

# Excerpt from the Terminal Report on the NACA Project<sup>1</sup>

## 1. PROJECT BACKGROUND

Funded by the United Nations Development Programme (UNDP), Project RAS/76/003, 'Network of Aquaculture Centres in Asia' (NACA), was identified by the 1975 FAO Regional Workshop on Aquaculture Planning in Asia, and adopted by the 1976 FAO Technical Conference on Aquaculture in Kyoto.

The need of regional aquaculture centres was recognized on both occasions as *essential to the coordination of cooperative research, training and information exchange in a collective effort to promote aquaculture development on a regional basis, especially with emphasis on sharing available resources in accordance with the concept of Technical Cooperation among Developing Countries (TCDC)*.

Despite diverse development emphasis on aquaculture systems in the Asia and the Pacific Region, there was a keen interest to transfer known aquaculture technologies among the countries making it possible for effective cooperation in the transfer of technical know-how under the project's mandate. Accelerated collective development was made possible through cooperative research, training and information exchange activities.

## 2. OUTLINE OF OFFICIAL ARRANGEMENTS

The project was signed on 7 June 1979 by 11 participating governments, namely, Bangladesh, China, Hong Kong, India, Indonesia, Malaysia, Nepal, the Philippines, Singapore, Sri Lanka and Thailand. Activity implementation started in August 1980 upon the appointment of the Project Coordinator, with the project office located at the National Inland Fisheries Institute, Bangkok.

Project activities were carried out in these RLCs, each being a national aquaculture research centre in China, India, the Philippines and Thailand, and five National Centres (NCs), one each in Bangladesh, Nepal and the Philippines and two in Indonesia. Each centre focused on the species and farming systems, in which it excels for transfer to participating countries.

To review the programmes and progress of the project and to provide guidance and assistance in fulfilling its objectives, an Advisory Committee (AdCom) was constituted comprising representatives of the participating governments. It was later reconstituted as the Provisional Governing Council (PGC) in November 1986. The PGC would later become the Governing Council (GC) which would take over the implementation mechanism of the project in furthering regional cooperative aquaculture development through an autonomous intergovernmental NACA.

Beginning from July 1987, NACA was also responsible to manage the first phase of the project RAS/86/024, 'Regional Seafarming Development', with eight member countries: China, India, Indonesia, Korea (DRK), Korea (Rep.), the Philippines, Singapore and Thailand, each with a seafarming research institution designated as a Nodal Centre. The regional network of 17 aquaculture centres then covered the range of inland, brackish and marine aquatic environment and culture systems.

The NACA project had three phases. Phases I and II, under project RAS/76/003, lasted from June 1979 to June 1987. Either contributed or pledged, contributions from the

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<sup>1</sup> From the Terminal Report of the UNDP/FAO Project to Establish the Network of Aquaculture Centers in Asia-Pacific, FAO Fisheries Department, 1992. <http://www.fao.org/docs/eims/upload/69925/V5673E00.htm>.

governments totalled \$US 404 500 from 1985 to 1989. In addition, a special contribution of \$US 400 000 was made by China (TCDC/IPF) and the Thai Aid Programme in support of special activities.

The project terminated on 31 December 1989, when a full-fledged Independent Intergovernmental NACA became established to receive the regional responsibilities and the implementation mechanism of the UNDP/FAO NACA project. To enable it to be fully functional, the intergovernmental NACA was attached to the second phase of the project 'Regional Seafarming Development and Demonstration' in 1989–91.

### **3. OBJECTIVES OF THE PROJECT**

The development objective of the project was to enable the expansion of viable aquaculture systems through the improvement of existing technologies and the development of necessary technical expertise for implementing aquaculture development programmes and achieving production targets.

The immediate objectives of the three phases of the project were to:

#### **Phases I and II**

- carry out multidisciplinary research on selected aquafarming systems to adapt, improve, or develop new technologies;
- train core personnel needed for aquaculture development; and
- maintain a regional information system for the provision of appropriate data and information for development planning, research and training.

#### **Phase III**

- consolidate the establishment of an expanded network of aquaculture centres together sharing the responsibilities of research, training and information exchange essential to aquaculture development in the region;
- strengthen institutional and personnel linkages between national and regional aquaculture centres and their staff through exchange of technical personnel, know-how and information;
- achieve regional self-reliance in aquaculture development through the establishment of a self-supporting network that would continue to operate within the framework of Technical Cooperation among Developing Countries (TCDC); and
- promote the role of women in aquaculture development.

### **4. RESULTS AND CONCLUSIONS**

#### **4.1. IMPLEMENTATION STRATEGY**

The overall strategy in project implementation emphasized five priority regional needs:

- increase aquaculture production through effective transfer of established technologies in the region;
- train senior personnel in the planning and execution of aquaculture development and production undertakings;
- help justify government financial support to national aquaculture project; and
- take on only relevant adaptive research that facilitates increasing production, leaving basic research though complementary for academic institutions.

Priority was thus given to producing early tangible results for increasing aquaculture production in the region, in an attempt to assert the economic and social importance of aquaculture for the attention of development policy planners in governments. This was achieved by effective transfer of established viable, commercial technologies and techniques to applied research in both host and recipient countries to adapt established aquaculture production systems to local conditions. Through the cooperation among the RLCs and NCs technical and managerial details of established aquaculture production practices were systematically transferred by way of training courses, workshops and seminars, as well as specialized technical assistance.

In Asia established aquaculture production technologies have each a long history of existence evolved through traditional trial-and-error practices. Modern-day institutional research to improve production practices or to increase yield was problem-rather than system-oriented. The project thus emphasized research that would promote scientific understanding of vital interrelationships of salient dependent and independent variables for the improvement of production systems of regional importance.

#### **a. RESEARCH**

Research and development were mainly done in the 4 Regional Lead Centres (in China, India, Philippines and Thailand). The studies were on systems and species that each RLC had distinctive competence to do. The research areas were of priority to the country in which an RLC is located. Results were exchanged among the countries through training and information. Some of the research results were adapted to country situations by the National Aquaculture Centres.

The studies done during the project period appear as Attachment A.

The strategy for research was to improve on known technology for immediate application with the aim to increasing commercial production. Initial emphasis was therefore placed on biotechnical aspects, a high priority in the newly developing field of aquaculture. Effective increase in production provided the justification needed by the government technical departments to obtain more financial support from their governments and enabled the researchers to buy time for them to produce results to fill the numerous gaps in technology.

Following the principle of TCDC through pooling of resources and sharing of responsibilities, the RLCs conducted studies related to their specific areas of competence and priority farming systems without deviating from their national priorities. Results from the RLCs were tested and adapted in-country by the national centres. A number of improved technologies were tested in the field by participating institutions. A total of 92 working papers based on their research and seven World Food Day Technical Series publications were generated by the centres.

#### **b. TRAINING**

The lack of trained personnel, especially those with broad-based knowledge and experience in the practical aspects of aquaculture production, was recognized as one of the major constraints in aquaculture development. While many short-term training courses were being conducted in the region for aquaculturists, technicians and extension workers, they were directed at specific topics or techniques, usually with little coverage of interrelated disciplines needed to make a whole complete and viable aquaculture system. Therefore, one of the immediate objectives of the project was to train key personnel, comprising senior aquaculturists and technicians needed for planning and implementing aquaculture development programmes in various countries of the region,

providing them with a better appreciation of the multidisciplinary approach to aquaculture development planning.

Another approach adopted by the project was to transfer well-developed traditional technologies in any country of the region to other countries. One such technology selected for regular training was integrated fish-livestock-crop farming, a technology developed in China through many years of practical experience. This technology was felt to be eminently suitable to extend to countries the additional sources of employment needed in rural areas. In addition, to increase the variety of food crops and effectively utilizing wastes from agriculture and ancillary industries, this mixed farming system can also contribute to overall integrated rural development.

See Attachment 2 for the description of the courses and workshops.

The manpower development programme under NACA focused on aquaculture development planning and project implementation, production skills, and transfer of known and improved technology for direct application in national production programmes.

From 1981 to 1989, 16 sessions of two regular training courses were organized along with nine short courses and two workshops. One of the two regular courses was the one-year postgraduate, broad-based and multidisciplinary training programme for senior technical personnel involved in planning and implementing national aquaculture development projects. The RLCP training course was held seven times and 137 Senior Aquaculturists from 20 countries graduated. The RLCC four-month training course on integrated fish farming was of special importance to rural development in developing countries as it showed how farm resources can be used efficiently and helped generate more rural employment. The course was organized nine times at RLCC and 207 Senior Aquaculture Personnel from 36 countries graduated.

Nine short courses and two workshops were organized in response to the needs of the governments. The nine-month postgraduate course on fish disease diagnosis and tropical fish health management conducted at the Universiti Pertanian Malaysia was held in 1988–89. The secondment programme of junior scientists for one-year practical attachment and participation in the research programmes of the RLCs was a cost-effective and relatively rapid method of upgrading research personnel in the centres.

Overall, the training programme fortified the manpower base for technology development, enhanced regional cooperation in aquaculture development through the strengthening of the human network, and helped aquaculture attain a higher priority in national planning and policy formulation. The latter was a result of many of the NACA alumni, particularly of the Senior Aquaculturists and the Integrated Fish Farming courses, being subsequently promoted to strategic positions in their governments, as feedback from former trainees has shown.

### **c. AQUACULTURE INFORMATION SYSTEM (AQUIS)**

Related to the strategy of project implementation, the information component was designed to serve aquaculture production needs. Complementing the training and technology transfer activities, it consisted of publications, audio-visuals, and a computer-based aquaculture information system called AQUIS. Besides the quarterly NACA Newsletter, more than 90 working papers and seven technical publications in the World Food Day Series were produced, including '*Integrated Fish Farming in China*', a book developed by the RLC in China for the regular training course on integrated fish farming. These were disseminated to the governments and centres, as well as other collaborative institutions, organizations and agencies.

The computerized AQUIS, a numerical database, was less effective due to lack of backstopping from ADCP, which cancelled the programme on the mistaken belief that it was a bibliographic database. Nevertheless, efforts were initiated to revitalize AQUIS, in particular the formulation of a proposal for technical and funding support to a regional activity followed by in-country activities with the main purpose of developing an application for the collection, analysis, storage and retrieval of farm performance data of selected farming systems and technologies.

The exchange of information and experts' visits accelerated the transfer of technology and expertise between different parts of the network. The pooling of resources and sharing of information and research results avoided the duplication of research projects, an advantage which is all the more significant in the light of the lack of resources and manpower and the inadequate research facilities in the developing countries. While AQUIS was unable to achieve its objectives fully, it provided a pioneering example in the field of a numerical information system, providing data for farm performance evaluation and farming systems studies.

## **4.2 CREATION OF THE INDEPENDENT INTERGOVERNMENTAL**

### **NACA ORGANIZATION**

#### **Establishment and institutionalization of the Intergovernmental NACA Organization**

Major efforts were directed to successfully transform NACA from a project to an intergovernmental organization. This was achieved when the Intergovernmental NACA Organization was established during the First Governing Council Meeting held in Dhaka in December 1989. The primary activities toward the attainment of the objectives were:

- The draft Agreement on NACA was finalized in 1987 by the Second Provisional Governing Council Meeting. It was adopted with some amendments on 8 January 1988 at the Conference of Plenipotentiaries convened by FAO at its Regional Office for Asia and the Pacific (RAPA) in Bangkok.
- Other preparatory work for institutionalizing NACA included the formulation of the following:
  - Schedule of Government Contributions;
  - Rules and Procedures for the Organization;
  - Financial Regulations;
  - Employment Conditions;
  - Staff Regulations; and
  - the development of a Five-Year Work Programme for Regional Aquaculture Development under the Intergovernmental NACA.
- Initiatives were taken to generate collaborative support from donor governments and agencies to implement priority field activities under the Work Programme.
- In another effort to lay a strong foundation for the intergovernmental organization, a consultative meeting of agencies and organizations implementing aquaculture and related development programmes was organized by the project. The meeting adopted a set of recommendations meant to foster closer collaboration among participating organizations and to assist and strengthen the governments in managing the intergovernmental body.
- A core group of five regional experts recruited under Special Services Agreements were trained to take over the operation of NACA. Specialists from the Network centres can also be called upon to assist countries of the region in various disciplines related to aquaculture research and development.
- The Headquarters Agreement between the Government of Thailand and NACA was developed, with Thailand continuing to host the project coordinating office of

NACA and provide various immunities and privileges for the organization and staff.

## **Conclusion:**

The overall result of this activity was the establishment of a self-reliant and autonomous Intergovernmental NACA Organization. The strengthening of the Network centres attracted the collaboration of other organizations and agencies. An autonomous NACA organization, with its core programme funded by member governments, created a more conducive environment for bilateral and multilateral agencies to channel their assistance, thereby supporting the governments at managing NACA and further strengthening their collective efforts in expanding aquaculture development in the region.

With further collaborative support, NACA could continue to offer an excellent opportunity for donor governments and agencies to work together on activities of mutual interest and achieve the common goal of improving the socio-economic well-being of the region's people.

## **5. RECOMMENDATIONS**

One of the major tasks of the project was to create a fully functional, autonomous Intergovernmental NACA, to take over the regional aquaculture development mechanism of the project. Having demonstrated the effectiveness of the network of regional collaborative efforts in developing aquaculture, it was recommended that the Intergovernmental NACA be strengthened further and continue to establish collaborative arrangements with UNDP/FAO and international and donor agencies.

The strengthening of the network centres had attracted the collaboration of other organizations and agencies. An autonomous NACA organization, with its core programme funded by member governments, offered a more conducive environment for bilateral and multilateral agencies to channel their assistance, thereby supporting the governments at managing NACA and further strengthening their collective efforts in expanding aquaculture development in the region.

With further collaborative support, NACA would continue to offer an excellent opportunity for donor governments and agencies to work together on activities of mutual interest and achieve the common goal of improving the socio-economic well-being of the people.

It was envisaged that the obligatory contribution of member governments, based on the approved formula, was only sufficient to maintain a core staff nationals seconded by the governments or recruited directly by Intergovernmental NACA at a special scale lower than international rates. Therefore, donors have to be found for most of the field programmes. In this connection, the Five-year Programme of Work of NACA approved by the Provisional Governing Council during its third session held in Bangkok in January 1989 proposed a number of ways for obtaining external funding support. One of these was for NACA to undertake the responsibility of implementing projects of international agencies like UNDP and FAO, as well as the World Bank and Asian Development Bank, that fall within the field of interest and competence of the organization.

The diversity of problems in the region calls for cooperative regional actions for solutions. The network mechanism has proven the effectiveness of pooling of resources and sharing of responsibilities, as well as results of research and development in approaching common problems.

Moreover, the level of aquaculture production in the Asia and the Pacific Region can further be raised by increasing the present production acreage and/or by replacing the

present traditional with intensive culture systems. In either case, there are the inter-related socio-economic and environmental constraints associated with the involved changes. Since further increases in production are imminent, the region's countries would need to adopt a collective approach in dealing with common problems through planning and adoption of realistic policies for orderly development.

NACA's work programme for 1990–94 was planned with the above issues in consideration. Proposals for the support of research and training activities in this direction were formulated.

For the fish disease and fish health management programme, support came from the AsDB for a regional study on fish disease control and fish health management. This regional study consists of expert visits to countries, consultations and a regional workshop expected to recommend a regional action programme on fish health management following a network mechanism for research and information exchange, a region-wide fish disease monitoring and reporting system, and a fish health management scheme that includes prevention, diagnostics, treatment and regulation.

The interrelationships between the impact of environmental changes on the development of aquaculture and the impact of aquaculture itself on the environment should be emphasized. The objective of an environmental programme must ensure the development of the aquaculture sector in harmony with the rest of the economy.

NACA has emphasized the importance of research on the improvement of important aquafarming systems in the region at its lead centres. Proposals have been made to obtain funding support from donors to carry out farm performance surveys of selected systems and technologies in different countries in the region to provide the basis for development planning, investment and successful farm management.

On the study of integrated fish farming systems, the processing and analysis of data collected from China is being carried out. In addition, data will be collected from other countries in the region, and further experimental studies implemented to delineate pond dynamics and waste recycling. Appropriate bio-economic models of integrated fish farming systems, as well as models of modified systems will be constructed for the different sub-regions for field trials. Results obtained will be disseminated for use in formulating appropriate rural development programmes.

Programmes on the socio-economic aspects of aquaculture development will be undertaken. These will help to develop the capability of national administrators and planners to ensure sustainable aquaculture for the betterment of economic growth and social development. Through these programmes, NACA will also provide assistance to governments in preparing national aquaculture development plans and undertaking feasibility studies for the formulation of investment projects in the region.

Through interdisciplinary research, it is possible to improve the efficiency of aquaculture production systems as in the case of animal husbandry, in which the interrelationships of various component disciplines (e.g., animal health, nutrition, reproduction and genetics) have been established and integrated into a multidisciplinary body of knowledge. Discipline-oriented studies on certain special areas are being done in NACA lead centres, but tertiary level education in the various disciplines, which can complement and strengthen aquaculture development programmes, is lacking in the region. However, certain universities and institutions do have strengths in some special areas within these disciplines. As indicated in its Work Programme for 1990–94 NACA will endeavour to assist in developing or upgrading tertiary level educational and advanced level research activities in selected institutions/universities within the region which would serve as centres of excellence in particular disciplines in meeting training needs.

The NACA and Seafarming (RAS/90/002) projects have shared management resources under a cost-effective arrangement. When the seafarming project terminates, its integration into the Intergovernmental NACA will expand the network with the addition of the eight seafarming nodal centres. This will also effectively bring coastal and marine aquaculture into the NACA umbrella.

Aquaculture has been largely traditional until a decade ago. The priority then was to increase production and therefore production technology was needed. At present, most of the technical skills and technologies are available for most culture systems. Research and development should now adopt a multidisciplinary approach in order to overcome the broader, non-biotechnical constraints. The network umbrella concept will be most appropriate in developing a regionally coordinated multidisciplinary research and development programme in which various centres of excellence will have the responsibility for specific disciplines. The same pooling of resources and sharing of responsibilities adopted by the NACA project will be followed.

One of the initiatives of the project to help lay a firm foundation for the Intergovernmental NACA was the organization in June 1989 of a consultative meeting among agencies and organizations in the region implementing aquaculture development and related projects. The meeting adopted a set of recommendations to assure collaboration among them, foster cooperation in areas of mutual interests and avoid duplication of efforts. The other initiative is liaising with donor governments and agencies with the view of seeking collaborative support for the implementation of some of the field activities under the NACA Programme of Work. These are essential preparatory actions for the establishment of a fully functional independent NACA organization.

As originally planned, the project was phased out by 1989. However, consultations with officials concerned with the participating governments and institutions showed the need for international assistance in the early stages of the NACA network operating independently for the first time as an intergovernmental organization. The assistance would firm up the foundation for the Intergovernmental NACA by providing advisory activities and funding support needed to consolidate and improve ongoing regional activities, initiate new programmes, mobilize funding support from various donor agencies and liaise with other institutions in and outside the region. It would prepare the Governments to fully assume the funding for the core programme through their mandatory contributions. It would also permit NACA to continue to engage the services of the regional and national experts who have been seconded to the project by their governments and therefore are already trained in the various activities required to operate the network.



## **Attachment A: Research under the Regional Lead Centres**

### **1. The Regional Lead Centre in China (RLCC)**

#### **1. Relationships between pond size and depth on yield in pig-manured ponds**

To further facilitate production management practices, the RLCC carried out a series of culture trials on the effect of five pond sizes and one depth of pig-manured waters on fish yield using fingerlings of the four major Chinese carps at 1 000 fish/mu, and later included Wuchang fish and Japanese koi at 1 000 fish/mu. Green fodder was given as supplementary feed.

Using water depth as a variable in similar trials, results conclusively showed that yield was directly correlated with water depth. Particularly silver carp, bighead carp and blunt-snout bream yielded significantly a greater biomass in deeper ponds.

#### **2 Effect of various animal wastes on fish yield**

In 1981 chicken, pig and cow manure was used in cement tanks for comparative growth trials using silver, bighead and common carps and tilapia. Results averaged 90% survival and a manure conversion coefficient of 5.7. Survival rate in chicken-manured tanks was higher (98%) as compared with 90.7% in tanks using pig manure and 87.3% in cow manure.

In 1982, similar trials were duplicated in larger 1 mu tanks, except that tilapia was replaced by Japanese Koi. Results improved showing 96% survival and a manure conversion coefficient of 2.5. The increment coefficient body weight was also higher at 3.58, 3.16, 3.15 and 2.53 for chicken, duck, pig and cow manure, respectively. Yield averaged 185 kg/mu for a stocking rate of 1 000 fish/mu, but only 137.5 kg/mu for a higher stocking rate of 2 220 fish/mu. In 1984 trials, pond water fertilized with cow as compared with pig manure resulted in significantly higher weight increment for silver, bighead, crucian and common carps, while pig manure was found to be much more superior than poultry manure.

#### **3 Comparative study of the effect of fresh and fermented animal manure on fish yield**

Studies carried out from 1986 onwards showed conclusively that fresh pig manure promoted significantly better fish growth than fermented pig manure and chemical fertilizers by 167% and 229%, respectively. Fresh chicken manure showed similar results. These studies showed that carps feed directly on the fresh manure, in addition to natural food.

#### **4 Comparative studies on the effect of fresh and fermented green manure on fish yield**

Preliminary studies conducted indicate that Sudan grass, water hyacinth, mugwort and alligator alternanthera can fertilize the pond water effectively. Experimental studies were carried out with fresh and composed green manure in ponds stocked with fingerlings of silver carp, bighead carp, common carp and crucian carp. The results obtained thereof were analysed.

#### **5 Effect of frequency of manure application on fish yield**

Initiated in 1986, fresh or fermented pig manure was applied in single or several split doses, keeping the total quantity applied in the course of a month the same in all the experimental ponds, which were stocked with two different combinations of silver carp, bighead carp and koi, and bighead carp, common carp and crucian carp. Ponds that

received daily doses of fresh or fermented pig manure gave better fish yields than those that received the manure at intervals. The bighead carp grew better than silver carp and koi in all the ponds.

At a stocking density of 6 333 fish/ha and with daily application of fermented pig manure at 41.8 kg/ha without any additional feed, the combination involving bighead carp, common carp and crucian carp yielded 30–40% higher than with manure application at intervals of five to seven days.

#### 6. Effect of manuring on fish disease and sanitation

Water and fish samples taken from ponds fertilized with pig, cow, chicken or duck manure and from the control pond were analysed for the presence of pathogenic bacteria. Out of more than 400 bacteria isolated, 110 were found to be pathogenic to fish.

Beginning in 1987, a survey was conducted on the relationship between human intestinal disease and bacteria on the body surface of fish raised in ponds treated with animal manure. The results show the presence of microbes which can be harmful to man if consumed. Among manured ponds, coliform bacteria count was highest in ponds treated with pig or cattle manure.

#### 7. Use of delta carbon as a tracer of nutrient pathways

Stable carbon isotope ( $\delta c$ ) was used to trace the carbon pathways of agriculture and livestock wastes as it enters the fish through the food web in integrated fish ponds. Analyses of  $\delta c$  values were made in fish, phytoplankton, periphyton, pig manure, vegetation and pond mud. The results indicated that delta carbon could be an effective tool in establishing a better understanding of the biological processes in an integrated fish farm. This would aid the transfer and adaptation of integrated fish farming technology in areas with different agro-climatic conditions.

Further studies with delta carbon in manured ponds showed that a third of the carbon found in the flesh of the omnivorous Japanese carp (*Carassius cuvieri*) was derived from the autotrophic production system and another third from the heterotrophic production system. In the case of common carp, more than half of the carbon was found to have been derived from the autotrophs and the rest from the heterotrophs. From these studies it appears that the Japanese carp feeds more on bacteria, organic detritus and plankton of size range 50–120  $\mu$  the common carp on periphyton, the silver carp on plankton smaller than 50  $\mu$  and the bighead carp on plankton of size range 50–120  $\mu$ .

#### 8. Modelling of integrated farming systems

A nationwide survey was conducted from 1982 to 1989 to collect input-output data from common integrated fish farming systems to study the economics of integrated fish farming as practised in China and to find the best systems. The study brought to light a variety of integrated fish farming practices, each of which was a system of balanced ecology and economics. Species composition using grass carp and Wuchang fish as the main species was found to be the most economic system, while the rotatory method of harvesting and stocking within a limited period was considered the most beneficial practice. The system of integrating fish farming with cultivation of grass on the dykes was found to be decidedly more economical than the mulberry-dyke fish pond system in some parts of China. Investigatory data were used to prepare a report on empirical models of integrated fish farming systems in China.

#### 9. Effect of stocking ratio on fish production in polyculture

With grass carp as the main species and silver carp and bighead carp as subsidiary species, using only green fodder as feed, the best ratio between them was found to be 10:2, followed by 10:3 and 10:4. It was also found that common carp could be added to these combinations at the rate of 5–8% of the total number of stocked fish.

Experiments were also undertaken with silver carp and bighead carp as the main species, using only pig manure as fertilizer.

#### 10. Effect of night aeration on fish yield

Since the demand for dissolved oxygen in a fish pond was greatest from midnight to early morning, aeration was applied from midnight till 06.00 a.m. This sustained the dissolved oxygen level above 4 mg/l, which enabled increasing utilization of available feed. Fish production in experimental ponds receiving aeration at night was invariably at a higher rate than in control ponds.

## 2. The Regional Lead Centre in India (RLCI)

### 1. Fish genetics

#### a. Gynogenesis

Artificial gynogenesis was successfully carried out in a number of Indian major carps and exotic carps, namely catla, rohu mrigal, common carp and silver carp, through fertilization of eggs with sperm genetically inactivated by ultraviolet rays, followed by thermal shocks. This retained only maternal inheritance in the offspring, since the gynogenetic fish could be used to produce inbred lines, and the intraspecific breeding of these would make it possible to obtain heterosis effect. Diploidy in eggs fertilized by irradiated milt could be restored in Indian major carps either through cold shock (12°C) or heat shock (39°–40°C). Fertilized eggs which were not subjected to thermal shocks resulted only in haploids, which were characterized by structural deformities (haploidy syndrome).

First generation gynogenetic progeny  $G_1$  of mrigal was successfully bred and  $G_2$  progeny obtained. Some of the  $G_1$  and  $G_2$  rohu were fed methyl testosterone (100 mg/kg of feed) for induction of sex reversal. Phenotypic males of  $G_2$  mrigal attained an average size of 290 mm of 550 g in ponds in one year. On the other hand, the growth rate of rohu was comparatively slow: the average size attained was only 250 g in one year and nine months.  $G_2$  rohu treated with methyl testosterone grew into males with normal tests and attained a size of 750 g in two years and nine months.

#### b. Androgenesis

Androgenesis was attempted by crossing distantly related carps and success was achieved in producing androgenetic progeny of common carp-catla, common carp-rohu and common carp-mrigal.

#### c. Polyploidy

Polyploid mosaics were successfully induced in rohu with colchicine treatment 25 minutes after fertilization. Polyploid-diploid mosaics were observed in the majority of treated fish.

Triploid and tetraploid rohu were also produced for the first time by subjecting fertilized eggs to thermal shocks prior to release of first polar body and first cleavage respectively. Their growth was rather slow, with the triploid attaining a size of 175 mm of 45 g in eight

months and the tetraploid 144 mm of 37 g in 10 months. In a later experiment, the rohu polyploids were found to attain average weights of only 75 g and 270 g in 18 months and 30 months, respectively.

#### d. Hybridization

Several intergeneric hybrids were successfully produced, viz., common carp x catla, common carp x rohu, common carp x mrigal, grass carp x silver carp, rohu x catla and some reciprocal crosses of the above combinations. Rohu-catla hybrids were found to be viable, attaining their first maturity when three years old. They could be successfully bred through hypophyztion. The high survival rate of the hybrids in some ponds indicated the possibility of producing them commercially. Rohu-catla hybrid was successfully backcrossed with catla male. The various intergeneric hybrids between Indian major carps and common carp grew better in monoculture than in polyculture and showed lower escapement during netting than common carp, but higher than catla. Catla-rohu, rohu-catla, common carp-catla and common carp-rohu hybrids were successfully produced on a large scale for use in production trials.

#### e. Karyological studies

Karyological studies of grass carp female x silver carp male hybrid indicated its diploid chromosome number to be 48, the same as that of the parent species. The chromosome number in common carp x rohu, common carp x catla, and common carp x mrigal hybrids was found to be intermediate between those of the parent species, varying from 74 to 76.

A modified chromosome methodology of colchicine - airdrying Giemsa staining technique for karyotypic studies in fish was developed. This made it possible to obtain a large number of good metaphase spreads in catla, rohu, mrigal, common carp, grass carp, silver carp and their hybrids, and in some types of catfish and air-breathing fish.

#### f. Cryopreservation of carp spermatozoa

Spermatozoa of rohu, mrigal, catla and magur (*Clarias batrachus*) were for the first time cryopreserved successfully for one year. However, hatching in fertility tests with cryopreserved spermatozoa was poor, not exceeding 20%.

## 2. Fish nutrition

### a. Composition of feed ingredients and feeds

About 100 potential feed ingredients obtained from different parts of India were analysed for their approximate composition.

### b. Feed formulation

A freeze-dried diet of micro-encapsulated whole egg, fortified with minerals and vitamins, was prepared for the first time for feeding spawn in the hatchery during the first 10 to 15 days. This feed induced better growth in grass carp and common carp than traditional feed, while the silver carp did better on the traditional feed. However, plankton plus traditional feed, and plankton alone, gave better results than micro-encapsulated feed for all three species.

Several feeds were formulated making use of locally available ingredients. Among the various compositions were:

- gingili oil cake + groundnut oil cake (GOC) + rice bran;
- GOC + rice bran;
- gingili oil cake + rice bran;
- GOC + wheat bran;
- GOC + salseed cake + rice bran;
- gingili oil cake + GOC;
- chicken feed; and
- GOC + goat liver + rice bran, all fortified with vitamins, minerals and trace elements.

Pelleted feeds were found to be superior to moist feeds in promoting fish growth. A balanced diet with 35% crude protein gave a high rate of survival in Indian major carp spawn. *Ottelia* powder as a protein source gave results comparable to those with conventional oil cake + bran mixture. Incorporation of commercial grade fish meal at 5–50% in diets compounded with soybean and groundnut cake as protein source showed adverse effects on protein deposition and an increase in fat level in rohu fry. This suggests that poor quality fish meal should be avoided, even in small quantities. Six different feeds were formulated with more than 47% crude protein for *Clarias batrachus*.

#### c. Protein and amino-acid requirements of carp fry and fingerlings

The crude protein requirement in feeds was found to be 40% for rohu and mrigal fry and 45% for catla fry. Specific growth rate, protein retention values, protein efficiency ratio, protein assimilation and urinary nitrogenous excretion were the highest and the trophic coefficient the lowest at these respective levels. These diets also were the most efficient for flesh production, flesh produced in assimilation and protein produced in assimilated protein. The protein requirement of silver carp fry was found to be 31% at 29°C.

The protein requirement in feeds for catla, rohu and mrigal fingerlings was found to be 40%, and 36% for grass carp fingerlings.

The essential amino-acid requirement of Indian major carps was found to be almost equal and similar to that of common carp. The total essential amino-acid requirement of rohu fingerlings was found to be 27% of the diet, which forms the basis for the formulation of synthetic diets. Different combinations of casein and gelatin were employed as protein source in studies to ascertain the amino-acid requirements of Indian major carps. Catla and rohu showed highest growth, feed efficiency and protein and energy deposition with the feed containing casein-gelatin in the proportion 35:10. Mrigal, however, was unaffected by changes in these proportions.

In studies on the protein-energy ratio in the diet of rohu fingerlings, the best growth was obtained with the diet containing 38% protein and 3.25–4.00 kcal/g energy.

#### d. Protein-saving effect of dietary carbohydrate

A combination of 35% protein and 38% carbohydrate gave the best performance in catla fry, while a 40% protein and 32% carbohydrate mix gave the best results for rohu fry.

### 3. Ichthyopathology and fish health protection

The various diseases encountered in Indian major carps were diagnosed. Among the parasitic genera detected *Trichodina*, *Ichthyophthirius*, *Dactylogyrus*, *Posthodiplostomum*, and *Sanguinicola* may be mentioned. Investigations on pathogenicity of newly identified parasites, as well as on histopathology, prevention and treatment of various diseases, were carried out. A communicable disease causing emaciation, haemorrhages on body surface and accumulation of ascitic fluid in the

coelomic cavity was encountered in silver carp and studies were undertaken to identify the bacterial pathogen isolated from kidney, liver, heart and coelomic fluid of affected fish. Infection of kidney and gills of catla fingerlings by *Myxobolus spp.* caused mass mortality. *Brachiomyces spp.*, a fungus caused large-scale mortality of mrigal fry in ponds with high organic load, and several antifungal drugs were tried to control the disease.

Prophylactic antibiotic treatment, followed by pond treatment with malathion, enhanced the survival rate of spawn and fry of Indian major carps by 35%. Malathion was found to be effective against copepods in ponds. Ascorbic acid-antibiotic combination in feed was found to be most effective in reducing post-handling mortality.

Acute cases of argillaceous in broodstock ponds were successfully controlled and its recurrence prevented by three successive malathion treatments at weekly intervals. It was also effectively cured by application of 0.25 ppm of malathion in the pond in combination with dip treatment of the fish in 500 ppm of  $\text{KMnO}_4$  solution. Columnaris disease, caused by *Flexibacter columnaris*, was recorded for the first time in Indian carps. Mortality caused by this disease was considerably reduced by prophylactic antibiotic treatment.

Epizootic Ulcerative Syndrome (EUS) disease has received special attention in recent years. The occurrence of this disease was seen to be mild in Indian major carps, but severe in *Puntius spp.*, *Channa spp.*, *Mastacembelus spp.*, *Clarias batrachus* and *Heteropneustes fossilis*. Only myxozoan parasites were seen, while the bacteria encountered were similar to *Aeromonas hydrophila*, *Salmonella spp.*, *Arthrobacter spp.*, *Actinomycetes spp.*, *Pseudomonas spp.* and *Clostridium spp.* Vital forms showing cytopathic effect in EPC and BB cell lines were also isolated, while the fungus *Aspergillus spp.* was encountered in liver. The water quality parameters in the affected water bodies did not show any abnormal values. Liming was found to be the only effective method of controlling the disease, the dosage being 600 kg of shell lime (unslaked) per hectare per metre depth, in three split doses of 200 kg each at seven-day intervals.

Several strains of *Aeromonas hydrophila* were tested for their drug sensitivity so that suitable preventive and curative measures could be planned. A vaccine using several strains of *A. hydrophila* was successfully developed. Success was also achieved in preparing antigen for *A. hydrophila* and in raising anti-sera in rabbits.

Four fish cell lines (EPC, FHM, BB and SCT) were successfully developed from Indian major carp tissues and maintained in various sub-passage levels for use in diagnostic virology.

#### 4. Broodstock management

This study was aimed at determining the stocking density, supplemental feeding and pond management measures for optimal growth, sexual maturation and quality and quantity of reproductive products.

Prophylactic antibiotic treatment of broodstock to ward off columnaris disease helped in reducing post-spawning mortality, while feeding with balanced formulated feeds led to better spawning.

Extensive work was carried out on the nutritional requirements of catla, rohu, mrigal, grass carp and silver carp broodstock of 1.5–2 kg, stocked at 1 500 kg/ha. In comparative feeding trials with the traditional diet of GOC + rice bran in equal proportions by weight and formulated pelleted feed (GOC + rice bran + dibasic

phosphate + multivitamins) with 30% protein, 35% carbohydrates and 11% fat, fed daily at 3% of body weight, gonadal development, condition and percentage of success in induced breeding appeared to be better with the formulated feed. Additional feed in the form of aquatic weeds was provided only for grass carp, at 25% of body weight. In further trials with this formulated feed, in which the above species were stocked together in equal proportions or in the ratio 3:2:2:2:1, gonadal maturation was significantly advanced and spawning took place as early as April.

Studies carried out on the effect of aeration and water replenishment on the maturation and spawning of broodstock of the above species revealed that the addition of 20% of original water volume twice a month for four months promoted better gonadal development and better response to hypophyztion than daily aeration for 5 h.

Using well water, mrigal could be induced to breed in late April, much ahead of the normal breeding season, which begins in late June. It was also possible to advance its maturity to April through photoperiod and temperature manipulation. A single dose of a homeopathic drug advanced maturity in catla to early May, while *Clarias batrachus* implanted with LHRH-A pellets attained early maturity and responded better to hypophyztion.

#### 5. Fry and fingerling rearing

Rearing of carp fry and fingerlings with aeration gave significantly better growth in all the treatments tried, viz., with feed or organic fertilizer (cow dung) or inorganic fertilizers (N-P-K) only.

A six-month experiment was carried out to rear rohu and silver carp fry to fingerlings separately and in combination (1:1), at a stocking density of 0.1 million/ha, to investigate the possibilities of holding the fish at fry and fingerling sizes over a prolonged period. The average stocking size of rohu was 47.2 mm/1.75 g, while that of silver carp was 55 mm/2 g. The results obtained are tabulated below.

Table 1

PERCENTAGE SURVIVAL AND SIZE OF ROHU AND SILVER CARP AFTER SIX MONTHS OF REARING

Culture method	Species	Survival (%)	Size (mm/g)
Stocked separately	Rohu	98–99	89.93/7.5–107.2/12.5
	Silver carp	51.9–52.2	128.9/18–135/20
Stocked together	Rohu	90.1–96.6	110.47/12–124/18
	Silver carp	69–86.2	109/11–133/16

Rohu showed better growth but a lower survival rate when cultured together with silver carp than when culture alone. Silver carp, however, showed better survival and poorer growth in combined culture. Rohu seed showed distinctly better survival than silver carp both in monoculture and combined culture, the overall survival percentages being 96.6 and 60.4 respectively. On the other hand, silver carp showed better growth than rohu in monoculture, though growth was marginally poorer in combined culture.

About 80% survival was obtained in rearing carp spawn at a stocking density of 5 million/ha, using a formulated diet of GOC + soybean oil cake + rice bran + fish meal, with a protein content of 32 %.

In an experiment to study the growth performance of carp fry in ammonia-treated ponds, the application of ammonia along with cobalt, manganese and single superphosphate gave significantly higher growth and survival than other treatments, which consisted of: (i) ammonia + single superphosphate; (ii) ammonia alone; and (iii) bleaching powder + urea + single superphosphate.

## 6. Management of undrainable ponds

A pond environment monitoring system involving 31 parameters was developed to study the chemical and biological characteristics of undrainable fish culture ponds. The initial survey indicated that the sediment was practically devoid of natural animal feed for benthophagous fishes, but was rich in nutrients. The basic production and decomposition processes were quantified in ponds receiving different levels and methods of organic manuring. Deep sediment layers rich in organic matter played an adverse role in community metabolism.

The application of fermented water hyacinth in ponds led to abundant plankton growth, which in turn promoted good fish growth.

A case of mass fish kill was found to be due to oxygen depletion caused by a temporary bloom of sulphur bacteria, resulting from the high rate of decomposition of organic matter.

The nitrogen fixation potential of fish ponds receiving organic and inorganic manuring was assessed with reference to bacterial populations involved in the nitrogen cycle. It was possible to improve nitrogen fixation levels through biofertilization with *Azolla* and heterocystous blue green algae (*Anabaena*, *Nostoc* and *Calothrix*). A culture system with a production capacity of 3 t/month was developed for *Azolla*, which could be applied to fish ponds as a biofertilizer. Poultry manure was found to be superior to cow dung at fixing nitrogen in water through aerobic nitrogen fixing bacteria.

Secondary sewage allowed to flow through two water hyacinth culture basins was found to be suitable for fish culture, because this reduced its excessive nutrient content by 70.8%. A net production of 1 765 kg/ha in only seven months of composite carp culture was attained.

In weed control experiments, prickly lily (*Euryale ferox*) was cleared in ten days with 2, 4-D sodium salt at 8 kg/ha, while *Typha* was killed by a spray of gramoxone.

Bleaching powder (at 5 mg/l chlorine) in combination with urea (at 5 mg/l ammonia) was found to kill all aquatic animals, including large murels. The residual fertilizing effect of ammonia serves to reduce management expenditure.

In manuring trials, cow dung slurry at 10 t/ha/year gave better fish yield than biogas slurry applied at 15 and 30 t/ha/year. Better fish production was obtained in composite culture of Indian major carps and Chinese carps when both the organic and inorganic fertilizers were applied in standard doses and methods than (i) when inorganic fertilizers were given in bags for leaching, and (ii) when organic manuring was not resorted to, but the pond bottom was raked on alternate days. The latter treatment yielded the least production, even though bottom raking released nutrients, which, however, appeared to be utilized by macrophyte communities.

In both monoculture and mixed culture of Indian major carps, rohu performed better than catla and mrigal in terms of both survival and growth. Rohu was also seen to perform better in weed infested ponds.



Feeding frequency appeared to influence fish yield significantly. When carp were stocked at 10 000 fingerlings/ha, production differed significantly between four and six feedings per day, the mean net production being 1 750 and 1 925 kg/ha respectively in three months of rearing.

## 7. Aquaculture economies

### a. Economic analysis of freshwater aquaculture operations

Empirical modelling work was undertaken for about 250 experimental and pilot scale freshwater aquaculture operations in India, taking into account six variables for categorizing input/output data:

- organic fertilizer;
- inorganic fertilizers;
- Mahua oil cake;
- rice bran;
- weeds; and
- stocking number.

The following model was developed to estimate total fish production:

$$\begin{aligned} \text{YFISH} = & -176013 + 83.2494 \text{ MAH} - 0.119558 \text{ MAH}^2 \\ & + 64.1404 \text{ COW} - 0.00515717 \text{ COW}^2 \\ & + 1502.58 \text{ CHE} + 233.644 \text{ FEED} \\ & + 14421.4 \text{ WEE} - 22.9051 \text{ WEE}^2 \\ & + 175.561 \text{ TONU} \end{aligned}$$

Where:

YFISH	=	Total fish in grams
MAH	=	Mahua oil cake in kilogram
COW	=	Cow dung in kilogram
CHE	=	Chemical fertilizers in kilogram
FEED	=	Feed in kilogram
WEE	=	Weeds in tons
TONU	=	Total number

The above model explained 80 % of variation in output. Feed was found to be the key explanatory variable with highest output elasticity, and therefore farmers would benefit by feeding the fish at optimal levels.

### b. Bio-economic modelling

In connection with bio-economic modelling to optimize the economics of carp culture, studies were carried out to estimate certain biological parameters, such as feed suitability, coefficient, growth coefficient and fasting catabolism.

### c. Economics of filling undrainable ponds

Windmill-drawn water was found to be the cheapest, followed by that drawn from irrigational canals. Water from dug wells proved to be the most economical.

## 8. Catfish culture

Techniques were successfully developed for the breeding and culture of *Ompok bimaculatus*, *Wallago attu* and *Clarias batrachus*.

All three species can be bred through hormone administration. *Clarias batrachus* was spawned by a single dose of 30 mg of Indian major carp pituitary or 4 000 I.U. of HCG/kg of female only. The maturity of this fish was substantially advanced through implantation of LHRH pellets. Through proper broodstock management and hormone treatment, it was possible to breed this fish from April to October, covering pre-breeding, breeding and postbreeding seasons. Mini hatcheries were set up for this species at two centres.

Molluscan meat was found to be suitable for both *Wallago* and *Ompok*. *Mystus seenghala*, another catfish taken up for culture, was found to feed nocturnally during late evening and the early night hours. It showed a marked preference for fish flesh, followed by prawns and molluscs.

#### 9. Prawn breeding and culture

Laboratory-reared seed of *Macrobrachium rosenbergii* produced in October 1988 attained first maturity in June 1989 and berried prawns were observed until September 1989. Meat of freshwater mussel was exclusively used as larval diet. Larval mortality in outdoor rearing could be minimized by using a chemotherapeutic drug. These findings could be applied in the management of giant freshwater prawn hatcheries in India.

#### 10. Culture of freshwater molluscs

The pearl mussels, *Lamellidens marginalis* and *L. corrianus*, recorded a survival of 83 % when suspended at pond surface, as against only 13 % when set at the bottom.

The highly migratory nature of *Achatina fulica*, *Pila spp.*, and *Laevicaulis alte* could be curbed by employing simple metallic copper or copper sulphate barriers.

#### 11. Fish breeding

Intraperitoneal injection of pimozide, a dopamine antagonist, at 10 mg/kg 4 h prior to the administration of LHRH-A at 20u/kg body weight, was highly effective in the induced spawning of catla, rohu and mrigal.

Administration of Winstrol, an anabolic steroid, at 4 mg/kg/month from May to July 1989 to mature rohu led to the development of the highest number of oocytes and highest percentage of fertilization, as compared to progesterone-treated and control groups. This indicated that Winstrol could be used as an inducer for final oocyte maturation.

### 3. The Regional Lead Centre in Thailand (RLCT)

#### 1. *Channa striatus* culture system

A survey was carried out to study the artisanal practices of *Channa striata* culture, their problems and economic aspects.

Detailed laboratory studies were later conducted to assess the lipid and vitamin requirements of snakehead fingerlings. The optimum level of dietary lipid and vitamin requirements of snakehead fingerlings. The optimum level of dietary lipid was found to be 6 % and no significant difference could be seen in survival, growth and conversion

between the lipid sources tried, viz., soybean oil, *Trichogaster* oil and *Pangasius* oil. Pantothenic acid was found to be a particularly important vitamin for snakehead growth and survival.

## 2. *Macrobrachium* culture system

By filtering the culture water through carbon and EDTA, it was possible to reduce the high rate of mortality of *Macrobrachium rosenbergii* postlarvae in hatcheries significantly. In trying to understand the effect of water quality on larval survival, a study on acute toxicity of ammonia was conducted, which revealed the 48 hours  $LC_{50}$  values for 5, 12, 23 and 35-day old larvae to be 1.53, 1.85, 2.05 and 1.77 ppm of unionized ammonia respectively.

A system-oriented and multidisciplinary research programme was conducted on the grow-out aspect of the *Macrobrachium* culture system, including studies on stocking density, feed and feeding, fertilization and environmental changes.

A small-scale hatchery successfully produced juveniles of this prawn using saline water prepared from salt-pan residues and 71–90 % survival from larval to juvenile stage was achieved. The great practical application of this would be *Macrobrachium* culture further inland from the coastal region.

An experiment conducted in grow-out ponds revealed that aeration had no effect on *Macrobrachium* production at a low stocking density of 6 individuals/m<sup>2</sup>, the mean stocking weight being 4.18 g. On the other hand, intrinsic biological characteristics (large variations in sex ratio and male phenotypes) greatly affected the yield and market values. The average yield over a seven-month cycle amounted to 1 312 kg/ha, which gave a net income of \$US 1 890, with 41 % profitability.

In experiments to ascertain the vitamin requirements of *Macrobrachium*, it was found that there was no difference in its growth rate and survival when vitamins were fed at 0.5 %, 1.0% and 1.5%. However, the growth rate was faster than that of the control group, which was not given any vitamin premix. Likewise, there was no significant difference in growth when prawns were fed at five levels of dietary protein, ranging from 20% to 40%.

Sex differentiation in *Macrobrachium* generally took 100–120 days after hatching, but was observed to start in 70 days in fast growing individuals.

Histopathological studies were made of prawns affected by some of the severe diseases encountered, viz., shell disease, idiopathic muscle necrosis and red discoloration disease. A variety of bacteria were isolated from the muscle tissue of prawns with red discoloration disease.

## 3. Catfish culture system

Disease infestation is responsible for the high mortality in artisanal catfish culture which may have resulted from high feeding rates, intensive stocking and poor water management. Improvement of existing practices of fry production and separation of nursery and grow-out activities. The traditional use artificial feed to raise fry attained a survival rate of 20–30%; whereas, the use of natural food produced through application of poultry manure resulted in better growth and higher survival of fry at 50%. Using large fingerlings in grow-out operation, it was possible to reduce the mortality rate from 60–90% to only 30% and achieve a yield of 37.5 t/ha in three months. This yield is comparable to that obtained by farmers in ponds with continuous water flow.

A mass selection technique was employed to determine selection response and realized heritability in *Clarias macrocephalus*. The selected line was 7.35% heavier and 3% longer than the control line after two generations of mass selection, the estimated realized heritability being 0.24 by weight and 0.28 by length.

Fed with isocaloric diets of five different protein levels for 14 weeks, the fry (7 g) of *Pangasius sutchi* showed the best growth and conversion rate at 28% and 32% protein levels.

#### 4. Trichogaster culture system

By the simple innovation of applying poultry manure over a period of two months at 156 kg/ha every ten days, it was possible to more than double the yield of *Trichogaster macrocephalus* in disused paddy fields. The maximum production of 850–930 kg/ha in eight months under artisanal practice was raised to 2 200 kg by this means.

To further upgrade the culture system, fry were raised in nursery ponds constructed within the paddy field up to fingerling stage for direct release into the paddy field for grow out. Fingerlings raised in nursery ponds fertilized with poultry manure showed better growth and survival than those raised in ponds treated with inorganic fertilizers.

Broodstock maintenance was another innovation introduced in *Trichogaster* culture. By feeding the brood fish with a specially formulated feed for one month before stocking, it was possible to reduce spawning period from two months to ten days, and get marketable fish of uniform size in six rather than eight months. Production was also 50% higher.

The cost benefit aspects of traditional and improved methods of *Trichogaster* farming were calculated and published.

Culture of *Trichogaster pectoralis* was taken up in the unutilized humic acid soil areas in southern Thailand, using the rearing method successfully practised in the central region of the country. Adult fish from elsewhere were released into ponds, where they spawned within a few days and the juveniles grew normally. However, the pH level was checked regularly and corrected when necessary, using lime and green manure. In subsequent trials, fingerlings bred in separate breeding ponds were stocked in grow-out ponds so that this method could be compared with the traditional method. The work shows that this type of soil can be used for *Trichogaster* culture without removing the rich surface soil and without disturbing the underground acid soil.

#### 5. Pen culture of carp and tilapia

With a view to utilizing extensive inland water resources, a culture trial was conducted in a reservoir using carps and tilapia in four pens, each of 200 m<sup>2</sup>. *Tilapia nilotica*, rohu, bighead carp and grass carp were stocked in a ratio 6:2:2:5 and fed with a commercial pellet feed in the morning at 1.5–2% body weight/day, and with water hyacinth, chopped and ground with rice bran, at 2.5% body weight. The yield obtained after six months of culture was 1.45 kg/m<sup>2</sup>, which compares favourably with fish production in pond culture. The overall survival rate was 64%, with bighead carp showing the highest (97%) and grass carp the lowest (12%). Apart from indicating the potential of such waters for fish culture, the experiments also showed that water hyacinth, an obnoxious weed, could be used as a supplementary feed.

#### 6. Paddy-cum-prawn farming

Preliminary studies showed that paddy fields, if properly prepared and if the trenches retain water for about three months, can be used for prawn culture, both for nursing postlarvae and grow-out.

## 7. Genetic studies

Genetic studies on tilapia showed that the Thai Chitralada tilapia strain was superior to Israel strains and to the cross between the Thai and Israel strains. Branding had no adverse effect on the growth and survival of tilapia, and the brand was visible for six months.

To improve the commercial production of *Crassostrea lugubris*, triploid oysters were produced using three different methods of genetic manipulation, i.e., temperature shock, hydrostatic pressure and Cytochalasin B treatment, of which the last proved to be most effective, resulting in 50–70% triploid induction. The growth, survival, glycogen content and level of sexual maturity of the triploid oysters were evaluated.

With a similar objective of improving the commercial production of the Java carp, *Puntius gonionotus*, all-female gynogenetic diploids were produced by using UV-treated sperms and temperature shocks. The female was preferred for culture because of its faster growth.

Weight-specific selection technique was employed to study the growth of the Thai strain of red tilapia. Weight and length of the selected line were found to be greater than those of the control in every generation. The selected line was 18.7% heavier and 8.57% longer than the control line after five generations. The realized heritability was estimated to be 0.17 by weight and 0.29 by length.

## 8. Fish hormone studies

Crude HCG, produced from the urine of pregnant women through acetone precipitation, was successfully used for the induced spawning of all types of catfish including *Clarias* spp.

A modified method of extraction and purification of HCG from urine was developed, in which the hormone was extracted with ammonium hydroxide and the precipitate dried with ether. HCG prepared using this method was found to be more effective in induced spawning of *Clarias macrocephalus* than the crude HCG extracted through acetone precipitation.

In comparative trials with fresh and acetone-preserved Chinese carp pituitary glands for induced spawning of *Puntius gonionotus*, the efficiency of the hormone was found to decrease with the length of preservation, which necessitated administration of higher doses of acetone preserved glands than of fresh glands.

## 9. Diseases

The RLCT actively participated in a regional research programme on the much feared epizootic ulcerative syndrome disease, which affects a wide variety of fish in Southeast Asia.

A new pathogenic bacterium similar to *Edwardsiella ictaluri* was isolated from haemorrhagic septicemia of skin and internal organs in *Clarias batrachus*. The morphology, physiology and biochemistry of another bacterium, *Streptococcus* sp., infecting sand goby (*Oxyeleotris marmoratus*) was studied and described.

The luminescent disease in giant freshwater prawn postlarvae was found to be caused by *Vibrio harveyi*, a luminescent bacterium. It was sensitive to chloramphenicol, erythromycin, gentamicin, novobiocin, trimethoprim, furazolidone, nitrofurantoin and sulfamethoxazole, but resistant to ampicillin and penicillin. Furazolidone and nitrofurantoin were found to be effective chemotherapeutics.

Intensive research was undertaken on the diseases of snakehead, catfish, seabass and grouper.

In studies on the relationship between giant freshwater prawn diseases and water quality in nursery ponds, it was found that the pH and unionized ammonia content of water were significantly higher in diseased ponds than in normal ponds.

Histopathological studies were carried out on shell disease, idiopathic muscle necrosis (IMN) and red discoloration disease of *Macrobrachium rosenbergii*. A progressive necrosis, inflammation and subsequent melanization of the cuticle were observed in shell diseased prawn, while a variety of bacteria were isolated from the muscle of prawn affected by red discoloration disease. The IMN disease was found to affect mostly postlarvae and juveniles. Microscopically it showed varying degrees of myofibrillar necrosis, haemocyte infiltration, calcification and sarcolemmal nuclear proliferation.

#### 10. Nutritional studies

Because of the seasonal abundance of trash fish, the traditional feed used in catfish and snakehead culture, the feasibility of preserving it by fermentation and using it as a feed round the year and in inland areas was investigated. It was found that a fermented feed mixture of trash fish and rice bran (70:30) with 5% salt, plus vitamin B complex and vitamin C, could keep well for several months without loss of nutritional value, and *Clarias* fry grew better than when fed with fresh trash fish.

Eight-day old *Puntius gonionotus* fry fed with minced water hyacinth and rice bran in various ratios, showed rates of survival mostly above 66%, the highest (96.25%) being with the 50:50 mixture. This work indicated the suitability of water hyacinth in diets for nursing fish.

In feeding trials with *Macrobrachium rosenbergii*, soybean meal was found to be a superior replacement for fish meal. The optimal level of soybean in the feed was found to be 29%.

Of the four different diets tried for *Clarias* fingerlings, the moist pellet (NIFI No. 12) appeared to be better than the sinking pellet, the floating pellet and trash fish-rice bran feed in the ratio 8:2.

In experimental studies to assess the n-3 HUFA requirements of seabass juveniles, it was found that this essential fatty acid (EFA) had a fundamental role in seabass nutrition, the dietary requirement being 1.8% in dry weight for good growth, high feed efficiency and freedom from EFA deficiency.

Vitamins B<sub>1</sub>, B<sub>2</sub>, B<sub>6</sub>, pantothenic acid and vitamin C were found to be essential for the health and growth of seabass fry. A deficiency in these led to loss of equilibrium, eroded fin, anorexia, etc. Seabass fingerlings were found to be more sensitive to a deficiency in vitamin C than in other vitamins, the vitamin C requirement being 0.5–1.0%.

#### 11. Breeding and rearing of short-necked clam, *Paphia undulata*

Short-necked clam was successfully induced to spawn and the larvae produced were reared in the hatchery up to planting size, with an average shell length of 1.0–1.3 cm. Temperature cycling, drying/immersion and Serotonin injection were the three methods used to induce spawning. *Isochrysis galbana*, *Thalassiosira pseudonana* and *Chaetoceros calcitrans* were found to be suitable food for the larvae. The average survival rate of larvae from D-shaped stage to 30 days old was 28.4%.

#### 4. The Regional Lead Centre in the Philippines (RLCP)

##### 1. Tiger shrimp (*Penaeus monodon*) culture

###### a. Larval-rearing techniques

Larval-rearing techniques practised in Japan, Taiwan, the United States, Thailand and the Philippines were tested, evaluated and refined, with the aim of improving, standardizing and packaging such techniques.

In feeding trials, no significant difference was observed in the growth and survival of postlarvae (PL5-PL 35) when fed on mussel meat and a commercial pellet feed with 40 % protein. Since the commercial feed was cheaper and readily available, it was tested further for its efficiency. The survival rate of juveniles with this feed was 33.4%, the length and weight attained being 45.65 mm and 0.83 g.

The use of an artificial diet for rearing the larvae from N<sub>1</sub> to P<sub>1</sub> stage gave very promising results with a survival rate of 65%, the moulting periods being normal. The larvae attained P<sub>1</sub> stage within nine days.

In further feeding trials with dry *Acetes*, *Chaetoceros sp.* and *Skeletonema sp.* L type under different climatic conditions and hatchery systems, the survival rates of larvae fed with dry *Acetes* and *Skeletonema sp.* from nauplii to postlarvae were comparable and fairly high at 62% in outdoor hatchery tanks. All three feeds gave a lower survival rate of only 51% in indoor tanks, but larvae fed on dry *Acetes* were seen to moult to postlarval stage about three or four days later than those fed with the other two feeds. There was no significant difference in the survival and growth of larvae between the summer and rainy seasons.

In another experiment on the nursing of postlarvae (P<sub>15</sub>-P<sub>60</sub>) in fertilized earthen ponds of 600 m<sup>2</sup>, it was found that the addition of *Artemia* nauplii at 400 g/pond three days prior to stocking led to significantly better growth, but a slightly lower rate of survival.

###### b. Culture techniques

Trials were conducted on the semi-intensive culture of tiger shrimp, aimed at raising production through manipulation of the traditional extensive culture techniques without substantially increasing initial investments. The trials were based on the assumption that the natural food in fertilized ponds could support a high density of shrimp larvae for the first two months, while supplementary feeding would then be required to provide adequate nutriment to the growing juveniles. Before stocking, the ponds were prepared in the usual manner, eliminating predators and competitors, liming and fertilizing. Stocking densities of 4, 8 and 12/m<sup>2</sup> were tried to determine the optimum. About 30% of the water was replenished daily, while the shrimps were fed with commercial pellets at 3–10% of biomass per day. Net yields ranged from 67.6 to 73.8 g/m<sup>2</sup>. Lower stocking densities gave the best results, with 90% of the harvest consisting of marketable size shrimps, at an average weight of 25 g. The 12/m<sup>2</sup> density yielded only much smaller shrimps, with an average weight of 11 g.

Another innovation tried was the transfer of shrimps to newly fertilized ponds after culturing them for 45 and 60 days. The latter proved to be better both in terms of growth and production.

The growth and survival of tiger shrimp were used to test the efficiency of several commercial feeds over a period of 120 days. The best average growth attained was 23.5 g, while the survival rates were not significantly different between different feeds.

#### c. Maturation of tiger shrimp

In order to ensure an adequate and constant supply of tiger shrimp spawners, studies were undertaken to induce maturation through eye stalk ablation in net cages fixed in ponds and in concrete tanks. The results obtained in net cages were not satisfactory. In the concrete tanks, the selected wild broodstock attained better maturation than the unselected fish. The criteria for selection were complete appendages, unbroken back, thelycum full of spermatophores and weight of female above 100 g.

### 2. Seabass (*Lates calcarifer*) culture

The seabass was induced to spawn in captivity by hormone injection in August 1983. The resultant larvae were reared successfully, and more than 1 million fingerlings were raised in four months.

#### a. Pond culture

The success in induced spawning of seabass facilitated the testing of its potential for pond culture. Studies were conducted to assess optimum stocking density, feed and feeding rates. Seabass was also cultured together with tilapia with a view to increasing production and reducing production costs.

A comparative study on hatchery-bred and wild fry, stocked at 3/m<sup>2</sup> in brackishwater ponds showed no significant difference in growth but, with a significantly higher rate of survival, the hatchery-bred fry yielded 9 t/ha as against only 3 t/ha yielded by the wild caught fry, over a period of six months.

#### b. Spawning and larval rearing of grouper (*Epinephelus tauvina*)

The grouper, *Epinephelus tauvina*, was successfully induced to spawn through hormone injection in October 1984 for the first time in the Philippines. While initially the fish had to be stripped, it was possible later on to induce natural spawning. Ovulation took place and the eggs were ready for stripping about 9–15 h after the final injection. An injection of HCG at 500 IU/kg body weight, followed after an interval of 24 h by a second injection of pituitary gland at 3 mg/kg body weight, resulted in natural spawning and the best rate of fertilization.

The larvae obtained from induced breeding were reared in 250 l capacity rearing tanks and fed with *Isochrysis*, *Tetraselmis*, *Artemia* and sea urchin eggs in different combinations. Newly hatched larvae fed with *Isochrysis* mixed with sea urchin eggs gave the best survival rate of 9%.



## **Attachment B: Training courses and workshops**

### **1. Regional Training Programme for Senior Aquaculturists**

This one-year programme was conducted seven times from 1981 to 1989 at the Regional Lead Centre in the Philippines, with parts of the programme conducted at other RLCs. It was a postgraduate programme, with the successful participants receiving a Master's Degree in Aquaculture from the University of the Philippines in the Visayas (UPV) and/or a Diploma in Aquaculture from NACA. The programme in the Philippines was organized in collaboration with the SEAFDEC Aquaculture Department and the UPV.

The programme also emphasized leadership training, as well as the establishment of links among scientists in the region. These links were to enable young scientists of the region to maintain close working relationships, especially to facilitate furtherance of regional cooperative and collaborative programmes.

The broad-based curriculum covered all aspects of aquaculture, including selection of suitable sites and species for aquaculture farms, design and construction of aquaculture facilities, production systems, breeding and rearing of cultivable organisms, relevant physico-chemical, biological and environmental aspects, post-harvest technology, socio-economic aspects, extension and planning. The programme was thus practical oriented with special emphasis on aquaculture systems having direct commercial application to the region. The programme comprised lectures and group discussions, practical classes including field surveys, laboratory work, hatchery work, farm training, seminars and workshops. Participants were required to be effective in writing field and laboratory reports, survey results, seminar and workshop proceedings and term papers.

Some parts of the programme were conducted at the RLCs in India, China and Thailand. The training in India focused on composite fish culture, evaluation of extension programmes and sewage-fed fish culture, while the focus in China was on field trials of aquaculture systems, the management of integrated farming systems and raising fish fingerlings in cages in reservoirs. The portion covered in Thailand related to the betterment of seed production of species of *Pangasius*, *Puntius* and *Clarias*, cage culture and sampling methods in reservoirs and field observation of major fish farming systems, especially those of regional importance such as *Macrobrachium*, snakehead, *Clarias spp.*, *Trichogaster spp.*, *Lates calcarifer*, oysters and *Penaeus spp.* Apart from RLCs, Hong Kong also offered the study of fish production in marine floating cages.

In all, 137 participants from 20 countries successfully completed the programme in a series of seven courses. Funding support for the various participants was provided by NACA, the Southeast Asian Fisheries Development Centre (SEAFDEC), the Brunei Government, the Sri Lanka country project, the Indonesia country IPF, FAO/UNDP, the International Development Research Centre (IDRC), the Commonwealth Secretariat (COMSEC), the World Bank, the Sri Lanka country IPF, the Asian Development Bank (AsDB), SEAFDEC-Japanese Cooperation International Agency (JICA), and the Viet Nam country project (VIE/83/002).

### **2. Training Course on Integrated Fish Farming**

This four-man course was conducted nine times during the period 1981–89 at NACA's Regional Lead Centre in China at Wuxi. In all, 207 participants from 36 Asia-Pacific, African, Latin American and European countries successfully completed the course.

In addition to lectures, this course emphasized practical study in both the laboratory and the field, visit to integrated fish farms, group discussions and seminars. The subject areas covered were:

- biological characteristics of major species of Chinese carp, their artificial propagation, culture of fry, fingerlings and marketable fish;
- fish farm management including preparation, fertilization, feeding, disease control, etc.;
- characteristics and significance of integrated pond fish farming;
- site selection and design of integrated fish farms;
- animal and agricultural crop production in integrated fish farms; and
- planning, management and economic evaluation of integrated fish farms.

In order to enable the participants to appreciate and grasp the essentials in the planning and management of an integrated fish farm, they were required to conduct a detailed study of all aspects of the operation and management of the Holei Fish Farm, including the efficient use of manpower, space and agricultural and animal wastes, and the economics of integrated fish farming in Holei commune.

Funding support for the various participants was provided by NACA, CPR/82/002, the World Bank, IDRC, NEP/79/030, the Mekong Committee, BUR/78/006, the Netherlands Government, TCDC, KEN/80/018, AsDB, GCP/ZAM/038/NET, IND/85/020, the Canadian International Development Agency (CIDA), the Government of Iran, DRK/88/002, LAO/82/014, NEP/85/034, MAG/84/002, TCP, FAO/Haiti Project and RAS/90/002. Only one participant from Italy was self-sponsored.

### 3. Training Course on Fish Disease Diagnostics

In view of the increasing incidence of fish diseases in intensive and semi-intensive aquaculture operations in the region causing enormous economic losses, the dearth of suitably qualified specialists to handle fish disease problems, and the need to establish quarantine systems for inter-country transfer of fish, a nine-month course on Fish Disease Diagnostics (Tropical Fish Health Training Programme) was organized at the Universiti Pertanian Malaysia (UPM) in 1988–89. Held in collaboration with UPM and IDRC of Canada, the course was designed to train core persons to support the expanding development of aquaculture in the region. The UPM was chosen because of the expertise and facilities available in its Faculty of Fisheries and Marine Science.

The course was aimed at making the participants fully conversant with:

- diagnostic procedures for viral, bacterial and mycotic diseases and protozoan and helminth parasitic diseases affecting the cultured food fishes;
- mechanisms of environmental stress and environmental-induced diseases;
- methods of monitoring fish health;
- immunoprophylactic and other preventive measures; and
- methods of disease control and requirements for the establishment of quarantine facilities and systems.

The participants also received instruction in systematic fish physiology, aquaculture science, pathology, parasitology, microbiology, immunology, fish quarantine, chemotherapy and statistics. The course consisted of detailed theoretical instruction and intensive practical work, including visits to fish farms in different parts of Malaysia and Thailand or Singapore.

Apart from the UPM faculty members, several regional resource persons were involved in the instructional programme.

While all the successful participants were awarded a Certificate in Tropical Fish Health by the UPM, those who did very well and satisfied the admission requirements were accepted to the master's degree programme at the UPM.

Of the nine nominees admitted to the course, four were sponsored by NACA, four by IDRC and one by the Bangladesh Agricultural Research Council.

#### 4. Secondment Programme for In-Service Training in Research

This one-year programme, which constituted one of the TCDC activities of the project, was to enable junior scientists from regional and national centres linked to NACA, to receive in-service training at RLCs in research methods and approaches required for aquaculture technology improvement and development through team work. In addition to pursuing their own particular fields of research in a culture system, the scientists were given the opportunity to learn at first hand the technology of the whole system and the methods used by researchers in other disciplines, thus enabling them to understand the advantages of a multidisciplinary approach in system-oriented research.

Between 1985 and 1989 all the 13 junior scientists from six countries (Bangladesh, China, Indonesia, Malaysia, the Philippines and Sri Lanka) successfully completed their research attachments in *Macrobrachium* culture, carp culture and *penaeid* shrimp culture as follows: RLCT (7), RLCI (4) and RLCP (2), respectively. Funding was provided under FAO's Government Cooperative Programme, GCP/INT/423/CAN.

#### 5. Short training courses

Aquaculture technologies used in selected aquafarming systems were improved by ways of interdisciplinary research. Technologies developed at the RLCs and elsewhere were packaged and transferred through short training courses to various countries in the region for adaptation and modification by national aquaculture institutions and centres, and subsequently to farmers through extension and training at the national level. These courses varied in length from three to ten weeks. A total of 180 trainees from 27 countries participated in nine such short courses in the period 1985–89.

##### a. Training Course on Small-Scale Shrimp Hatchery/Nursery Operation and Management

This course was conducted at the RLCP from 2 September to 30 October 1985. From ten countries, 19 trainees were in attendance: 11 sponsored by NACA, two Nigerians by their Government, and six by private sources.

Instruction was given on shrimp hatchery practices in Asia, the reproductive biology of *penaeid* shrimps, species and site selection, the cultivation of live feed organisms, shrimp maturation, larval rearing, disease control, broodstock development, formulated feeds, production economics and hatchery design and construction. There were practical exercises in most of these subject areas.

This course was repeated from 4 August to 28 September 1986 with 13 participants from eight countries, all sponsored by NACA.

##### b. Training Course on *Macrobrachium* Seed Production

Sixteen participants from nine countries attended this RLCT course from 25 February to 4 April 1986. Participants were all sponsored by NACA. The training course consisted of lectures, practical work and field trips.

#### c. Training Course on Composite Carp Culture

This ten-week RLCT course was held from 1 September to 15 November 1986 with 21 trainees from ten countries sponsored by NACA.

#### d. Training Course on Marine Finfish Cage Culture

A total of 21 participants from nine countries attended this six-week training course, conducted in Singapore from 1 September to 11 October 1986. All participants were sponsored by NACA. This was the only NACA-sponsored short training course organized in a non-member country.

#### e. Training Course on Mass Seed Production of Chinese Carps

This five-and-a-half week course was held from 20 April to 27 May 1987 in China with 16 participants from seven countries. Ten of the participants were sponsored by IDRC, the remaining six by NACA. The course was conducted partly at the RLCT in Wuxi and partly at the Pearl River Fisheries Research Institute in Guangzhou. The course objective was to enable the participants to acquire adequate theoretical and practical knowledge of mass production of Chinese carp fry and fingerlings for stocking purposes, including the biological basis of breeding of cultivated carps, culture of brood fish, development of sex glands, artificial breeding technology, rearing of fry and fingerlings, seed transport and disease control.

#### f. Training Course on Aquaculture Economics and Socio-Economics

This course was to train aquaculturists, and to conduct social and economic research analysis of aquaculture production. It included micro-and macro-economics, covering both production and marketing aspects, as well as social interactions. The course was jointly conducted by the RLCT and the Kasetsart University in Bangkok from 7 May to 5 June 1987. The 22 participants, from 11 countries, were all sponsored by NACA.

#### g. Training Course on Environmental Monitoring and Ulcerative Syndrome in Fish

This four-week course (3–30 November 1987) was organized by the RLCT. All the 18 participants from 11 countries were sponsored by NACA. This prepared participants in research on the relationships between environment and ulcerative disease syndrome in fish, with emphasis to upgrade the participants' technical skill in water chemistry and analysis, design of sampling programmes, identify common fish pathogens and symptoms, and interpret the relationships between environmental factors and fish disease occurrence. All the participants had subsequently initiated a regional research programme on the subject.

#### h. Training Course on Backyard Shrimp Hatchery Operation and Management

Six sessions on this highly intensive on-the-job training were conducted between April and November 1988 by the Department of Fisheries of Thailand through its Brackishwater Aquaculture Division. The first five sessions were held over 26 days and the last over 23 days. Each session had two trainees, except the last, which had four. All 14, who came from eight countries of the region, were sponsored by NACA.

#### i. Training Course on Aquaculture for Economists

Twenty participants from 15 countries of the region took part in this three-week course held at the RLCT in November 1989. All the trainees were sponsored by NACA.

## 6. Workshops

Two workshops were organized by NACA between 1986 and 1989, both in Bangkok. The breakdown of participants by country is given in Appendix 2.

### Workshop on Socio-Economics of Aquaculture Development

This nine-day workshop, organized at the RLCT in October 1986, was attended by ten participants from nine countries of the region. All the participants were sponsored by NACA.

### Workshop on Ulcerative Disease Syndrome in Fish and the Environment

In connection with the ongoing regional research programme on ulcerative disease syndrome in fish and its relationship with the environment, a five-day workshop was held at the RLCT in Bangkok in March 1989 to analyse and review research results and plan the continuation of the programme. The workshop was attended by 18 participants from 11 countries. All the participants were sponsored by NACA. The workshop provided valuable and interesting data and helped in further refinement of the research programme.

### Regional Workshop on Fish Health Management

As part of an AsDB-funded project, an eight-day regional workshop was held at Pusan, Republic of Korea from 8 to 15 October 1990, attended by 35 participants from 17 countries of the region, in addition to six regional and ten international resource persons and 20 representatives of ten international and regional organizations and agencies. Hosted by the Government of the Republic of Korea, it marked an important milestone in the area of fish health management in the Asia and the Pacific Region. In addition to gather relevant data on the existing situation relating to incidence and magnitude of diseases, economic losses thereof, availability and requirement of infrastructure facilities and manpower, role of environment and other factors in the incidence of fish diseases, the deliberations of the workshop helped in identifying future courses of action to ensure viable fish health management in the region.