# Pond Fertilization
# Algal Bioassay Testing
# Workshop

Presented in
Bangladesh, Cambodia, Laos, Nepal, Thailand, and Vietnam
June/July 2002

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Pond Dynamics/Aquaculture Collaborative Research Support Program
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# READ ME FIRST

Greetings:

During June-July 2002, the Pond Dynamics/Aquaculture Collaborative Research Support Program (PD/A CRSP) sponsored 3-day workshops on the ecology of pond fertilization, and the application and use of the Pond Fertilization Algal Bioassay Test Kit. These workshops were presented by Dr. Christopher F. Knud-Hansen (Dept. of Fisheries and Wildlife, Michigan State University, USA) at the following locations:

- Aquaculture and Aquatic Resources Management (AARM) program, Asian Institute of Technology, Pathumthani, Thailand (31 May - 3 June, 25 June 2002).
- Udornthani College of Agriculture and Technology (UCAT), Udornthani, Thailand (5-7 June 2002).
- Regional Development Coordination for Livestock and Fisheries Development in Southern Laos (RDC), Savannakhet, Laos (10-12 June 2002).
- Faculty of Fisheries at the University of Agriculture and Forestry (UAF), Ho Chi Minh City, Vietnam (17-19 June 2002).
- School of Agriculture Prek Leap (SAPL), Phnom Penh, Cambodia (20-22 June, 2002).
- Regional Aquaculture Center No. 1 (RIA-1), Dinh Bang, Tuson, Bacninh, Vietnam (27-29 June, 2002).
- Fisheries and Aquaculture Department, Institute of Agriculture and Animal Science (IAAS), Tribhuvan University, Rampur Campus, Chitwan, Nepal (3-5 July 2002).
- Department of Fisheries Management, Bangladesh Agricultural University (BAU), Mymensingh, Bangladesh (9-11 July 2002).

The main focus of the workshop was the practical application and understanding of the Algal Bioassay Test Kit (kit) for identifying pond- and time-specific fertilization requirements. Some components of the kit were brought from the USA, while other components were purchased in Thailand. A complete list of kit's components is on page 2 of the kit's user manual, a copy of which is included in a side pocket of this workshop package. The kits provided concentrated solutions of nitrogen (N), phosphorus (P), and carbon (C) for algal nutrient spikes to be put in pond water collected in 300-500 ml clear plastic drinking water bottles. These bottles were readily available at each workshop site. Each pond required 8 bottles for the test - 7 bottles for spikes (N, P, C, N+P, N+C, P+C, N+P+C) and 1 bottle for a non-spiked control. After 2-3 days incubation under indirect sunlight, relative algal growth in the bottles was examined by visual comparison of filters (a mechanical filtering apparatus was provided in the kit) and by simple visual comparison of water color in the bottles following mixing (which was easier and more reliable than filtering). Fertilization recommendations were based on whether a nutrient was found to be primarily limiting, secondarily limiting, or did not limit algal productivity at all. Fifteen kits were left at each workshop site. All workshop presentations were given from a laptop computer using PowerPoint software.

The general format for each workshop was as follows:

Day 1 (am): Introduction: opening remarks and introductions, workshop overview, use of the algal bioassay test kits, sampling on-site ponds (5-12 different ponds) and beginning algal bioassay analyses on the ponds.
Day 1 (pm): 1st technical presentation: Pond Ecology
Day 2 (am): 2nd technical presentation: Choosing Fertilizers - focusing on the economics and ecology of fertilization. Beginning with the 2nd workshop, we had a field trip to local farms to conduct additional algal bioassay tests. Also beginning with the second workshop, we developed a spreadsheet to calculate and compare fertilizer costs.
Day 2 (pm): 3rd technical presentation: Pond Characteristics and Structures which Affect Pond Fertilization Decisions.
Day 3 (am): End algal bioassays that started on days 1 and 2. 4th technical presentation: Fertilization Methods - a comparison and analysis of methods used to determine fertilization rates. The workshop ended around lunchtime after an overview/ summary of the workshop objectives/conclusions, and the presentation of workshop certificates to participants.

Participants at all the workshops requested a printed version of the Introduction and four technical presentations. The difficulty with this request, however, was that the workshop materials, format, and technical content were constantly being revised with each presentation. Slides were constantly being modified, particularly those that presented more technical concepts. Digital photos taken at the workshops were also added and incorporated into the presentations. My hope and plan was to produce a final workshop presentation by August 2002 to send back to host country workshop coordinators and participants.

There are a number of reasons why it has taken much longer to finish the workshop presentations than anticipated. The primary reason was a need to first finish a manuscript on field trials where algal bioassays were used to identify pond fertilization requirements. This paper, titled "A comparative analysis of the fixed-input, computer modeling, and algal bioassay approaches for identifying pond fertilization requirements for semi-intensive aquaculture" (by C.F. Knud-Hansen, Kevin D. Hopkins, and Hans Guttman), was recently published in the international journal Aquaculture in 2003 (vol. 228, pp. 189-214), and a reprint is now included in this workshop folder.

Although lessons learned through the workshops were incorporated into the paper, lessons from the experimental field trials conducted at the Asian Institute of Technology (AIT, Thailand) discussed in the paper are also now incorporated into the final workshop presentation. The two main contributions are 1) a formal name for the algal bioassay approach: i.e., the **Algal Bioassay Fertilization Strategy (ABFS)**, and 2) a 5-step fertilization strategy which modifies the ABFS to include a fixed-input fertilization rate for nitrogen. Appreciating that many workshop participants do not have access to the journal

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Aquaculture, I have provided below both the Abstract and Recommended Fertilization Strategy as presented in the paper. This same information is now summarized in the workshop's 4th technical presentation on Fertilization Methods.

**Abstract (Knud-Hansen et al. 2003)**

This paper compares three different strategies/treatments for determining fertilization rates for producing natural foods in semi-intensive aquaculture ponds. The first strategy used a predetermined, fixed-input rate of nitrogen (N) and phosphorus (P) based on results from previous yield trials. The second strategy was based on algal nutrient concentrations, and used biweekly water quality measurements in combination with a microcomputer-based expert system, PONDCLASS©, to determine fertilization rates. The third approach, the algal bioassay fertilization strategy (ABFS), was based on algal growth responses to nutrient (i.e., N, P, and carbon (C)) enrichment, and used weekly, pond-specific algal bioassays to determine both nutrient requirements and associated rates of nutrient inputs. The three fertilization strategies were applied to Nile tilapia (*Oreochromis niloticus*) growout ponds over a 120-day period, with five ponds per treatment. All ponds were fertilized weekly with urea, triple superphosphate, agricultural lime, and/or chicken manure in amounts determined by each strategy.

Results indicated that net fish yields (NFY) were not significantly different ($\underline{P}$ = 0.094) between treatments, with the fixed-input treatment giving the highest but most variable yields. Average NFYs ± s.e. for the 120 day growout period were 2,124 ± 276 kg $ha^{-1}$, 1,476 ± 151 kg $ha^{-1}$, and 1,651 ± 133 kg $ha^{-1}$ for the fixed-input strategy, PONDCLASS©, and ABFS treatments, respectively. The relatively lower NFYs for PONDCLASS© and ABFS indicate that neither approach maximized fish production.

Nitrogen utilization efficiencies of fertilizer inputs were similar for all three strategies. Although the fixed-input approach used approximately 20% more N than the other two approaches, mean algal productivities and NFYs were also proportionally higher with this treatment. This result is consistent with the observation that algal productivities in PONDCLASS© and ABFS ponds were nearly always limited by N availability.

However, both P utilization and fertilization cost efficiencies were significantly better with PONDCLASS© and ABFS than with the fixed-input treatment. The fixed-input approach not only used a higher P input rate than necessary, it did not account for ecological differences between ponds within the same treatment (e.g., nutrient and light limitation of algal productivity, inorganic turbidity, etc.), which can affect a pond's response to fertilization. In particular, the fixed-input treatment did not add carbon to compensate for non-uniform losses in alkalinity, which resulted in relatively high soluble P concentrations in treatment ponds where C availability apparently limited algal productivity. Including C fertilization in the fixed-input treatment would have likely reduced NFY variability and improved P utilization efficiency in those ponds.

Because both PONDCLASS© and the ABFS adjusted pond-specific fertilization requirements throughout the study, they provided increased fertilization efficiencies and profitability over the fixed-input strategy. However, the ABFS is more practical than PONDCLASS© for rural application because it is far simpler and does not require water chemistry, computers, laboratory equipment, technical expertise, or electricity to implement. Based on this study, the recommended fertilization strategy designed to

achieve cost-efficient, consistently high yields is a modified ABFS approach which uses a fixed-input fertilization rate for N, and algal bioassays to determine time- and pond-specific fertilization requirements for P and C.

**Recommended Fertilization Strategy: (Knud-Hansen et al. 2003)**

To effectively and efficiently produce natural foods through the stimulation of algal productivity, a recommended fertilization strategy must account for both pond- and time-specific algal growth limitations of light, N, P, and C. Furthermore, the approach must be easy to understand and simple to apply. The latter requirement eliminates computer models such as PONDCLASS© as practical tools for determining pond fertilization requirements for semi-intensive aquaculture. Subsequent modifications to PONDCLASS© may improve its simulation of ecological relationships and produce greater yields, but the technical requirements will remain. Results from this study, however, indicate that a modification of the ABFS to include a fixed-input component satisfies all of the above requirements.

The ABFS is not a rigid approach, but refers to a strategy where pond- and time-specific algal bioassays are conducted to identify one or more algal fertilization requirements. The ABFS used in this study was effective at identifying nutrient/light limitations and producing more consistent yields, but the recommended N inputs were apparently too low for maximizing yields. Compared to the ABFS, the fixed-input approach added about 20% more N and produced about 20% greater NFYs without any comparative loss of N fertilization efficiency. But because the fixed-input approach over-fertilized with P and did not recognize pond-specific C or light limitations, the yields were considerably more variable with less predictability or economic efficiency. Therefore, the recommended fertilization strategy is a hybrid of the fixed-input and ABFS approaches, and incorporates the benefits of both.

1. N fertilization: fixed-input of about 30 kg N $ha^{-1}$ $week^{-1}$

This should be considered a maximum weekly input rate of available N. With sufficient light, P and C, there is a good relationship between N inputs and algal/tilapia productivity. A farmer need not add N at the maximum fertilization rate, but average yields should decrease proportionally with lesser rates. Beyond about 30 kg N $ha^{-1}$ $week^{-1}$, the ponds may be so green that algal self-shading promotes light limitation and N utilization efficiencies decrease. Nevertheless, N should be included in routine algal bioassays (see below) to monitor N limitation and to avoid over-fertilization if N is found to be neither primarily nor secondarily limiting.

2. P fertilization: variable inputs based on algal bioassays

P limitation was not observed frequently in this study; but when it does exist, P fertilization is essential to maintain high algal and fish yields. A maximum P fertilization rate of about 10 kg $ha^{-1}$ $week^{-1}$ of available P should satisfy short-term P limitations of algal productivity. This rate may be increased to about 15 kg $ha^{-1}$ $week^{-1}$ if the earthen pond is new, or lowered if the pond is more "experienced" and has sediments more saturated with P. Routine algal bioassays will indicate when a particular pond does or does not need additional P inputs.

3. <u>C fertilization: variable inputs based on algal bioassays</u>
In addition to N and P, inorganic C fertilization may be necessary to achieve high productivities if ponds are rain-fed, are built on acid-sulphate soils, have a large population of clams or other mollusks, or have low alkalinities (i.e., below about 75-100 mg $CaCO_3$ $L^{-1}$) for any other reason. The maximum recommended C fertilization rate is about 500 kg agricultural lime ($CaCO_3$) $ha^{-1}$ $week^{-1}$. This is an amount typically used for satisfying pond lime requirements prior to filling, and appeared sufficient to satisfy C fertilization requirements when C limitation was indicated in this study. Animal manures release $CO_2$ upon decomposition, and can also be used to help satisfy algal C requirements if applied in amounts moderate enough not to exert a deleterious biochemical oxygen demand. Routine algal bioassays will reveal if/when the occasional lime/organic supplement should be added to maintain high yields and algal nutrient utilization efficiencies.

4. <u>Light:</u>
For the modified ABFS approach to produce consistently high yields as efficiently as possible, inorganic turbidity in ponds must be minimized. For example, a 1 m deep earthen pond stocked with common carp (*Cyprinus carpio*) will likely never turn green because these fish stir up bottom sediments which block light for algal photosynthesis and growth. Without either removing the carp or making the pond deeper, no fertilization strategy will overcome the light limitation induced by resuspended inorganic turbidity. Adding rice straw to the pond's bottom and stabilizing pond banks with vegetation can also reduce inorganic turbidity caused by storm water runoff. If a pond's source water has high inorganic turbidity, then much of the suspended clays may settle out as the pond becomes more productive. If manures are used as fertilizers, however, green manures and animal manures from ruminants (e.g., buffalos and cows) should be used with caution because tannins and other dissolved organic compounds released from the previously-consumed vegetation will add a dark color to the water and reduce light availability to algae.

5. <u>Records:</u>
Keeping good, pond-specific fertilization records is the final component of the recommended fertilization strategy. Records should include what fertilizers were used, when they were added, how much were added, pond color, and fish yields. As ponds mature with successive culture periods, pond-specific fertilization records may reveal trends of P and C (and possibly N) fertilization requirements. Ultimately, the farmer should be able to establish pond-specific fixed-input rates for N, P and C based on prior fertilization histories and observed relationships between noted inputs, pond color, and measured yields. At this point, algal bioassays would be necessary only when a pond is not visibly responding to nutrient input, or when the farmer suspects that fertilizations may be unnecessarily excessive.

In conclusion, pond fertilization recommendations typically have been institutionally derived and regionally applied. Differences between recommended fixed-input recipes often reflect their locations of origin rather than trying to account for actual ecological differences between ponds. Because differences in regionally-independent factors such as pond depth, inorganic turbidity, alkalinity, and fertilization history do affect a pond's response to fertilization, fixed-input recipes usually give highly variable results - even at the research institutions which created them. For example, the fixed-input rate used in this study was developed at the Asian Institute of Technology (AIT) in Thailand. Yet, NFY variability observed in these same AIT ponds was about two times greater with the fixed-input treatment than with the ecologically-based ABFS and computer modeling treatments.

By adopting an ecologically-based strategy, fertilization rates can be adjusted on a per pond basis while accounting for temporal changes in each pond's fertilization requirements during growout. The simple algal bioassay enables each individual pond to show the farmer what nutrient(s) its algal community need(s) - and does not need - for growth and natural food production. Research presented here supports the logic of modifying the ABFS approach to include a fixed-input rate for N, and routine algal bioassays to identify pond-specific fertilization requirements for P and C. This is particularly important for P, which is relatively expensive but recycled within older ponds more efficiently than generally appreciated. By keeping careful records, eventually farmers should be able to develop their own pond-specific fertilization rates.

The recommended fertilization strategy is applicable anywhere a farmer wishes to stimulate algal productivity for efficient natural food production. Given suitable temperatures, algae will grow as long as they have sufficient nutrients and light availability. Providing algae less nutrients than they can use unnecessarily reduces natural food production; providing algae more nutrients than they need is economically wasteful. By essentially eliminating both possibilities, the modified ABFS helps the farmer by promoting consistently high yields, greater economic efficiencies, and more sustainable, semi-intensive aquaculture production systems.

The recommended fertilization strategy should also benefit aquaculture researchers when fertilization is part of the experimental design, e.g., supplemental feed studies. Fertilizing as described above standardizes the experimental protocol on outcome rather than fertilizer inputs. Although each experimental pond may receive different amounts of fertilizers, natural food production should be high and less variable between ponds. By reducing within-treatment variability (i.e., experimental error), the benefits and costs of adding different supplemental feeds can be more accurately assessed.

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In summary, the compact disc included in this workshop folder contains the following electronic files modified from the 2002 workshops:

- ReadMeFirst.doc (Word file)
- 1.Introduction.ppt (PowerPoint file)
- 2.PondEcology.ppt (1st technical presentation: PowerPoint file)
- 3.Fertilization.ppt (2nd technical presentation: PowerPoint file)
- 4.PondCharacteristics.ppt (3rd technical presentation: PowerPoint file)
- 5.FertMethods.ppt (4th technical presentation and summary: PowerPoint file)
- Workshop.FertCalc.xls (calculates fertilizer costs, amounts and profits, Excel file)

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The fertilizer calculation Excel spreadsheet was initiated during the second workshop, and included in subsequent workshops. The spreadsheet provided here is more flexible and easier to use – both input information and results are time- and site-specific, and written directions are provided. Although very useful for comparing different fertilizer costs and identifying most economical fertilizer combinations, calculations of resulting yields and profits should be considered reasonable estimations at best.

It was an honor and a privilege to present this workshop to so many fine men and women, students and professors, institutional staff and researchers, farmers and extension workers, and good people everywhere. I hope everyone is well, and that you will find the following workshop materials (i.e., electronic copy of workshop files on a compact disc, hardcopy of these electronic files, a copy of the test kit's user manual, and a reprint of the 2003Aquaculture paper on fertilization methods) both familiar and very useful. I also hope that the 5-step ABFS as described above will be particularly beneficial to farmers. Towards that end, I recommend the further simplification of the ABFS test kit – the filtration component is not necessary, and nutrient spikes can be made from local fertilizers or other easily available chemicals. If I can assist in anyway, please do not hesitate to contact me at:

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With fond memories and kind regards to all,
Chris Knud-Hansen
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