Develop and compare fry to fingerling production in earthen ponds and tanks.
4 Examine economic viability of culture of copepods and of semi-intensive seed production in earthen ponds and tanks.

Various combinations of organic and inorganic fertilizers will be tested in earthen ponds and will be correlated with chlorophyll *a* and copepod production. Adults and stage 4 to 6 copepodids of *Acartia* and/or *Pseudodiaptomus* will be collected from brackishwater ponds, isolated, and scaled up to 1–2-tonne fiberglass tanks. Copepods will be fed daily mixed microalgae at a final density of 24,000 cells/ml or a combination of mixed algae and bread yeast (0.5 g/50,000 copepods). Following the best fertilization scheme, ponds will be prepared for zooplankton production. About a week after filling up the ponds, 1- or 2-day old grouper larvae will be stocked at 0.25,

0.50 or 1.0 million larvae/ha. To sustain copepod nauplii production in semi-intensive larval tanks, adults and copepodids of *Acartia* and/or *Pseudodiaptomus* will be added 3 days before stocking of larvae and every week thereafter until Day 17. Copepods mass-produced from ponds or tanks will be added into the larval tanks daily from Day 25 until harvest to minimize the use of *Artemia.* Food abundance, larval growth, and gut content of the larvae will be monitored every 3 days until harvest (completion of metamorphosis). Fry to fingerling production in concrete tanks or in net cages set in ponds will be developed using either fish bycatch or SEAFDEC formulated diet for carnivorous fish. Economic analysis to estimate production cost for copepods, grouper fry and fingerlings will be done.

Prior to semi-intensive seed production of grouper, various methods to mass-produce copepods in tanks and earthen ponds were tested. Six units of 1-tonne fibreglass tanks were initially stocked with *Acartia* spp. copepodids and adults at 100 ind/l. These were fed daily with microalgae at a final density of 24,000 cells/ml or a combination of half the ration of microalgae plus baker's yeast. The population of copepods in each tank was monitored daily for two weeks. In a parallel study, the efficacy of various combinations of organic and inorganic fertilizers in the mass production of copepods were tested in 17 units 200-m^2 earthen pond at Dumangas. In addition to the natural population of copepods in the incoming water, nine ponds were inoculated with *Acartia* copepodids and adults at a density of 10 ind/l. Physico-chemical parameters were monitored four times a week while samples to examine copepod population and *chlorphyll a* levels were taken twice a week. Collected samples are presently being analyzed. Construction of earthen ponds allotted for this study was completed in April 2000.

3 Grow-out nutrition

3.1 Inventory of feed ingredients

Site selection of the local ingredients producers area in South Sulawesi was based on the consideration on availability and potentialities for imported ingredients substitution. The raw materials were observed in abattoir, cold storage, soybean cake producers, CPO producers, cassava mill, and fish landing site.

The volume of trash fish in Luwu Regency is 9,691 MT/year, and the price in the season (Rp 1,500/kg) was not so much different with the price in off season (Rp 2,000/kg). Luwu Regency is the main trash fish producer followed by Takalar, Barru, and Selayar Regencies. Makassar, is the main producer of blood meal, the abattoir's produce more than 38.000 MT followed by the ones in Tana Toraja, Bulukumba and Bone Regencies. Makassar is also the main producer of shrimp head and palm oil waste (41,354 MT) after Luwu (125,000 MT) and Mamuju (100,000 MT). Palm oil waste is a good source of plant fat. Rice bran is found all over South Sulawesi, but the main producers are Luwu, Bone, Wajo, and Sidrap. The high quality rice bran is a good source of plant protein, fat and fibre. Cassava cake, a source of carbohydrate, is produced mainly in Gowa and Enrekang regencies.

Based on the regional availability of ingredients, Luwu Regency is the main producer of local ingredients for either fish or poultry feed. Unfortunately, Luwu Regency is the furthest Regency from the province capital city, Makassar. Full details of the feed ingredient survey are appended (Appendix 3).

3.2 Nutritional composition

3.2.1 Chemical analyses of feed ingredients in South Sulawesi

A variety of ingredients was collected from throughout southern Sulawesi and analysed for moisture, crude protein, crude fat, glucose, ash and fatty acid. Proximate analysis was carried out at RICF while fatty acid analyses were carried out at Gajah Mada University, Yogyakarta. Full details of the results are appended (Appendix 3).

Protein content of animal sources ranged from 19.5 to 83.0% and in plant sources from 5.3 to 10.6%. Blood meal contained the highest protein (83.0%) but the lowest fat (0.2%). The high protein content and availability (94,189 ton/year) makes blood meal an ingredient with good potential for fish meal substitution. The other material containing high protein is shrimp head meal (50.4%) but its ash content is relatively high: 25.1%. Of three species of fish analysed (anchovy, sardines, and pony fish), protein content of pony fish is the lowest: 55.6%.

Protein content of plant sources materials is very low. Despite the low content of protein, palm oil cake and rice bran are potential sources of fat with fat content of 11% and 12 % respectively. To increase the protein content of plant sources, a fermentation process is suggested.

3.2.2 Digestibility of key ingredients

At SEAFDEC, protein digestibility studies have been carried out with *E. coioides*. Apparent protein digestibility (APD) values for fish meals were generally high andcomparable with those of *Acetes* sp. and defatted soybean meal. No significant differences were observed between the APD of squid meal, white cowpea meal, *Acetes* sp. and defatted soybean meal, but APD for ipil-ipil leaf meal was significantly lower. The digestibility of rice bran protein by grouper juveniles was very poor. The growth performance of fish fed white fishmeal and white cowpea meal- based diets was comparable with that of the control fish. Fish fed ipil-ipil leaf meal-based diets had the poorest growth performance. White fish meal-based diet had the highest APD value, followed by control. The APD values for white cowpea meal and ipil-ipil leaf meal-based diets were comparable but significantly lower than that of the control diet. The present findings suggest that APD can be used as an indicator of protein quality. Also, white cowpea meal can be incorporated as protein source in practical diet for grouper at 20.5% of the diet without affecting their growth.

At RICF Maros, digestibility studies are being carried out with *C. altivelis* which are sourced from RSCF Gondol. Digestibility studies carried out in Year 1 examined the digestibility of shrimp head, soybean, and palm oil cake. A standard feed (46.1% protein, 11.2% fat, and 5.02 kcal/g) was used as a reference diet. The results showed that the APD of soybean, shrimp head, and palm oil cake was 65.1±3.26; 73.1±2.80 and 77.8, respectively.

3.3 Nutritional requirements

3.3.1 Protein; P:E

Research into the optimal protein requirement of humpback grouper *C. altivelis* has been conducted at RSCF Gondol and at RICF Maros. At RSCF Gondol, the experiment on dietary protein requirement was conducted in 15 polycarbonate tanks, each 30 litres in volume. All tanks were equipped with flow-through water system. Hatchery produced juveniles of 5.5 ± 0.2 g in body weight were randomly selected and 10 fish stocked in each tank. Fish fed experimental diets containing different levels of protein, i.e., 36, 42, 48, 54, and 60% for 50 days. The experiment was designed using CRD with three replicates for each treatment. Broken line analysis of body weight gain data showed that optimum dietary protein requirement for maximum growth of juvenile humpback grouper was 54.2 %. Details of this experimental work are appended (Appendix 3).

At RICF Maros, these experiments were carried out at the floating net cage site at Barru. Diets were formulated to provide crude protein contents ranging between 30 and 55% at 5% increments. Juvenile *C. altivelis* sourced from RSCF Gondol were selected on weight uniformity and assigned randomly into 18 of 1×1×1.2 m net cages at a stocking density of 15 fish/cage. The mean weight (± SD) of the fish was 17.3 ± 2.10 g. Fish were fed twice daily at 0800 and 1600 h.

Growth rate and food conversion ratio improved curvilinearly with increasing dietary protein content. Survival rate was not significantly different over the dietary range. Broken line analysis of the growth rate data detected a break point at a dietary protein content of 45.3%, suggesting that this level appears to be the optimal for maximising growth rate of *C. altivelis*. Full details of this experiment are appended (Appendix 3).

3.3.2 Fatty acids

Experimental work at Gondol has investigated the n-3 HUFA requirements of *C. altivelis*, with the objective of identifying the minimum dietary requirement to prevent n-3 HUFA deficiency. The results indicated that growth of *C. altivelis* was significantly affected by the level of n-3 HUFA in diets. Fish fed diet without n-3 HUFA supplementation had significantly lower growth than those fish fed diets with n-3 HUFA level of 1.0% – 3.0%. Growth of fish that were fed diets with levels of n-3 HUFA 1.0%, 1,5%, 2.0% and 3.0% were not significantly different ($P>0.05$). This experiment shows that the minimum dietary n-3 HUFA requirement for growth of humpback grouper juveniles is 1.0%. Details of this experimental work are appended (Appendix 3).

3.3.3 Phospholipids

This component has not yet commenced. Based on the results of the inter-laboratory calibration exercise, which demonstrated substantial differences between laboratories for phospholipid analyses, some additional cross-checking of phospholipid analyses will be necessary.

3.4 Fishmeal replacement

A series of eight experimental diets was tested at SEAFDEC to determine the effects of replacement of fish meal with meat solubles at proportions ranging from 0 to 100%. Generally, weight gain and SGR tended to decrease as the percent replacement of fish meal with processed meat solubles increased. The fish-meal-based control diet gave significantly better growth than diets with 60% to 100% replacement. Those fish given diets with 100% meat solubles, 0% fish-meal exhibited the lowest growth rate compared to the rest of the dietary treatments.

Survival ranged from 83–97% and did not differ significantly among treatments. Feed conversion efficiency was best at 30% fish-meal replacement and poorest at 80% fish meal replacement. Fish fed the diets containing the three highest fish meal replacement levels (60–100% meat solubles) exhibited significantly lower growth and inefficient feed conversion compared to the control group. These results indicate that while partial replacement of fish meal with meat solubles may be practical for grouper diets, total replacement may result in significant degradation of growth rate and feed conversion. Full details of this experimental work are appended (Appendix 3).

Research at RSCF Gondol with *C. altivelis* has demonstrated that there is a limited capacity to replace fish-meal with non-animal protein in this species. Five experimental diets containing different levels of soybean meal (0, 10, 20, 30 and 40%) and fish meal (63, 55.3, 47.6, 39.9 and 32.2%) were prepared as dry pellet. Diets were isonitrogenous (51% CP) and isocaloric (4.4 kcal/100 g diet). Polycarbonate tanks of 30-litre volume were each stocked with 11 fish (3.0 ± 0.02 g) , and the experimental diet fed three times daily to satiation for 35 days. Results of the experiment showed that fish fed diet containing 10% soybean meal had the highest weight gain and feed efficiency. Inclusion of 20% or more soybean meal in the diet significantly retarded the growth of fish.

3.5 Diet validation

This component has not yet commenced. Diet validation work will be undertaken later in the project when further laboratory studies of diet composition and nutrition have been completed.

3.6 Economic evaluation

Bill Johnston (DPI) has drafted separate economic models for the three major phases of grouper aquaculture:

1 Hatchery;
2 Nursery;
3 Grow-out (in ponds and cages).

The models are written in Excel 97® and provide a detailed assessment of the capital and operating costs for each phase of grouper aquaculture. Outputs include a breakdown of annual costs and a profitability analysis (using a discounted cash flow model).

These models will be provided to participating institutions at the project meeting in Cairns in late July. Bill will run through the models at that meeting to demonstrate the method of operation. The second stage of the economic evaluation will be for participating institutions to check that the components of the model are applicable to hatcheries, nurseries and grow-out farms in their country. In addition, we need to develop 'model farms' for Australia, Indonesia and the Philippines. These farm models will be used to evaluate the economic impact of our research on grouper aquaculture in each of the participating countries. More evolved versions of the economic models will be widely distributed through the Asia-Pacific Grouper Network.

4 Communication and coordination

The communication and coordination component of the project was developed from a recommendation of the Grouper Aquaculture Research Workshop held in Bangkok in 1998 that communication and coordination between grouper aquaculture researchers

in the Asia-Pacific region needed to be improved in order to increase the efficiency of the existing research effort in this field by reducing overlap and outright duplication of research effort. Because the communication and coordination functions fall within NACA's mandate, NACA is the central point for these activities.

4.1 Research program development

The overall research program for the Asia-Pacific Grouper Network was presented for discussion at the APEC–NACA Collaborative Grouper R&D Workshop held in Medan, Indonesia, on 17–20 April 2000. The research program was accepted, with some modifications, by the workshop participants. The research program outline is now:

1 Production technology
  1.1 Broodstock
  1.2 Larviculture
  1.3 Nursery
  1.4 Grow-out
  1.5 Post-harvest
2 Environment
3 Marketing
4 Food supply, certification
5 Socio-economics, livelihoods
6 Fish health

4.2 Research coordination

Coordination of the above program is being undertaken by NACA, in conjunction with ACIAR (Mr Barney Smith), AFFA (Ms Paula Shoulder and Mr Matthew Dadswell), SEAFDEC (Dr Clarissa Marte), DPI (Dr Mike Rimmer) and CSIRO (Dr Kevin Williams).

Additional funding to support the APGN has been provided by APEC through the Fisheries Working Group, and this supports annual workshops and staff exchanges. These mechanisms are being used to 'value-add' the ACIAR Grouper Project, and to expand the networking component of the project.

4.3 Dissemination of results

Results are disseminated through five mechanisms:

1. The Grouper Electronic Newsletter, compiled and sent by Sih-Yang Sim (NACA). Yang has sent out six editions of the newsletter to date and it currently has over 160 subscribers. The E-Newsletter provides updated information on grouper aquaculture in the region without the need to access the grouper website, which appears to be a major constraint for some parts of the region due to slow downloading. The 'E-Newsletter' was designed for people who are unable to connect to the internet, but who may benefit from the wide range of information available in the region. In fact, with every new issue of the Grouper E-Newsletter, several requests are received in NACA for documents to be sent via e-mail. Copies of the E-Newsletter are appended (Appendix 4).
2. The 'Grouper News' segment in regional magazines and newsletters, particularly 'Aquaculture Asia' and 'The Live Reef Fish Bulletin'. This has appeared regularly since July 1998.
3. The ACIAR Grouper Project web site (http://naca.fisheries.go.th/aciar/). This is linked to the NACA grouper web site (http://naca.fisheries.go.th/grouper/) and

contains the outline of the project and detailed project documentation. The site is maintained by Sih-Yang Sim and is updated regularly.
4. Publications – see Appendix 5 for details.
5. Presentations at regional conferences, workshops and meetings – see Appendix 6 for details.

Although the grouper network was initially targeted on the Asia-Pacific region, the network has also attracted attention from the African region (Tanzania, Mozambique), and more recently the United States of America.

Recent publications and reports prepared by NACA and associated grouper network participants:

1. Identification Guide to Fishes in the Live Seafood Trade of the Asia-Pacific Region.
2. Research and Development: The Seed Production Technique of Humpback Grouper, *Cromileptes altivelis* (prepared by Gondol, Bali).
3. Diagnosis and Treatments for Parasitic Diseases in Humpback Grouper, (*Cromileptes altivelis*) Broodstock (prepared by Gondol, Bali).
4. Proceedings of the ACIAR/NACA Grouper Aquaculture Research Workshop, Bangkok, Thailand 7-8 April 1998.
5. Draft Report for the Regional Workshop on Grouper Aquaculture, Hat Yai, Thailand, April 1999.
6. Draft Report for the Regional Workshop on Sustainable Seafarming and Grouper Aquaculture, Medan, Indonesia, 17-20th April 2000. Contributed papers also available on the web site (www.naca.fisheries.go.th/grouper).

Importance of results

Future research plans

Results to date have indicated no major change to the proposed research program.

Results achieved at Gondol indicate the application of high-quality larval feeds in grouper larviculture, and this is an area of research that shows considerable potential. We plan to address this area of research in more detail than originally proposed, through post-graduate study by Ketut Suwirya (GRSCF) at James Cook University. Mr Suwirya's PhD would be carried out under the supervision of Dr Paul Southgate and Dr Peter Appleford (Aquaculture Department) who are internationally acknowledged experts in the field of larval feed development and fish nutrition. Both the ACIAR project and JCU have strong linkages with Dr Sagiv Kolkovski (Fisheries Western Australia) who is also an acknowledged expert in the field of feed development for marine finfish larvae, and who would have some involvement in this research.

Future project budget

The research activities of the project are currently being managed within budget. At a later stage, some additional funds may be necessary to support an enhanced attendance at the end-of-project workshop (planned for mid-2002). This will particularly depend on the location and format of the workshop.

Conduct of other research projects

The ACIAR work is strongly linked with other projects in place at all the participating laboratories. A major closely-linked project is the APEC Collaborative Grouper R&D Network Project (FWG 01/99), which is administered by AFFA and coordinated by NACA. The objectives of the APEC project are to:

1. Through the development of a regional research network develop the capacity to establish a sustainable grouper aquaculture industry which will benefit all collaborating economies.
2. Provide an alternative source of income / employment to people currently engaging in dangerous and illegal fishing practices.
3. Protect endangered reefs and reef fish from the pressures of illegal and dangerous fishing practices.
4. Develop a new aquaculture industry with significant export potential and economic benefit to a diversity of stakeholders.
5. Reduce substantially the current reliance on wild-caught fingerlings for aquaculture purposes because capture of wild juveniles is probably unsustainable, and is sometimes carries out using destructive fishing techniques which can have significant impact on the long-term status of reef fish stocks.

Through APEC involvement, the expansion of an existing South-East Asian initiative on collaborative research into grouper culture can be extended to more economies in the Asia-Pacific region. The role of APEC will be to enhance the extension of grouper research and facilitate the development of a network throughout the APEC region and beyond, to ensure that all economies in the region can benefit from the development of improved technology in live reef fish culture techniques.

The APEC Collaborative Grouper R&D Network Project has held two regional workshops to date :

1. Hat Yai, Thailand, 7–9 April 1999

This workshop was attended by 43 delegates from 14 APEC and NACA member economies. Economies represented included Australia; Brunei-Darussalam; China; Chinese Taipei; Hong Kong, China; Indonesia; Japan; Korea; Malaysia; New Caledonia; Peru; Philippines; and Thailand.

The major outcomes from the workshop were:

- Agreement on the need to expand and strengthen the grouper aquaculture research and development network, particularly through technical exchanges.
- Development of a strategic research plan for to support grouper aquaculture development; improve survival and food safety of live fish during handling and transport; and address destructive fishing practices.
- Preparation of three projects for consideration by the APEC FWG.
- Submission of a proposal for APEC to work with other regional bodies to develop a cooperative and equitable means of addressing the issue of cyanide fishing.

2. Medan, Sumatra, Indonesia, 18–20 April 2000.

This meeting was held in conjunction with the Regional Seafarming Workshop and was hosted by the Government of Indonesia in cooperation with the Bay of Bengal

Programme (BOBP/FAO), and NACA. The workshop involved 53 participants from APEC economies from throughout the Asia-Pacific, including Australia; Hong Kong, China; Indonesia; Japan; Korea; Malaysia; Philippines; Singapore; Thailand; and Vietnam. The meeting was attended by representatives from NACA, the Secretariat for the Pacific Community (SPC), the Solomon Islands, Myanmar, INFOFISH, and a non-governmental organisation, The Nature Conservancy (TNC).

The meeting was very successful, with a number of key recommendations being made in support of APEC FWG and NACA objectives for grouper aquaculture. Specifically, the Workshop recommended further expansion of activities to cover coastal livelihoods, improved environmental management of cage aquaculture, and most importantly the formalisation of the participation of the centres/institutions involved in the network.

In addition to the Grouper Collaborative R&D Network project, APEC is supporting a number of associated projects:

- Regional survey of grouper fry collection methods (Hong Kong University).
- Production of a 'farmer-friendly' Grouper Health and Husbandry Manual (SEAFDEC AQD).
- Development of a regional disease research project, concentrating on viral diseases in groupers (AAHRI).

Additional projects are likely to be developed as a result of the Medan workshop. All these projects are being coordinated through NACA as part of the coordinated R&D program of the Asia-Pacific Grouper Network.

Related research grants received or applied for

PhD scholarship – Japan Society for the Promotion of Science

Joebert Toledo (SEAFDEC AQD) has been awarded a PhD scholarship from the Japan Society for the Promotion of Science at the Faculty of Applied Biological Science, Hiroshima University. Joebert's dissertation will be titled: 'Studies on the seed production of grouper *Epinephelus coioides*' and will cover a range of research activities at SEAFDEC, including some ACIAR-funded activities. The scholarship is for 4 years, from 2000 to 2004. Joebert is required to attend Hiroshima University for at least 30 days each year to undertake some laboratory work and to consult with his supervisor, but most of the research will be undertaken at SEAFDEC.

Capacity-Building for Agriculture and Rural Development (CARD) Program – CSIRO / NTUF Feeds Development Project

Under a newly established bilateral arrangement between Australia and Viet Nam, AusAID is supporting a project to build *Aquafeeds R&D capacity for intensive aquaculture in Viet Nam*. Participants in the Project are CSIRO Marine Research (Kevin Williams) and the University of Fisheries (UoF), Nha Trang, Viet Nam (Le Anh Tuan). The overall objective of the project is to increase food security and income of rural Vietnamese in coastal communities by improving the profitability, and environmental sustainability, of intensive aquaculture in Viet Nam. This objective will be achieved by:

- Developing a collaborative aquafeed research project between CSIRO and UoF to facilitate the transfer of aquafeeds technology between Australia and Viet Nam.

- Instituting a training program to increase the aquaculture skill base at UoF and to provide Vietnamese post-graduates with opportunities to study aquaculture nutrition.
- Disseminating and show-casing project achievements at scientific forums, by the holding of a technical workshop in Viet Nam and extension of research findings to the aquaculture industry.

In the CARD project, grouper and rock lobsters are priority species for which feeds and feeding management research will be carried out. There is an obvious advantage for the CARD project to be closely linked with the ACIAR Grouper project and this will benefit both projects. Wherever possible, key Vietnamese staff engaged in the CARD project will be invited to attend Project meetings of the ACIAR Grouper project.

FRDC Project – Development of a National R&D Plan for Hatchery Feeds

In 1999, FRDC commissioned a Hatchery Feeds R&D Plan, to provide guidance in the area of hatchery feeds R&D for the corporation in the period 2000–2005. The R&D plan was developed at a workshop held in Cairns, Queensland, on 9–10 March 2000. The objectives of the workshop were:

- To assess the status of hatchery feeds, including live and compounded feeds, and to identify research in progress.
- To assess priorities for research and development needs in the area of hatchery feeds.
- To identify constraints to the continued development of Australian aquaculture in the area of hatchery feeds.
- To identify opportunities to enhance collaboration and information exchange amongst researchers and industry.
- To develop a national R&D plan for hatchery feeds.

The aquaculture community was widely polled to establish industry priorities for future research. A questionnaire was sent to all stakeholders, together with an invitation to attend the workshop, which was held in Cairns on 9–10 March 2000. Researchers were invited to present the results of work in progress, and industry needs were canvassed in open forums.

For convenience, the subject was divided into 5 main areas of research: microalgae, rotifers, brine shrimp, copepods and formulated diets. Status reviews were commissioned in each of these areas, and priorities in each defined in the workshop. In all areas, the need to benchmark best practice and to more efficiently transfer research results to industry were highlighted. In addition to these common priority areas, the following specific areas were identified as worthy of further research:

- Microalgal production systems
- The role of microalgae in green-water systems
- Assessment and production of Australian rotifer strains and alternative feeds
- Production of brine shrimp in Australia rather than depending on imported product
- Early weaning of larvae on to formulated feeds
- Scaling up existing systems for copepod production
- Development of a knowledge-base for copepod production

- Improvement of diets for copepod production
- Identification of appropriate copepods as food for individual species
- Development of local microdiets.

Further details, and copies of the draft Hatchery Feeds R&D Plan and the workshop proceedings, are available from the web site: http://www.aims.gov.au/hatchery-feeds.

Many of these priority areas have direct linkages with the research being carried out in this ACIAR project. There are thus excellent opportunities to link the ACIAR research with FRDC projects in the field of hatchery feeds.

APEC Collaborative Grouper R&D Network Staff Exchange

A collaborative project staff exchange has been funded for Dr Inneke Rumengan (Laboratory of Marine Biotechnology, Faculty of Fisheries and Marine Science, Sam Ratulangi University) to visit Northern Fisheries Centre, Cairns, to undertake collaborative research to selectively breed SS-strain rotifer (*Brachionus rotundiformis*) for application in marine finfish hatcheries, particularly grouper hatcheries.

The project will investigate the development of selective breeding procedures for *B. rotundiformis* to determine whether it is practical to select for size as a genetic trait. If rotifers can be selectively bred for size, then a population of the desired size (say, <90µm width) could be established for routine use in hatcheries. This would preclude the need to sieve rotifers, reducing wastage of rotifer production and increasing hatchery productivity by making more rotifers available for larviculture using the same rotifer culture facilities.

CARD Program – DPI / RIA1 / RIA2 Developing marine finfish hatchery technology for mariculture in Vietnam

This activity will promote the development of sustainable aquaculture for marine finfish by providing training for Vietnamese personnel in barramundi / seabass (*Lates calcarifer*) hatchery technology, and in copepod culture technology. This proposal seeks to address the current major constraint to the development of sustainable finfish mariculture in Vietnam, which is the availability of seedstock (fish fingerlings). Training will be provided in the following areas:

- Broodstock management and spawning induction;
- Live prey production, including a specific component on the development of copepod production technology;
- Hatchery rearing of marine finfish, concentrating on barramundi / seabass.

This application was not successful in the first round of CARD applications, but will be submitted in a revised form for consideration in subsequent funding rounds.

Development of linkages with collaborating-country organisations

DPI and GRSCF have developed strong linkages with The Nature Conservancy, a US-based NGO, which is providing natural resource management services to the Government of Indonesia in Komodo National Park. Although most KNP inhabitants mainly derive their income from a pelagic lift net fishery targeting squid and small pelagic fish, several surrounding communities, are involved in fishing with cyanide

and hook-and-line for valuable fish species (groupers and Napoleon wrasse) to supply foreign markets (mainly Hong Kong) with live food fish. The extremely high exploitation pressure on the grouper stocks, and the poisoning of the coral reefs through the use of cyanide severely threaten the marine biodiversity in and around KNP.

To abate these threats, TNC's Indonesia Coastal and Marine Program together with the Indonesian Park authority implemented an extensive marine conservation program for Komodo National Park. Development of a fish culture enterprise that involves local communities was identified as a possibility to steer fishermen away from destructive and unsustainable fishing methods. Also, the development of a fish culture enterprise would serve as a model for other rural areas in Indonesia, thereby contributing to the market transformation of the life reef fish trade from unsustainable, capture-based to sustainable, culture-based.

In July 1999, an expert team consisting of Mr. Bill Rutledge (Director of Aquaculture for NSW Fisheries), Dr Mike Rimmer (DPI) and Dr Ketut Sugama (GRSCF) visited the Komodo area to collect data for the compilation of a business plan for the fish culture enterprise. The business plan concluded that grouper aquaculture in the Komodo region was potentially highly profitable. The business plan was reviewed by Dr Stephen Battaglene, previously with ICLARM and now a Senior Research Fellow with the Tasmanian Aquaculture and Fisheries Institute, and has been accepted by TNC. Presently, part of the funds to start up the hatchery project have been secured by TNC, and a suitable location for the hatchery has been identified. It is expected that the construction of the hatchery will start in July 2000. Application for funding to continue this collaboration to develop mariculture for the Komodo region has been made to APEC and to AusAID.

In addition, linkages have been formed with Sam Ratulangi University, Manado, northern Sulawesi, through the APEC Collaborative Grouper R&D Network staff exchange outlined above.

RICF Maros has developed enhanced linkages with Indonesian universities by involving undergraduate and postgraduate students in the research:

Student Contributions to RICF Activities

1. Makmur, M; undergraduate of University of Moeslim Indonesia (Makassar) – protein requirement of humpback grouper (first two of five chapters completed).

2. Syafrianto; undergraduate of Hasanuddin University (Makassar) – protein digestibility of local raw material on humpback grouper. The experiment just finished in June 2000

3. Hasmadi; undergraduate from Diponegoro University (Semarang, Central Java) – digestibility of blood meal and ensiled blood on humpback grouper. His research is still ongoing until August 2000.

4. Muchlis, M; Master degree of Hasanuddin University (Makassar) – protein/lipid ratio of diet on growth of humpback grouper. His study includes the digestibility of the diets.

5. Ridwan, M; Master degree of Hasanuddin University (Makassar) – replacement of fish meal on humpback grouper diet including the digestibility of replacer.

In addition, RICF Maros has hosted field trips and visits from local educational institutions:

- Student from Hasanuddin University (undergraduate and postgraduate) - discuss the sea farming in floating net cage particularly humpback grouper and milkfish.

- Student from Aquaculture Program of Polytechnic in Pangkep Regency - discuss the sea farming aspects included the site selection, design construction of raft and cage, nutrition and feeding management as well as diseases.

- TVRI Station of Makassar; hunting the location that potential for sea farming development particularly for export commodity.

- Mayor of Barru Regency, studying the model of sea farming of exportable fishery community.

Optimal methods / channels of extension / outreach of results to end-users

All the participating organisations in FIS/97/73 have well developed extension facilities to assist with the application of research results.

DPI has a range of effective extension activities in the field of aquaculture. Regular (currently six-monthly) workshops / conferences are held in association with the Australian Barramundi Farming Association which represents the tropical marine finfish farming sector in Australia. Technical outputs are provided to industry at these workshops / conferences. In addition, dedicated technical workshops on specific aspects of marine finfish culture are held on an 'as needed' basis. Other extension activities provided by QDPI are:

- publication three times per year of 'Queensland Aquaculture' Magazine, allowing widespread dissemination of research outcomes to the Queensland aquaculture industry;
- publication of technical material in a range of formats, such as the 'Going Farming' series and the Information Series publications.

DPI is holding a dedicated Reef Fish Aquaculture Symposium in conjunction with the ACIAR FIS/97/73 Project Meeting to be held in Cairns in late July 2000. The Reef Fish Aquaculture Symposium will provide intending farmers and aquaculture investors with a 'snapshot' of the current status of reef fish / grouper aquaculture. About 30–40 people are expected to attend this symposium.

CRIFI utilises the services of the Assessment Institute for Agricultural Technology, which has an office in each province in Indonesia. AIAT is administered by the Agency for Agricultural Research and Development, which also administers CRIFI. CRIFI research progress and outcomes are reported to AIAT at seminars and by direct contact. AIAT then extends research results to

farmers through meetings and workshops. The development of small scale backyard fish hatcheries for milkfish and seabass has provided RSCF Gondol with experience in extending research results to industry. About 800 of these hatcheries now operate in Bali, with many in the Gondol-Singaraja area. RSCF staff are confident that these hatcheries will readily adopt grouper production techniques once the technology is commercially viable, due to the high profitability of grouper larviculture.

SEAFDEC operates a Technology Verification Program (TVP), which is effectively a farm extension service. There are five SEAFDEC staff assigned to TVP. Most emphasis at the moment is on providing alternative technologies for the shrimp farms in The Philippines that have gone out of operation because of disease problems. One alternative to shrimp farming that is being actively promoted by TVP is grouper aquaculture. SEAFDEC held a 'Grouper Festival' on the island of Negros in March 1998, to promote grouper aquaculture specifically. According to SEAFDEC TVP staff, there is huge interest in grouper aquaculture in The Philippines and the development of additional grouper farms is constrained only by the poor availability of fingerlings.

A major component of this project is dedicated to ensuring that there is greater cooperation and exchange of information amongst grouper researchers and industry throughout the Asia-Pacific region. Research outcomes from the ACIAR project, as well as results from other research projects, are publicised in the dedicated 'Grouper News' section of 'Aquaculture Asia' (published by NACA) and the 'Live Reef Fish Bulletin' (published by the Secretariat for the Pacific Community). The widespread readership of these established and respected publications will ensure widespread dissemination of the results of the ACIAR project. This information is also provided on the NACA Grouper web site (http://naca.fisheries.go.th/grouper/) and outcomes of the ACIAR project are provided on the ACIAR project web site (http://naca.fisheries.go.th/aciar/). These mechanisms will ensure the widest possible extension of the outcomes of this project, and will 'value add' the ACIAR project by accessing the outcomes of other regional research projects to broaden the knowledge base relating to grouper aquaculture technology.

In addition, transfer of results to farmers and coastal communities is being actively supported under the APEC grouper research and development project.

Environmental impacts

The proposed research will potentially provide positive environmental impacts with regard to alleviating pressure on wild stocks that currently supply the bulk of the live reef fish markets. Currently, the demand for live groupers for the high value live fish markets of Hong Kong and southern China is being met largely by the capture fishery, and this fishery has been associated with unsustainable fishing practices, particularly the use of sodium cyanide as a capture technique.

The development of sustainable grouper aquaculture will contribute to the alleviation of adverse environmental impacts associated with unsustainable fishing practices by providing an alternative source of supply which will assist in meeting demand for grouper product. A specific strategy to develop grouper mariculture to reduce pressures on wild stocks is being developed in collaboration with TNC in the Komodo region of Nusa Tenggara Timur, Indonesia.

Increased cage and pond culture of groupers is likely to have some localised adverse environmental impacts, particularly related to water quality degradation associated with uneaten fish feed and fish wastes. Although published studies indicate that the overall contributions of nutrients and organic matter from coastal cage and pond culture of finfish are small compared with other coastal discharges, localised water quality changes and sediment accumulation may occur [Phillips, 1998 #544]. Such impacts tend to be greater from cage farms than from ponds because wastes can be assimilated to some extent within the pond environment, while wastes from cage farms are discharged directly into the local environment [Phillips, 1998 #544]. However, such impacts are highly localised and the overall impact of marine cage culture in coastal environments is minor [Phillips, 1998 #544; Wu, 1995 #545].

This project will assist in reducing adverse environmental impacts by developing a dry diet for grouper culture to replace the now commonly used trash fish diet. Losses associated with feeding trash fish are around 20–38% [Wu, 1995 #545] compared with around 10% for pelleted feeds [Wu, 1995 #545; Beveridge, 1996 #551]. Furthermore, this issue will be addressed directly through the development of guidelines on environmental management of grouper cage aquaculture planned for 2000–2001 under the APEC grouper research and development project.

Gender impacts

The impacts of this project will be gender neutral. Both men and women are employed in finfish aquaculture in the affected countries. The outcomes of the project will result in increased development, and improved sustainability, of grouper aquaculture. These outcomes will not impact on one gender exclusively, but will promote the economic development of the community in general.

Research problems

There have been relatively few research problems encountered to date. Some of the activities associated with the larval nutrition research have been delayed because of equipment breakdowns, particularly with the gas chromotagraphs at SEAFDEC and at RSCF Gondol. The RSCF GC has been fixed, and SEAFDEC have purchased a new Shimadzu GC that should be fully operational soon.

Some scientists at partner laboratories have had difficulty in accessing chemicals for their experimental work. Where possible, some chemicals have been purchased in Australia and supplied to facilitate the research at partner laboratories.

An on-going constraint is the limited success of larval rearing of groupers for the planned nutrition work in Australia. It is unlikely that the required numbers and sizes of juvenile groupers can be collected from the wild. CSIRO's Cleveland laboratory is an accredited quarantine station, so we will probably resolve this problem by importing juvenile groupers from the overseas partner laboratories, running the required nutrition experiments under strict quarantine procedures, and disposing of the fish once the experiments are finished.

Publications and other communication activities

Publications

The proceedings of the ACIAR–NACA workshop on Grouper Aquaculture Research, held in Bangkok on 7–8 April 1998 have been published by NACA. This document provides a valuable reference, since it includes overviews of the status of grouper aquaculture in most countries in the Asia-Pacific region; detailed technical information on grouper production technology; and an assessment of research needs and priorities for the sustainable development of grouper aquaculture in the Asia-Pacific region. Copies of the proceedings are available from NACA.

Project results and progress have been reported at a range of conferences, workshops and meetings throughout the region. These include:

- 9th International Symposium on Nutrition and Feeding in Fish, held at Seagaia, Miyazaki, Japan, 21–25 May 2000.
- The APEC Collaborative Grouper R&D Network Workshops held in Hat Yai, Thailand, 7–9 April 1999; and at Medan, Sumatra, Indonesia, 17–19 April 2000.
- The Annual Conference and Exposition of the World Aquaculture Society, held in Sydney, Australia, 26 April – 2 May 1999.
- First International Symposium on Cage Aquaculture in Asia, Taiwan Fisheries Research Institute, Tungkang Marine Laboratory, Tungkang, Taiwan, 2–6 November 1999.
- Fifth Asian Fisheries Forum, held in Chiang Mai, Thailand, 11–14 November 1998.

A full list of project publications is appended (Appendix 5).

Other communication activities

Grouper electronic newsletter

The grouper electronic newsletter, compiled and sent by Sih-Yang Sim (NACA). Yang has sent out four editions of the newsletter to date and it currently has over 160 subscribers.

'Grouper News' in regional aquaculture magazines

We have arranged for the regional aquaculture magazine 'Aquaculture Asia' (published by NACA) to carry a 'Grouper News' section that provides updates on activities within the region on the development of sustainable grouper aquaculture. This has appeared regularly since July 1998. Many of these articles are also included in 'The Live Reef Fish Bulletin'.

Web site

We have established a web site for the ACIAR grouper aquaculture project (http://naca.fisheries.go.th/aciar/). This is linked to the NACA grouper web site (http://naca.fisheries.go.th/grouper/) and contains the outline of the project and detailed project documentation. The site is maintained by Sih-Yang Sim and is updated regularly.

Value of the research

Social benefit

The development of sustainable grouper aquaculture technology will have impacts at all levels of the community in those countries involved in this industry. In Indonesia, many hatcheries are of the 'backyard' type – relatively small hatcheries that are basically family-run operations. Income from these hatcheries is reported to be of the order of Rp20,000 (AUD$3.50) for every three week production cycle (Dr A. Giri, pers. comm.). RSCF Gondol staff feel that production technology for groupers will be rapidly taken up by these small hatcheries, because of the highly profitable nature of grouper aquaculture.

In the Philippines, SEAFDECs TVP has found that there is excellent potential for the adaption of disused shrimp farms for grouper aquaculture. Currently, over 90% of Philippine shrimp farms lie idle because of shrimp disease problems. However, development of grouper farms is constrained by the limited availability of grouper fingerlings, which are available only from wild capture. Supply of hatchery-reared fingerlings would enable these disused farms to be productively employed for grouper culture. Because of the intensive nature of finfish farming (in Australia, barramundi farms employ one person for about every 8 tonnes of production, and this would likely be higher in the Philippines) the development of grouper farms from disused shrimp farms would be an important source of employment for local people.

Through the development of sustainable grouper aquaculture, employment opportunities would be provided for people with a wide range of skills. Hatcheries have a requirement for relatively skilled technicians, while farms utilise a wider range of skills, including relatively unskilled labour. Consequently, the development of improved grouper aquaculture technology will benefit a wide range of people in the community.

Economic benefit

Economic benefits resulting from this researched will be estimated during later stages of the project. As detailed above, the first year of the project has involved developing separate economic models for the three major phases of grouper aquaculture: hatchery, nursery and grow-out (ponds and cages). Future work will concentrate on further refining these models, and developing 'model farms' for Australia, Indonesia and the Philippines using realistic commercial data. The model farms will then be used to estimate the benefits of the research outcomes of the ACIAR project.

Existing information on the economic benefits of grouper aquaculture is summarised below.

Australia
DPI's Reef Fish Aquaculture Feasibility Study, carried out in 1995-96, predicted that considerable benefits would flow from the development of reef fish aquaculture in Australia. Costs to establish this industry were estimated at about $15 million over 10–13 years. The benefit/cost model showed that a reef fish aquaculture industry in Queensland has the potential to be highly profitable, generating revenue in excess of $1 billion within 30 years under favourable conditions. Using an 8% discount rate over 40 years, the net present value of the research project could be of the order of $170 million, with a benefit/cost ratio of 17:1, and an internal rate of return of 29%.

An additional aspect of the development of a reef fish aquaculture industry in northern Australia is the additional employment that would be generated. Northern Australian fish farms employ one person for every 8-10 tonnes of production (QDPI, unpublished data), so the development of a reef fish aquaculture industry is potentially a valuable source of employment for rural areas of northern Australia.

Indonesia
In Indonesia, RSCF Gondol has developed the technology for small-scale backyard fish hatcheries and about 800 of these hatcheries now operate in Bali. Milkfish are the species most commonly cultured, but Asian seabass are also cultured. RSCF staff expect that these backyard hatcheries will diversify into grouper species when the technology is suitable. The success of the backyard hatcheries is based on economics. The land now used for backyard hatcheries was previously used for coconut production. However, production of marine fish fry produces incomes several orders of magnitude higher than coconut production – e.g. an income of about Rp80,000 (AUD$14) per annum with coconuts compared to Rp20,000 (AUD$3.50) every three weeks for milkfish fry.

Philippines
SEAFDEC has undertaken economic evaluations of grouper production in both ponds and cages in the Philippines. For pond culture, based on a production unit of 5,000 pieces per ha per year with 80% survival to harvest at 5-7 months, the return on investment is about 82% and the payback period is 1.22 years, with an annual net profit of PHP244,210 (AUD$10,950) [Baliao, 1998 #552]. For floating net cages, one module of six cages holding 3,000 pieces with a survival rate of 80% to 5–7 months will produce a net profit of PHP 111,230 (AUD$4,850) per annum, with a return on investment of 59% and a payback period of 1.68 years [Baliao, 2000 #553].

Travel and meetings

The following travel has been undertaken:

- Mike Rimmer and Kev Williams travelled to Indonesia and the Philippines in June – July 1999 to visit laboratories participating in the ACIAR grouper project and to discuss the proposed work schedule for the first year of the project with project staff.

- Clarissa Marte, Joebert Toledo (SEAFDEC), Taufik Ahmad, Rachmansyah and Neltje Palinggi (RICF Maros), travelled to Bali to attend the initial project meeting held at the Gondol Research Station for Coastal Fisheries on 12–14 July 1999.

- Kev Williams travelled to Taiwan to attend the First International Symposium on Cage Aquaculture in Asia in November 1999. Kev presented two papers at the symposium: one on his ongoing feeds development and nutrition research, and another on the ACIAR grouper project and the Asia-Pacific Grouper Network. Kev also visited GRSCF, Bali, Indonesia, and SEAFDEC, Iloilo, the Philippines, for discussions with project staff on the ACIAR grouper project.

- Mike Rimmer travelled to Vietnam and Thailand in November – December 1999 to investigate opportunities to more closely involve Vietnamese aquaculture institutions in the Asia-Pacific Grouper Network. Mike spent several days at the NACA headquarters in Bangkok working with Mike Phillips and Sih Yang Sim on aspects of the ACIAR project and related activities (APEC projects and up-coming workshops).

- Kev Williams travelled to Vietnam and Indonesia in January – February 2000. The Vietnam component was funded by DFID to evaluate opportunities to replace trash fish in aquaculture with formulated feeds. Kev also visited RICF Maros, Sulawesi, Indonesia, for discussions on the ACIAR grouper project activities.

- Mike Rimmer travelled to the Philippines (SEAFDEC AQD) and Indonesia (RSCF Gondol, Bali, and RICF Maros, Sulawesi) in April 2000 to visit partner laboratories and to participate in the APEC–NACA–GOI–BOBP Sea Farming Workshop held in Medan, Sumatra, Indonesia, on 17–19 April 2000.

- The second project meeting will be held in Cairns, Queensland, Australia on 24–27 July 2000 and will be attended by project leaders from the partner laboratories.

Budget discussion

The initial distribution of funds was held up by the delay in finalising the Project Memoranda of Understanding with Indonesia and the Philippines. The initial six-monthly allocation was distributed in January 2000. The second six-monthly allocation was forwarded to the participating laboratories in May 2000. An acquittal for the first year of the project is appended (Appendix 8).

Conclusions

Despite delays in implementing the formal project arrangements, participating laboratories are to be congratulated for commencing work on the project immediately on its commencement. In several cases, this has meant that the laboratories have had to absorb the initial costs of the research while waiting for the ACIAR funds to be disbursed.

As this report demonstrates, significant progress has been made in a number of areas of the project. Overall the project is very close to on-target, although a few areas are slightly lagging – this is particularly the case with the training activities.

Appendix 1 – Recruitment advertisement for Fisheries Biologist (Grouper Culture) Position, Queensland Department of Primary Industries

Dept Account:	Primary Industries (PRI131)
Contact:	Mrs Kayleen Giezendanner
Phone:	47 222 634
Fax:	47 783 634
Ref No:	NR
Newspaper:	Courier Mail, Weekend Australian
Date:	Saturday -
No of Insertions:	One
Classification:	**Queensland Government Employment Opportunities**
Classification Level:	**PO3**

TEMPORARY FISHERIES BIOLOGIST (LARVICULTURE)

Aquaculture and Industry Development
Fisheries
North Region
Northern Fisheries Centre, Cairns
This position is temporary until 30 June 2002
Salary p.a. $ 42 901 – 46 849 VRN: NR
Key Duties:
Undertake research, development and extension associated with the ACIAR project 'Improved hatchery and grow-out technology for grouper culture in the Asia-Pacific region'.
Skills/Abilities:
Possession of a degree in agricultural science, veterinary science, applied science, rural science or equivalent qualifications in a field relevant to the position. Sound knowledge of, and demonstrated experience in, marine finfish larviculture research, including one or more of the following areas: larval finfish nutrition; histology; enzyme histochemistry.

Position Description: 07 47 222 634
Internet: http://www.dpi.qld.gov.au/dpi_vacancies
Closing date: 5.00 pm, Monday.