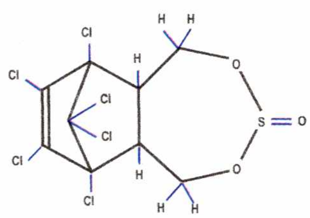
SEAFDEC Asian Aquaculture

Volume XXV Number 2 April - June 2003 ISSN 0115-4974

Endosulfan: a hidden menace

Relicardo M. Coloso, Ph.D.
Scientist II, SEAFDEC/AQD
rmcoloso@aqd.seafdec.org.ph

The use of pesticides in agriculture and human health has been successful in controlling pests and diseases. The application of the organochlorine pesticides such as DDT [1,1,1-trichloro-2-2-bis(parachlorophenyl)ethane] against the malaria mosquito and many other insect pests provided a cheap and effective control for most insect problems. The new pesticide technology also brought in other effective agents such as herbicides (for weeds), avicides (for birds), piscicides (for fish), and molluscicides (for snails) that contributed to the success of farming systems worldwide. But pesticide application has many problems such as the emergence of new pests, persistence in the environment, environmental contamination, and subsequent effect on non-target organisms including humans.



$C_9H_6Cl_6O_3S$

The chemical structure of endosulfan. *Endosulfan is highly toxic; it is either restricted, not allowed in ricefields, or banned in Southeast Asian countries. Illegal trade and incorrect use (eg. to control golden apple snail in rice paddies) always pose added danger*

Endosulfan (hexachlorohexahydromethano-2, 4, 3-benzodioxathiepine-3-oxide), a cyclodiene pesticide, also commonly known as Thiodan™ in many countries and Benzoepin in Japan, has been used effectively in the production of fruits, grains, nuts, and vegetables since 1954. New reports say that endosulfan

☞ page 2

ASEAN-SEAFDEC 5-YEAR PROGRAM
Integrated Regional Aquaculture Project

To Temburong (Brunei Darussalam) with one of IRAP's technical coordinators, Ms. Noraini

IRAP activity launched

*by **WG Yap** and **VT Sulit***

After some delay due to various reasons the latest of which was the SARS outbreak, the *Integrated Regional Aquaculture Project* (IRAP) under the ASEAN-SEAFDEC Special 5-year Program had a soft launching with the first phase of the site visitation and survey conducted from 12 to 23 May 2003.

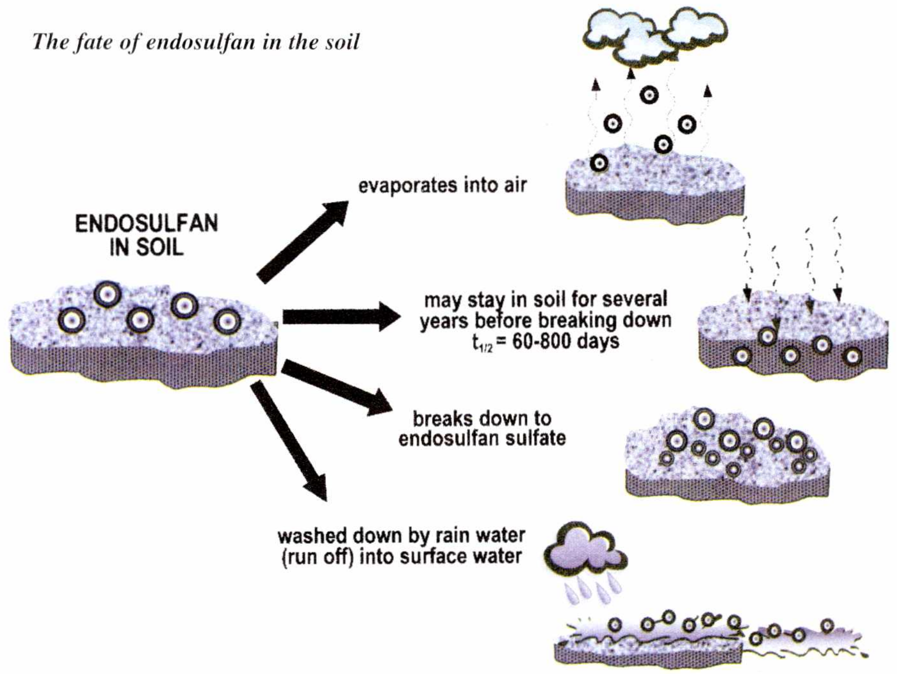
The initial countries covered were Indonesia, Brunei Darussalam and Malaysia. The SEAFDEC/AQD survey team consisted of these writers and three experts from the Thailand's Department of Fisheries (DOF) and one from AQD who joined the survey in different places depending on the expertise required. Ms. Sunee Payomjamsri, Food Technologist from the DOF Fisheries Technological Development Division came on board for Jambi, Indonesia; Mr. Sombhong Suwannatos, Senior Aquaculture Advisor of the DOF Fisheries Foreign Affairs Division for Brunei Darussalam; and for Malaysia Mr. Nareupon

☞ page 10

www.seafdec.org.ph

A publication of the Aquaculture Department of the Southeast Asian Fisheries Development Center [SEAFDEC/AQD] Tigbauan 5021, Iloilo, Philippines

The fate of endosulfan in the soil



is also used against snail pests in rice paddy ecosystems causing alarm among scientists and environmentalists regarding its impact on the surrounding aquatic ecosystem. Perhaps looking again into the use of endosulfan in enhancing agricultural production and its impact on the environment especially when it is used indiscriminately would promote a delicate balance between the demand for higher food production on one extreme and a cleaner and safer environment on the other.

Endosulfan use in agriculture

The endosulfan preparation that is commercially available is a mixture of two chemical forms, α- and β-endosulfan. It is colored cream to brown, may be crystalline or in flakes, may be an amber liquid, and smells like turpentine. The commercially available product Thiodan 3 EC contains aromatic hydrocarbons (less than 37%), 1, 2, 4-trimethylbenzene (less than 19%), surfactant blend (less than 3.4%), xylene (less than 1.8%), cumene (less than 1.2%), ethyl benzene (less than 0.6%), and 1-butanol (less than 0.3%) aside from the active ingredient endosulfan (33.7%) (*1*).

Endosulfan is used as a pesticide in vegetables such as beans, broccoli, cabbage, cauliflower, eggplant, pepper, sprouts, spinach, squash, and tomato; row crops such as cotton, potato, sweet corn, and tobacco; fruit and nut trees such as apple, apricot, blueberry, nectarine, and peach; grains such as barley, oats, rice, rye, and wheat; and vines such as grape. It is effective against a variety of insects such as aphids, army cutworm, beetles, boll weevils, leafhoppers, leafminers, tree and twig borers, stink bugs, tobacco cutworm, and others (insecticide). It is also effective against mites and ticks (acaricide) and as a wood preservative.

Endosulfan is one of the top 40 pesticides imported into the Asia and Pacific region. Its consumption (4,680 metric tons of active ingredient) in the region in 1995 was third only to DDT and monocrotophos, an organophosphorus insecticide (6,017 and 5,250 metric tons of active ingredient, respectively) (*2*). It is presently banned in Indonesia, Taiwan, and Singapore; not allowed in rice fields in Bangladesh, Indonesia, Korea, and Thailand; and restricted or severely restricted in the Philippines, Sri Lanka, and Thailand.

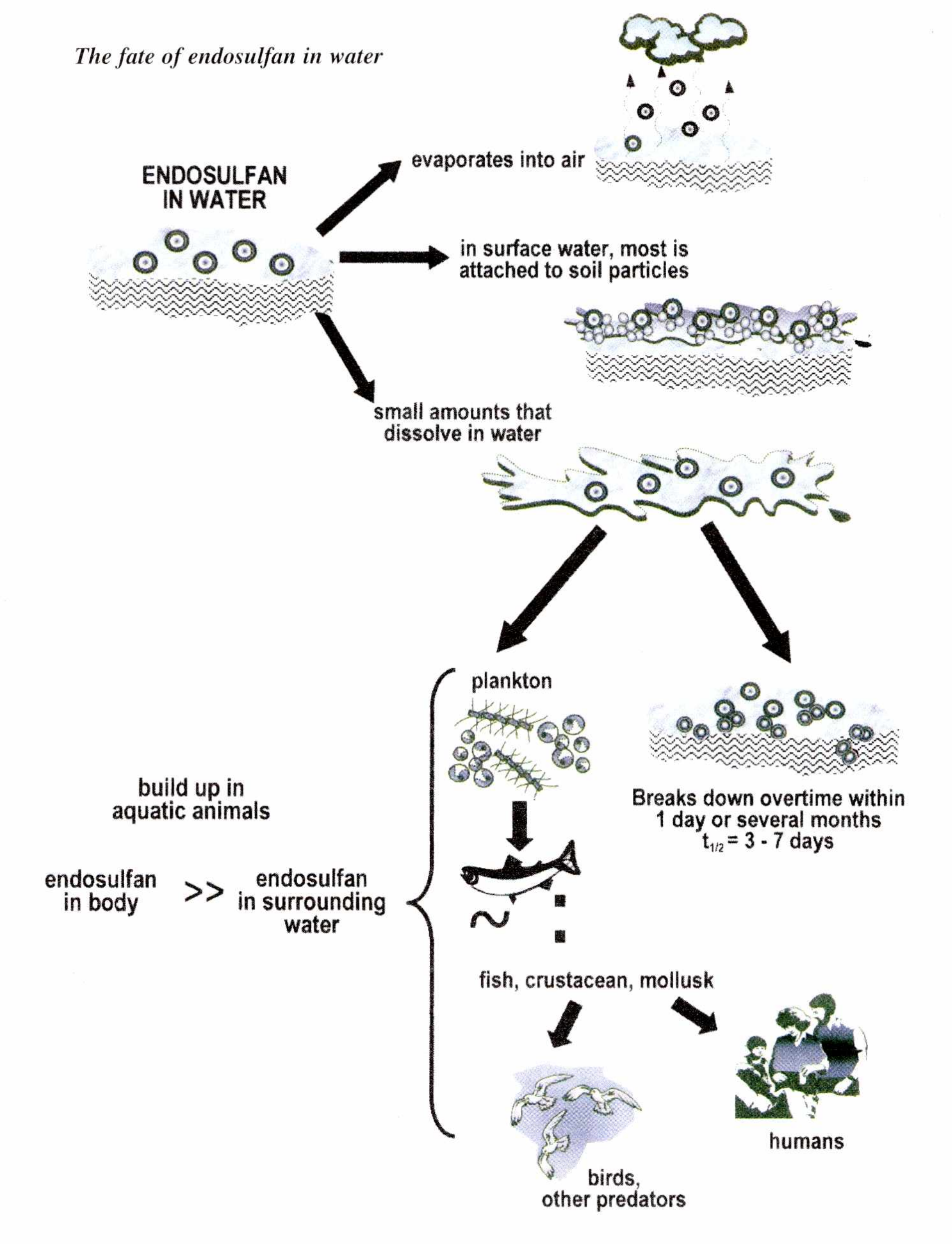
Acute toxicity on humans and wildlife

Endosulfan is highly toxic. The WHO classifies endosulfan as a moderately hazardous (category II) pesticide while the US EPA classifies it as highly hazardous (category 1b). In humans, it is highly toxic if ingested orally, inhaled, or absorbed through the skin. The FAO/WHO acceptable daily intake for endosulfan is only 0.000008 g per kg body weight (8 micrograms/kg of body weight per day). Endosulfan poisoning can result from eating contaminated food, drinking contaminated water, breathing contaminated air and touching soil and other surfaces where endosulfan has been sprayed, smoking cigarettes made from contaminated tobacco, and working extensively in an industry using endosulfan. Poisoning results in convulsions, tremors, paralysis, and vomiting, all symptoms of disorder of the central nervous system. As a class, organochlorine pesticides alter both sodium and potassium levels in neurons, thus interfering with normal neurotransmission and triggering muscles to twitch involuntarily. Furthermore, the other ingredients of the endosulfan formulation such as the aromatic hydrocarbons can produce severe pneumonitis and fatal pulmonary edema.

The toxicity of endosulfan varies according to the manner of exposure. In acute experiments, the lethal oral dose (LD_{50}) to rats is 45 milligram/kg. It is less toxic by the dermal route with a dermal LD_{50} to the rabbit of 256 milligram/kg. The lethal inhalation (LC_{50}) in the rat is 87 micrograms/liter/4 hours. However, endosulfan is highly toxic to fish and other aquatic wildlife. Endosulfan earned its notoriety for spectacular fish kills in the Rhine River in June, 1969. Endosulfan residues in the water reached 5 micrograms/liter in the German section and 0.7 microgram/liter in the Dutch section, values which were in the range of the acute lethal dose in fish of 1-10 micrograms/liter. Compounded by the high water temperature of 19 °C, fish kills occurred in both sections (*3*). Crustaceans and molluscs are less sensitive to endosulfan (acute LC_{50} value of 10-1600 microgram/liter). However, some species of shrimp such as the adult grass shrimp *Palaemonetes pugio* may be just as sensitive as fish with LC_{50} value for endosulfan of 0.62 microgram/liter. Furthermore, endosulfan may preferentially affect the male shrimp while endosulfan-exposed females may produce embryos with delayed hatching time (*4*).

Environmental fate

Endosulfan enters the environment when it is sprayed on crops or when it is accidentally spilled into the soil or water. After spraying, it could reach far distances (spray drift) before it settles on crops, soil, or water. In the soil, it is attached to soil particles especially near dump sites where the pesticide may be inadvertently disposed of. It could then evaporate into the air or stay in the soil for several years before degrading into endosulfan sulfate which is even more persistent than the parent compound. The time required for one-half of endosulfan concentration to decay (half-life or $t_{1/2}$) in sandy loam is 60-800 days. The metabolite endosulfan sulfate is highly persistent (*3*). In farms regularly sprayed with the pesticide, a significant level of endosulfan sulfate is already present in the soil prior to spraying as residues from previous applications. Lastly, endosulfan in the soil could reach surface water through runoff after a heavy rain (see figure on page 2). However, by staying bound into soil particles, the threat of endosulfan leaching into ground water is low.



Endosulfan does not readily dissolve in water. It forms an emulsion with water. In surface water, it could evaporate into the air or be attached to soil particles that are either suspended in the water column or settled in the sediment. The small amount of dissolved endosulfan breaks down overtime. The half life or $t_{1/2}$ of endosulfan in water and in fruits and vegetables is about 3-7 days. However, despite its fast degradation in water, it can still persist for a long period when bound to soil particles which later become a source of contamination. For instance, contaminated sediment can significantly change the abundance of several groups of macroinvertebrates in freshwater (*5*). Endosulfan can also build up in aquatic organisms that live in endosulfan-contaminated water, the concentration of endosulfan in the body of the organism becoming higher than its concentration in the surrounding water over time (see figure on this page). Endosulfan is rapidly accumulated and concentrated from surrounding water by freshwater green algae *Pseudokirchneriella subcapitatum* [Bioconcentration factor (BCF) of 2700 in algae exposed to 100 micrograms/liter for 16 hours] and the freshwater cladoceran *Daphia magna* (BCF of 3300 using the same exposure). Direct uptake of endosulfan from water is the major route for its bioconcentration by freshwater plankton (*6*).

Fish are a good biological indicator for pesticide pollution of aquatic ecosystems. Endosulfan residues have been reported in fish obtained from natural bodies of water near regions where there is widespread use of the pesticide for agricultural purposes. In Kolleru Lake in the Andra Pradesh region of India, extensive use of pesticides in surrounding farms have resulted in detectable levels of endosulfan in tissues of fish species like *Channa striata* and *Catla catla* as high as 77 micrograms/g (*7*). Endosulfan residues have also been detected in market fish samples in and around Calcutta, India (*8*). In Mar Chiquita coastal lagoon in Argentina, significant endosulfan and endosulfan sulfate levels were found in tissues and digestive tract of the freshwater fish silverside *Odontesthes bonariensis* indicating recent and past endosulfan use

☞ page 16

ENDOSULFAN ... from page 3

in this region (*9*). In two coastal ecosystems (Ensenada del Pabellon and Bahia de Santa Maria) in the Gulf of California, Mexico, considered to be the biggest shrimp producers in the region, residues of endosulfan and other pesticides were detected in shrimps presumably coming from surrounding farms. Pesticide use has largely been blamed for the slow growth, diverse pathologies, and low survival of shrimps in this region in recent years (*10*). Thus, the impact of endosulfan contamination of aquatic ecosytems on wildlife is presumably confined to regions with high pesticide use and does not appear to be widespread in the coastal environment. But with endosulfan having one of the highest hazard ratings among pesticides, ill effects on the living organisms in affected areas can be significant.

Chronic and sub-lethal effects

The chronic and sub-lethal effects of endosulfan contamination of aquatic biota is only beginning to become known due to recent advances in the laboratory tools used in the biomonitoring of aquatic pollutants. While lethal effects are observed immediately as toxicity, chronic effects on organs and tissues of organisms are not readily apparent except when investigated in detail after toxicity or mortality in the population has already occurred. Since the digestive tract is the first organ of aquatic organisms that comes in contact with food- or water-borne pollutants and the liver is the site for storage, biotransformation, and excretion of endosulfan, characterization of the cellular and subcellular degenerative effects is a sensitive tool for monitoring the sub-lethal effects of the pesticide. Other physiologic functions such as osmoregulation, reproduction, and molting in the case of crustaceans may also be adversely affected by sub-lethal concentrations present in the water, suspended solids, or sediment.

Degenerative changes occurring through time have been observed in the cellular and subcellular structures of the intestine and/or liver of fishes such as rainbow trout *Oncorhynchus mykiss*, carp *Cyprinus carpio*, and mosquito fish *Gambusia affinis* exposed to endosulfan (*11, 12, 13*). Some of these toxic cellular effects have been observed at endosulfan concentrations as low as 0.01 microgram/liter similar to those measured in natural water bodies. Endosulfan also promotes peroxidative damage on the lipids of the liver, kidney and the gills of the freshwater fish *Channa punctatus* Bloch exposed to 5 micrograms/liter endosulfan (*14*). In the catfish *Clarias batrachus*, sub-lethal concentrations of endosulfan reduce the specific activity of liver malate dehydrogenase, a key enzyme in energy metabolism (*15*). Endosulfan is also toxic to isolated adrenocortical cells of rainbow trout *Oncorhynchus mykiss* and possesses the potential to disrupt the normal process of adrenal steroid production in the species (*16, 17*). Endosulfan is also capable of altering the gonad structure in juvenile and adult zebrafish which presumably results from alterations in primordial germ cells of embryos (*18*).

In other vertebrates, endosulfan and other pesticides have been instrumental in the decline of several species of frogs (anuran amphibians). Reduced cholinesterase activity, a key enzyme in the central nervous system function, in tadpoles *Hyla regilla* has been observed in areas with poor populations of these amphibians as in the case of those found near vast agricultural areas in the Sierra Mountains east of Central Valley, California. Measurable endosulfan and organophosphorus pesticide residues were seen in majority of sample frog populations (*19*).

In invertebrates, exposure of the prawn *Macrobrachium malcolmsonii* to sub-lethal concentrations of endosulfan (11-32 nanograms/liter) alter the cells of the hepatopancreas and cause swelling and necrosis of the gill lamellae. These disruptions likely affect the absorption, storage, and secretion of nutrients in the hepatopancreas; respiration and osmoregulation in the gills; and growth and survival of the prawn (*20*). In the fiddler crab *Uca pugilator*, endosulfan significantly inhibits chitobiase activity in the hepatopancreas which could slow down the normal molting process (*21*). Thus, sub-lethal concentrations of endosulfan disrupt the absorption, storage, and metabolism of nutrients in fish and crustaceans. Reproductive performance in fish and other vertebrates and osmoregulation in crustaceans are also adversely affected by low levels of the pesticide.

Chronic exposure to low levels of endosulfan has been shown to affect higher animals and humans. Endosulfan treatment decreases testicular function in the rat (*22*). Residues of endosulfan are found in yolk or embryos of white leghorn hens treated with endosulfan, which suggests that exposure of the chick can occur as a consequence of maternal exposure to the pesticide (*23*). Furthermore, protein malnutrition in the rat makes it susceptible to intestinal tissue damage by endosulfan (*24*). Recent studies done in human cell lines have supported the finding that endosulfan is a xenoestrogen, an exogenous chemical that mimics the action of natural, endogenous estrogen, which can potentially disturb sex hormone action. Xenoestrogens are suspected of causing a host of human diseases including cancer. Endosulfan can act as both an estrogen agonist (mimics the function of the hormone) and an androgen antagonist (opposes the function of the hormone) and its effects can be amplified by the presence of other pesticides (*25*). It has also been shown to activate the estrogen receptor in normal and cancerous cells of the smooth muscles of the uterus (*26*) and is genotoxic to human HepG2 cells *in vitro* by causing chromosomal damage (*27*). Endosulfan can also affect human immune function by inducing cell death or apoptosis in T-cells and thymocytes (*28*). Lastly, endosulfan residues have been demonstrated in biological samples coming from people who have been exposed to the pesticide. Detectable and significant levels of endosulfan have been found in human milk (*29*), semen from normal males (*30*), cord blood samples taken after delivery (*31*) suggesting in utero exposure to pesticides, and fat samples from children living near farming areas (*32*).

Incorrect use against snail pests in rice paddies

Endosulfan has been indiscriminately used as a molluscicide to control the golden apple snail *Pomacea canaliculata* in rice paddies in the Thai provinces of Pathum Thani, Suphan Buri, Nakorn

Pathom, Nonthaburi, and Chanchoengsao, where supposedly its use is disallowed. Although endosulfan is not recommended for use as a molluscicide, majority of the 234 rice farmers surveyed found it effective against the snail pests after 1-3 applications during the pre-planting or post-planting stages. Farmers had used endosulfan for 1-5 years or more prior to the survey after favorable recommendations from the supplier and their neighbors. Spraying or splashing of the water-based emulsion was the preferred method of application. Farmers seldom drained the water from the rice paddies, but when they did, they usually drained the water 1-3 days after the first application into the next field, irrigation canal, or river. As expected, mortality in fish, snakes, and frogs were observed in the rice paddies and in irrigation canals. Those who applied the pesticide complained of headache, skin irritations, and nausea (*33*).

Although endosulfan degrades rapidly in water, significant residues in the water, soil, rice, and straw remain after application. The biological effect of such residues depends on concentration. Right after application, the concentration of the pesticide is high, which is very toxic to target and non-target organisms in the paddy. In time, the pesticide is degraded, the concentration is diminished, and the apparent effects on aquatic organisms considerably reduced. However, effects on water and soil microorganisms and plankton are usually overlooked and the sub-lethal effects on aquatic animals are difficult to determine without more detailed and extensive investigations. With the more widespread adoption of the rice-fish farming system during the growing season for rice, concern about bioconcentration of endosulfan residues in the fish and its hazard to human health is warranted.

In experiments conducted to monitor endosulfan residues in the rice paddies after application, total endosulfan residues (sum of α-, β-endosulfan and endosulfan sulfate) as high as 342 micrograms/liter was observed right after application (day 1). At day 1 after the second application (at day 10), total residues in the water were as high as 44 micrograms/liter and at day 1 after the third application (at day 20), residues were as high as 56 micrograms/liter. At day 48 after the first application, total residues in the water was 3 micrograms/liter, which was still well within the LC_{50} for fish of 1-10 micrograms/liter, but way above sub-lethal concentrations known to affect some physiologic functions in aquatic animals. Residues of endosulfan in the soil at harvest were detectable at 130 micrograms/kg. Endosulfan residues were also detected in rice and straw (1 and 20 micrograms/kg, respectively), the CODEX MRL for rice and straw being 100 micrograms/kg (*34*). How much endosulfan residue is in the fish coming from such a rice-fish farming system perhaps needs to be determined to ensure safety to human health.

While endosulfan application may be limited to the rice paddy, pesticide residues may be transported after the release of the paddy water to other aquatic ecosystems such as irrigation canals, creeks, streams, rivers and fishponds that receive their water supply from these rivers, and lakes. Pesticide residues will affect the resident aquatic organisms in these areas as well as predatory animals such as birds and even humans. Volatility of the pesticide especially under hot and humid tropical conditions and subsequent transport by air and wind is difficult to assess (*35*).

Endosulfan residues in rivers near the paddy field areas in the Thai central plain were also monitored. Several water samples were obtained from Thachin river (from Suphan Buri to Nakorn Pathom), Chaophraya river (from Pathumthani to Nonthaburi), and Bangpakong river (from Chachengsao). Highest endosulfan residues were obtained in Thachin river (0.05-0.97 microgram/liter), followed by Chaophraya river (0.05-0.29 microgram/liter), and then by Bangpakong river (0.01-0.24 microgram/liter) (*36*). Although these concentrations did not exceed the LC_{50} value of endosulfan for fish of 1-10 microgram/liter and no fish kills have so far been observed, sub-lethal effects on non-target organisms are likely to occur as a result of these measurable levels of endosulfan. Residues of endosulfan in fish, crustaceans, and other aquatic commodities for human consumption coming from these rivers should also be determined.

Summary and conclusion

A clean and healthy environment is paramount to human existence. While pesticide use has successfully sustained agricultural and food production in our lifetime as well as safeguarded human health by controlling insect pests, it has also caused many tragedies including population declines in our wildlife, fatalities in workers exposed to pesticides in its manufacture and use, and the increasing incidence of dreaded human illnesses such as cancer. A delicate balance should be achieved to mitigate the adverse impact of pesticide use to the environment and at the same time ensuring short- and long-term agricultural productivity.

Endosulfan has been effectively used as a pesticide, but much evidence on its chronic and sub-lethal effects on humans and wildlife have been gathered in recent years. More research still needs to be done to determine its effects from long-term exposure at very low levels. Endosulfan is highly toxic to fish and other aquatic animals and, thus, not recommended for use in aquatic ecosystems. However, in some countries, it has been incorrectly used as a molluscicide in rice paddies, which could have an adverse impact on the rice-fish farming systems and on other surrounding aquatic ecosystems. It is clear that such practices should be stopped and users must strictly observe the recommended application methods.

Agricultural productivity should be achieved with much less pesticide by using integrated pest management programs which make use of biological, cultural, and physical control agents and lower doses of safer pesticides on a need only basis. The benefits of biotechnology should also be used to develop more effective and safer products and techniques. This is a valid approach and one that will require a unified and concerted effort among suppliers and users of pesticides in order to ensure that resources are used to our best advantage with minimal risk.

Acknowledgment

The author is grateful to Nelson Golez for the illustrations, Ma. Teresa de Castro-Mallare for critical comments, and Marilyn Surtida for technical editing of the manuscript.

☞ next page for reference list

References

1 Material Safety Data Sheet (MSDS). 2001. Reference No. 115-29-7-8, FMC Corporation, Philadelphia, PA, USA

2 Chen Li G. 2002. Regional report on the production and utilization of pesticides- Summary of country reports. Production and Utilization of Pesticides in Asia and the Pacific. Asian Productivity Organization, Tokyo, Japan, p 27-73

3 McEwen FL, Stephenson GR. 1979. The Use and Significance of Pesticides in the Environment. John Wiley and Sons, Inc. New York, USA, 538 p

4 Wirth EF, Lund SA, Fulton MH, Scott GI. 2001. Determination of acute mortality in adults and sublethal embryo responses of *Palaemonetes pugio* to endosulfan and methoprene exposure. Aquat. Toxicol. 53: 9-18

5 Hose GC, Lim RP, Hyne RV, Pablo F. 2002. A pulse of endosulfan-contaminated sediment affects macroinvertebrates in artificial streams. Ecotoxicol. Environ. Saf. 51: 44-52

6 DeLorenzo ME, Taylor LA, Lund SA, Pennington PL, Strozier ED, Fulton MH. 2002. Toxicity and bioconcentration potential of the agricultural pesticide endosulfan in phytoplankton and zooplankton. Arch. Environ. Contam. Toxicol. 42: 173-181

7 Amaraneni SR, Pillala RR. 2001. Concentrations of pesticide residues in tissues of fish from Kolleru Lake in India. Environ. Toxicol. 16: 550-556

8 Kole RK, Banerjee H, Bhattacharyya A. 2001. Monitoring of market fish samples for endosulfan and hexachlorocyclohexane residues in and around Calcutta. Bull. Environ. Contam. Toxicol. 67: 554-559

9 Menone ML, Aizpun de Moreno JE, Moreno VJ, Lanfranchi AL, Metcalfe TL, Metcalfe CD. 2000. PCBs and organochlorines in tissues of silverside (*Odontesthes bonariensis*) from a coastal lagoon in Argentina. Arch. Environ. Contam. Toxicol. 38: 202-208

10 Reyes GG, Villagrana LC, Alvarez GL. 1999. Environmental conditions and pesticide pollution of two coastal ecosystems in the Gulf of California, Mexico. Ecotoxicol. Environ. Saf. 44: 280-286

11 Arnold H, Pluta H-J, Braunbeck T. 1996. Cytological alterations in the liver of rainbow trout *Oncorhynchus mykiss* after prolonged exposure to low concentrations of waterborne endosulfan. Dis. Aquat. Organ. 25: 39-52

12 Braunbeck T, Appelbaum S. 1999. Ultrastructural alterations in the liver and intestine of carp *Cyprinus carpio* induced orally by ultra-low doses of endosulfan. Dis. Aquat. Organ. 36: 183-200

13 Cengiz EI, Unlu E, Balci K. 2001. The histopathological effects of thiodan on the liver and gut of mosquitofish, *Gambusia affinis*. J. Environ. Sci. Health B. 36: 75-85

14 Pandey S, Ahmad I, Parvez S, Bin-Hafeez B, Haque R, Raisuddin S. 2001. Effect of endosulfan on antioxidants of freshwater fish *Channa punctatus* Bloch: 1. Protection against lipid peroxidation in liver by copper exposure. Arch. Environ. Contam. Toxicol. 41: 345-352

15 Mishra R, Shukla SP. 1997. Impact of endosulfan on cytoplasmic and mitochondrial liver malate dehydrogenase from the freshwater catfish (*Clarias batrachus*). Comp. Biochem. Physiol. C. Pharmacol. Toxicol. Endocrinol. 117: 7-18

16 Bisson M, Hontela A. 2002. Cytotoxic and endocrine-disrupting potential of atrazine, diazinon, endosulfan, and mancozeb in adrenocortical steroidogenic cells of rainbow trout exposed in vitro. Toxicol. Appl. Pharmacol. 180: 110-117

17 Leblond VS, Bisson M, Hontela A. 2001. Inhibition of cortisol secretion in dispersed head kidney cells of rainbow trout (*Oncorhynchus mykiss*) by endosulfan, an organochlorine pesticide. Gen. Comp. Endocrinol. 121: 48-56

18 Willey JB, Krone PH. 2001. Effects of endosulfan and nonylphenol on the primordial germ cell population in pre-larval zebrafish embryos. Aquat. Toxicol. 2001. 54: 113-123

19 Sparling DW, Fellers GM, McConnell LL. 2001. Pesticides and amphibian population declines in California, USA. Environ. Toxicol. Chem. 20: 1591-1595

20 Saravana Bhavan P, Geraldine P. 2000. Histopathology of the pancreas and gills of the prawn *Macrobrachium malcolmsonii* exposed to endosulfan. Aquatic Toxicol. 50: 331-339

21 Zou E, Fingerman M. 1999. Effects of estrogenic agents on chitobiase activity in the epidermis and hepatopancreas of the fiddler crab, *Uca pugilator*. Ecotoxicol. Environ. Saf. 42: 185-190

22 Chitra KC, Latchoumycandane C, Mathur PP. 1999. Chronic effect of endosulfan on the testicular functions of the rat. Asian J.Androl. 1: 203-206

23 Bargar TA, Scott G I, Cobb GP. 2001. Uptake and distribution of three PCB congeners and endosulfan by developing white leghorn chicken embryos (*Gallus domesticus*). Arch. Environ. Contam. Toxicol. 41: 508-514

24 Labana S, Bansal RC, Mahmood A. 2001. Effects of endosulfan on intestinal functions in protein-malnourished rat. Indian J. Exp. Biol. 39: 1017-1021

25 Raun Andersen H, Vinggaard AM, Hoj Rasmussen T, Gjermandsen IM, Cecilie-Bonefeld-Jorgensen E. 2002. Effects of currently used pesticides in assays for estrogenecity, androgenecity, and aromatase activity in vitro. Toxicol. Appl. Pharmacol. 179: 1-12

26 Hunter DS, Hodges LC, Vonier PM, Fuchs-Young R, Gottardis MM, Walker CL. 1999. Estrogen receptor activation via activation function 2 predicts agonism of xenoestrogens in normal and neoplastic cells of the uterine myometrium. Cancer Res. 59: 3090-3099

27 Lu Y, Morimoto K, Takeshita T, Takeuchi T, Saito T. 2000. Genotoxic effects of alpha-endosulfan and beta-endosulfan on human HepG2 cells. Environ. Health Perspect. 108: 559-561

28 Kannan K, Holcombe RF, Jain SK, Alvarez-Hernandez X, Chervenak R, Wolfe RE, Glass J. 2000. Evidence for the induction of apoptosis by endosulfan in a human T-cell leukemic line. Mol. Cell. Biochem. 205: 53-66

29 Campoy C, Jimenez M, Olea-Serrano MF, Moreno-Frias M, Canabate F, Olea N, Bayes R, Molina-Font JA. 2001. Analysis of organochlorine pesticides in human milk: preliminary results. Early Hum. Dev. 65: S183-190

30 Kumar R, Pant N, Srivastava SP. 2000. Chlorinated pesticides and heavy metals in human semen. Int. J. Androl. 23: 145-149

31 Cooper SP, Burau K, Sweeney A, Robison T, Smith MA, Symanski E, Colt JS, Laseter J, Zahm S. H. 2001. Prenatal exposure to pesticides: a feasibility study among migrant and seasonal farmworkers. Am. J. Ind. Med. 40: 578-585

32 Olea N, Olea-Serrano F, Lardelli-Claret P, Rivas A, Barba-Navarro A. 1999. Inadvertent exposure to xenoestrogens in children. Toxicol. Ind. Health 15: 151-158

33 Purivirojkul W, Arayarungsarit L. 2001. Survey on the use of endosulfan for golden apple snail control in farmer fields of Central Plain. Proceedings from the Rice Research Institute, Department of Agriculture, Meeting No. 5, November 21-23, Bangkok, Thailand. p 119-128

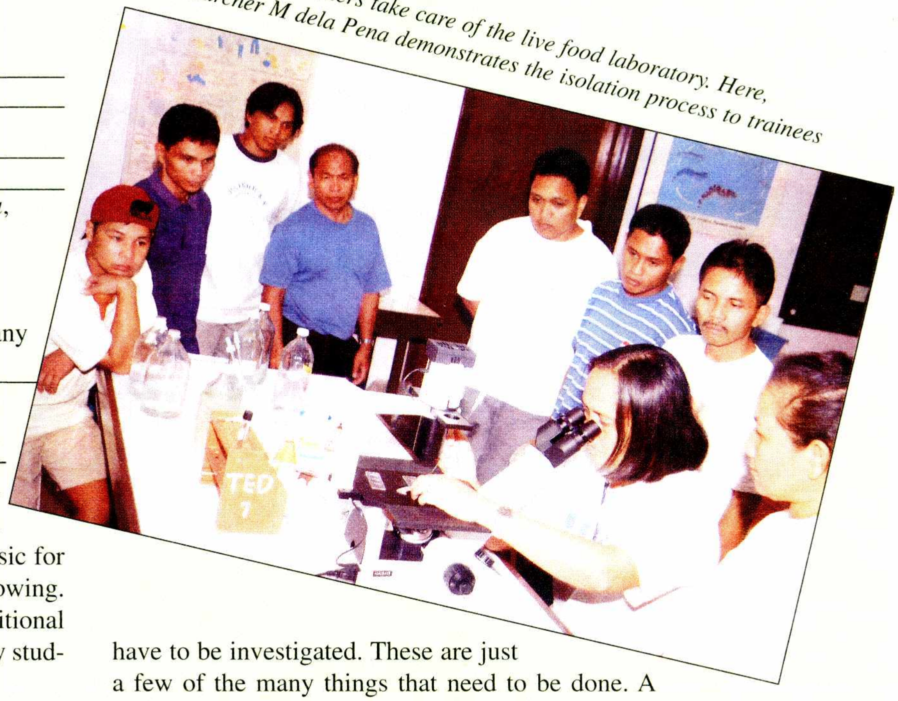
34 Suvaparp R, Chawengsri V, Tayaputch N, Chernchaivachirakul A, Yokyongsakul N, Sitchawat T, Doichuanngam K. 2001. Dissipa-

☞ page 20

Present prices of algal starters/media are:

Algae (any species)	
P100/liter aerated	
P 100/test tube unaerated in liquid or solid media	
Rotifer (*Brachionus*)	
P100/million	
Media/nutrients	
P1,170/set for F-medium - for *Isochrysis*, *Skeletonema*, and *Chaetoceros*	
P700/liter for Conway medium - for *Nannochlorum*, *Tetraselmis*, *Nannochloropsis oculata*	
P200/set for TMRL medium - for bigger cultures of any species	

At AQD, researchers take care of the live food laboratory. Here, researcher M dela Pena demonstrates the isolation process to trainees



Future activities

No doubt in the future, live food would become a new market niche along with the other profit-making production components of aquaculture. AQD has never been ready for this than now. The present microalgal collection, considered basic for any growing live food laboratory, has constantly been growing. Indeed, indigenous live food has always been used by traditional aquaculture practitioners but have never been systematically studied.

Taxonomic studies have to confirm the species studied, culture techniques have to be established for newly isolated species and strains, ecological studies such as population dynamics and determination of environmental factors as well as the nutritive value have to be investigated. These are just a few of the many things that need to be done. A bioreactor or fermentor has to be acquired and harvest and storage techniques established. Screening and extraction of bioactive compounds with nutritive and pharmaceutical values are possibilities for the live food industry. *- MBS*

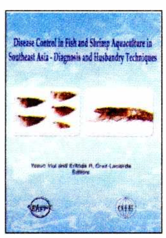
Endosulfan: a hidden menace by rm coloso

ENDOSULFAN ... from page 18

tion of endosulfan in rice fields. Proceedings from the Rice Research Institute, Department of Agriculture, Meeting No. 5, November 21-23, 2001, Bangkok, Thailand, p 129-139

35 Magallona ED. 1995. Impact of pesticides used in lowland rice production. Selected Papers on Pesticide Management and Regulation in the Philippines (ED Magallona, AC Sumalde, AC Rola, P MacQuarrie), Dalhousie University, Canada and University of the Philippines at Los Banos, Philippines, p 1-45

36 Chatsantiprapa P, Haruthaithanasan P. 2001. Distribution of endosulfan residues in the rivers along paddy field area. Proceedings from the Rice Research Institute, Department of Agriculture, Meeting No. 5, November 21-23, 2001, Bangkok, Thailand, p 141-148 ###

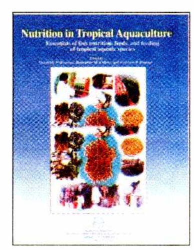


Disease Control in Fish and Shrimp Aquaculture in SEA - Diagnosis and Husbandry Techniques, 215 pages, edited by Yasuo Inui and Erlinda Cruz-Lacierda. This volume contains the 14 papers presented at a seminar-workshop held December 4-6, 2001 that was organized by SEAFDEC and the Office International des Epizooties. For a copy, contact Dr Erlinda Cruz-Lacierda at *eclacier@aqd.seafdec.org.ph* or fax (63-33) 5118709, 3351008.

Some of SEAFDEC/AQD numbers are new!

SEAFDEC/AQD trunkline
63 (33) 511 9171, 336 2965, 336 2937
Office of the Chief
(33) 511 9174, 511 9170
Training and Information
(33) 511 9172, 511 9050, 511 9049
FAX
63 (33) 5118709, 3351008, 511 9070

SEAFDEC websites on the internet
www.seafdec.org
www.seafdec.org.ph
www.mangroveweb.net
www.agrolink.moa.my/dof/seafdec



Nutrition in Tropical Aquaculture is edited by SEAFDEC/AQD scientists Dr. Oseni Millamena, Dr. Relicardo Coloso, and Dr. Felicitas Pascual. The textbook is the second title released from AQD's textbook writing program. <sales@aqd.seafdec.org.ph>