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Assessment and Reduction of Insect Infestation of Cured Fish in South East Asia, with Laboratory Studies on *Chrysomya megacephala* (Fab.), a Principal Causative Agent.

by

John R. Esser

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being a thesis presented in the candidature for the degree of Doctor of Philosophy in the University of Durham, 1988.

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ACKNOWLEDGEMENTS

I wish to thank my supervisor, Dr. Lewis Davies for his encouragement and advice throughout this study and for his help in preparing this thesis. I am very grateful to Dr. W.R.B. Arthur for providing support and departmental facilities during the laboratory investigations and to Dr. S.W. Hanson for his valuable advice.

I am indebted to the Overseas Development Administration for funding the projects that made this research possible, in particular the senior fisheries adviser, Mr. John Stoneman, for his full and consistent support throughout the study.

I am grateful to the Overseas Development and Natural Resources Institute for assisting with the pirimiphos-methyl evaluation trials in Indonesia and for performing the pirimiphos-methyl residue analyses.

I am also grateful to the Shell Research Laboratory for performing the alphacypermethrin residue analyses.

I wish also to thank the cured fish processor in Cirebon, Hadji Sonja and my Indonesian and Thai counterparts, who always responded willingly and with good humour to my seemingly eccentric requests, during the field trials.

Finally, I must thank my wife Anne, for her perseverance and patience and for proof reading the manuscript.

ABSTRACT

A survey of cured fish establishments in 5 countries of South East Asia, revealed that cured fish is a nutritionally and economically important commodity in the region and that insect infestation, in particular blowfly infestation during processing and dermestid beetle infestation during storage, are major causes of losses in cured fish. Many processors have responded by illegally applying household and agricultural insecticides to their fish.

Field investigations in Indonesia and Thailand, identified *Chrysomya megacephala* (Fab.) as the most widespread cause of infestation during processing. *Lucilia cuprina* (Wied.) was also a common cause of infestation. *Dermestes maculatus* (Degeer), *D. carnivorus* (Fab.), *D. ater* (Degeer) and *Piophila casei* (L.), were the most common causes of infestation during storage. These species were able to tolerate the relatively high salt concentrations of the processed fish.

Field infestation reduction trials, demonstrated that salting the fish for an extended period failed to provide protection against insect infestation.

Guarding the salting tank with a closely fitting lid, prevented blowfly infestation during salting. Flyscreens were found to reduce blowfly infestation during drying, but the design used, presented practical difficulties and was not acceptable to the processor.

The pyrethroid insecticide Fastac (alphacypermethrin), prevented blowfly infestation during processing at concentrations as low as 0.003% and had a marked repellent effect against blowflies at a concentration of 0.001%. Fastac, applied at a concentration of 0.006%, protected fish against dermestid beetle infestation and damage. Fastac residues in fish treated with a 0.006% dip decreased to less than 2 mg/kg after drying and 1 week´s storage.

The pyrethroid insecticide deltamethrin, prevented insect infestation during processing and storage, when applied as a 0.003% dip before drying.

The FAO/WHO approved insecticide pirimiphos-methyl, reduced blowfly infestation and prevented damage during processing and reduced dermestid beetle infestation during storage, when applied as a 0.03% dip before drying. This treatment resulted in residues, after processing, that were within the FAO/WHO maximum residue limit of 10 mg/kg.

Spray applications of pirimiphos-methyl, at dosages of 5-20 mg/kg and deltamethrin, at dosages of 1-3 mg/kg, were effective in reducing dermestid beetle infestation of smoked fish during storage.

Laboratory investigations demonstrated that *C. megacephala* produced similar numbers of male and female offspring and that there was no difference between the mortalities of the 2 sexes. Female flies greatly outnumbered male flies at the processing site.

Mean lifespans of *C. megacephala* cage populations ranged from 47-54 days and the maximum survival time ranged from 80-98 days.

C. megacephala eggs matured within 3 weeks of adult emergence and the mean egg count for the adult female flies was 221.

The presence of *C. megacephala* eggs on fish, stimulated oviposition by *C. megacephala* and freshly laid eggs were found to have a higher stimulatory effect than eggs which had been previously boiled.

Fish being salted exerted a marked, differential attractive effect on gravid, female flies.

When presented with fish of a range of salt concentrations, *C. megacephala* preferentially oviposited on the fish with the lowest salt concentration. In the absence of choice, *C. megacephala* readily oviposited on fish with relatively high salt contents of 30-40% (dwb). A feeding medium salt content of 33.8% was necessary to significantly reduce larval growth rate and salt contents in excess of 39.5% were necessary to obtain high larval mortalities. Salt contents of up to 39.5% had no effect on pupal mortality.

CONTENTS

1. INTRODUCTION AND BACKGROUND

The Food and Agriculture Organisation (FAO), has forecast that demand for fish, for human consumption, will continue to increase throughout this century, with increased demand being greatest in South East Asia. Although higher landings will account for some of the increased demand, many fish stocks are already being fully exploited and, in some cases, overexploited. A short-fall in supply, of at least 10 million tonnes+, is anticipated by the year 2000 (Whittle, 1985). This short-fall can only be met by improved utilisation of the catch. An important aspect of improved catch utilisation, is the reduction of post-harvest losses, which are thought to account for about 10% of the total world production of fish and are believed to be highest in small scale fisheries, where fish are processed by traditional methods, such as salting, drying and smoking.

Few detailed studies have been carried out into losses of cured fish during processing and storage, but losses of 25% are thought to be common and in some instances, estimates as high as 50% have been made (Poulter *et al.*, 1988). Many of these points are discussed in detail in a review of the assessment and prevention of losses in cured fish, published by FAO (FAO, 1981).

One of the major conclusions of the FAO review, was that losses of cured fish are high, particularly in small scale operations, but it was pointed out that this conclusion was based on a limited amount of data, mainly from Africa. A summary of losses of cured fish, recorded by various authors, is given in Table 1. It must be appreciated, however, that the majority of the loss figures, quoted in the table, were obtained from field trials, in which the experimental methods used, were not the same for all studies. In many cases, loss estimates are simply based on visual assessment, which, although useful, may be misleadingly inaccurate. The lack of data on losses, is particularly apparent for the South East Asian region, an

+ Approximately 14% of world annual production

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Table 1. Physical losses caused to cured fish by insect infestation in the Tropics. (Adapted from Poulter *et al.*, 1988)

Cause	Country	Type of Fish	Method of assessment	% losses	Source
Blowflies	Bangladesh	DU	E	25	Doe *et al.* (1977)
	Bangladesh	DU	E	30	Ahmed *et al.* (1978)
	Indonesia	DS	M	5-19	Esser *et al.* (1985)
	Malawi	DU	M	10-27	Meynell (1978)
	Malawi	DU	M	22	Walker and Donegan (1987)
	Sudan	DU	E	15-30	Mastaller (1981)
	The Gambia	DU	M	16	Walker and Evans (1984)
	Senegal	DU	M	34.5	Wood and Walker (1986)
Beetles	Burkina Faso	DU	E	25	Guggenheim (1980)
	Global		E	25	James (1977)
	Kenya	DU	M	1-15	Wood and Walker (1986)
	Kenya	DU	M	16	Golob *et al.* (1987)
	Malawi	DU	M	18	Walker (unpub)
	Mali	DS	M	23	Aref *et al.* (1965)
	Niger	SD	E	40	Bouare (1986)
	Nigeria	DU	E	50	Rollings and Hayward (1963)
	Nigeria	DU	M	22	Mills (unpub)
	Senegal	DU	E	20	Toury *et al.* (1970)
	Senegal	DU	E	10-30	Diouf (1980)
	Thailand	DS	M	25	Rattagool *et al.* (1988)
	The Gambia	DU	M	14	Walker and Evans (1984)
	Zambia	DU	E	10	Watanabe (1971)

E = Estimated loss DU = Dried, unsalted SD = Smoked, dried
M = Measured loss DS = Dried, salted

area of the world, where cured fish forms an important part of the diet. The total catch of the South East Asian region is about 8 million tonnes, of which approximately 30%, is preserved by curing, mainly at the small scale production level.

Fish curing, may be achieved by salting, drying and smoking, or a combination of these treatments. Extended shelf life is obtained, in all cases, by reducing the water activity (abbreviation Aw) of the product and consequently inhibiting the growth of spoilage microorganisms. Processing methods, used by different processors, vary considerably according to fish species, local tradition and taste. Large fish species are usually split, gutted and may be beheaded before salting. Small species are usually processed whole, but are sometimes split and gutted. Salting methods can be classified into three basic types:- kench curing, pickling and brining. Kench curing consists of rubbing salt into the fish flesh and placing alternate layers of fish and salt on top of each other, forming a stack. Fluids formed during salting, are allowed to drain away. Pickling differs from kenching, in that the fluids produced during salting, are not allowed to drain away and therefore remain in contact with the fish during the salting treatment. Both kench and pickle cures are commonly used in Indonesia, particularly with large species of fish. Brining simply consists of immersing the fish in concentrated salt solution and is the most widespread method of salting used in Thailand.

The salt content, of the fish after salting, varies according to fish size, species and salting method, although it rarely exceeds 30% dry weight basis (abbreviation dwb). After salting, the Aw may be further reduced by solar drying. In South East Asia, the fish are usually dried on trays or mats, either supported by racks, or placed on the ground. The drying period varies, according to fish size, species, and weather conditions. Under favourable drying conditions, small species may dry within a day and large species may take 3 to 4 days.

Smoked fish may or may not be salted, or partially dried, before being smoked by a hot or cold smoking technique. The smoking treatment further dries the fish and imparts a smoky flavour. In the case of hot smoking, the pasteurising effect, achieved by the cooking, together with the smoke acting as an inhibitor of microbial growth, help extend the shelf life of the product.

After processing, the cured fish is usually packed in bamboo baskets or boxes, before distribution or storage. Cold stores may sometimes be used for extended storage periods, but in general, the cured fish is stored, sometimes for many months, at ambient temperature and humidity, by wholesalers and retailers.

Traditional fish processing, particularly at the small scale level, is often carried out under basic, unhygienic conditions and is subject to a number of constraints, that limit the quantity and quality of fish produced by the processor. Fish availability is often erratic, making planned processed fish production impossible. Adverse weather conditions, particularly during the rainy season, disrupt drying and result in considerable physical and financial losses, caused mainly by blowfly infestation during processing and beetle infestation during storage. Insect infestation is often regarded as the major problem experienced by cured fish processors', many of whom, have resorted to applying household insecticides to their fish, in order to control the problem. Table 2 lists species of Diptera that have been found to infest cured fish and illustrates the widespread nature of the insect infestation problem in the tropics.

Table 2. Diptera which cause postharvest infestation of fish in the Tropics. (Adapted from Walker, 1988)

Family	Species	Country	Source
Calliphoridae			
	Chrysomya albiceps	Malawi	Meynell (1978)
		The Gambia	Walker and Evans (1984)
	Chrysomya bezziana	The Gambia	Walker and Evans (1984)
	Chrysomya chloropyga	Malawi	Meynell (1978)
		The Gambia	Walker and Evans (1984)
	Chrysomya megacephala	Indonesia	Esser *et al.* (1985)
		Thailand	Rattagool *et al.*(1988)
		PDR Yemen	Sachithananthan et al.(1986)
	Chrysomya regalis	Malawi	Meynell (1978)
		The Gambia	Walker and Evans (1984)
		Uganda	McLellan (1963)
		Zambia	Proctor (1972)
	Chrysomya rufifacies	India	Walker (unpub.)
	Lucilia cuprina	Indonesia	Esser *et al.* (1985)
		Malawi	Walker and Donegan (1984)
	Lucilia sericata	The Gambia	Walker and Evans (1984)
Ephydridae			
	Discomyza maculipennis	India	Soans and Adolph (1971)
Milichiidae			
	Leptometopa latipes	India	Pillai (1957)
Muscidae			
	Musca domestica	Indonesia	Indriati *et al.* (1985)
	Ophyra capensis	Malawi	Walker and Donegan (1987)
Phoridae			
	Megaselia scalaris	Malawi	Walker (unpub.)
Piophilidae			
	Piophila casei	Indonesia	Indriati *et al.* (1985)
		Malawi	Anon (1963)
Sarcophagidae			
	Parasarcophaga misera	Indonesia	Esser and Warren (1983)
	Sarcophaga nodosa	Malawi	Walker and Donegan (1987)
	Sarcophaga tibialis	Malawi	Walker and Donegan (1987)
	Wohlfartia sp.	PDR Yemen	Green (1967)

During the initial survey (see Section 2), it became apparent that large species of fish were particularly susceptible to insect infestation and that the problem was very evident in Indonesia and Thailand. It was decided to concentrate most of the research effort on reducing insect infestation of large species of fish, processed by a pickle cure followed by sun drying, because the relatively high moisture, low salt content product, that results from this process, was seen to be highly vulnerable to blowfly infestation. The majority of the field trials were conducted at the premises of a small scale cured fish processor in W. Java, Indonesia, where infestation during processing and early storage were monitored and infestation reduction methods were developed.

The major aims of the research, detailed in this thesis, were to:-

1. Assess the losses that occur in cured fish, processed at the small scale level in South East Asia.
2. Investigate the causes of losses and collect information on the biology of the insects responsible for them, together with details of their methods of attack.
3. Develop practical and safe methods of reducing these losses.

The work commenced with an initial field survey of the cured fish industry in South East Asia in 1982, which was followed by a further series of field trips to Indonesia and Thailand, between 1983 and 1988, when loss reduction trials and further survey work were carried out. During the initial survey, it was found that virtually all of the blowfly attack on salted, dried fish during processing, was due to 1 species only, namely *Chrysomya megacephala* (Fabricus) (Calliphoridae). It was therefore decided to set up and maintain cultures of this species, in the laboratory at Grimsby. The cultures, derived from specimens collected in W. Java, were essential for the laboratory, experimental work done on the oviposition behaviour of the blowflies, on the relationship between salt concentration and larval growth and mortality and on other aspects of the biology of this important species. This laboratory work, provided the essential biological basis for an understanding of the principal insect problems that occur during processing, a basis essential to designing methods of controlling blowfly infestation.

The problem of insect infestation during storage was also investigated, as this was found to be a major cause of losses during distribution, when the fish may be stored for many months at ambient temperature and humidity and exposed to attack by *Piophila casei* (L.) and *Dermestes* beetles.

2. SURVEY OF TRADITIONAL FISH PROCESSING SITES AND RETAIL OUTLETS IN SOUTH EAST ASIA.

2.1 Introduction

The initial survey was carried out in 1982, when 5 countries, Thailand, Burma, Malaysia, Indonesia and the Philippines were visited (Fig.1). Further surveys, in Indonesia and Thailand, were undertaken between 1983 and 1988.

The principal aims of the surveys, were to obtain information on traditional fish processing in the region and to identify and assess the problems faced by cured fish processors, wholesalers and retailers. At each location, special attention was given to the details of processing technique, fish species being processed, processing equipment used, situation of the premises, standards of hygiene, details of insect attack, methods used to deter insects, including insecticide use, and magnitude of financial and physical losses, resulting from insect infestation. The detailed observations are specified further in the appropriate parts of the thesis. Insect samples were collected, for identification and cured fish samples, were taken for chemical analysis at Grimsby.

2.2 Survey of cured fish establishments in Thailand

Thailand is a major producer of fish and with a marine production of 2.12 million tonnes, ranked 8th in the world in 1985. This figure represents 94% of the total fish production, with only 150,000 tonnes coming from inland fisheries. Fish forms an important part of the Thai diet and represents 60% of the animal protein supply, with an average consumption of 22.5 kg/capita/annum.

The Thailand fishery is facing a major problem, in that the maximum sustainable yield of the fish resource, is being approached, or even exceeded in many areas. Thailand's marine catch rose to 2.2 million tonnes in 1977, but since then has fluctuated between 1.8 and 2.3 million tonnes, of

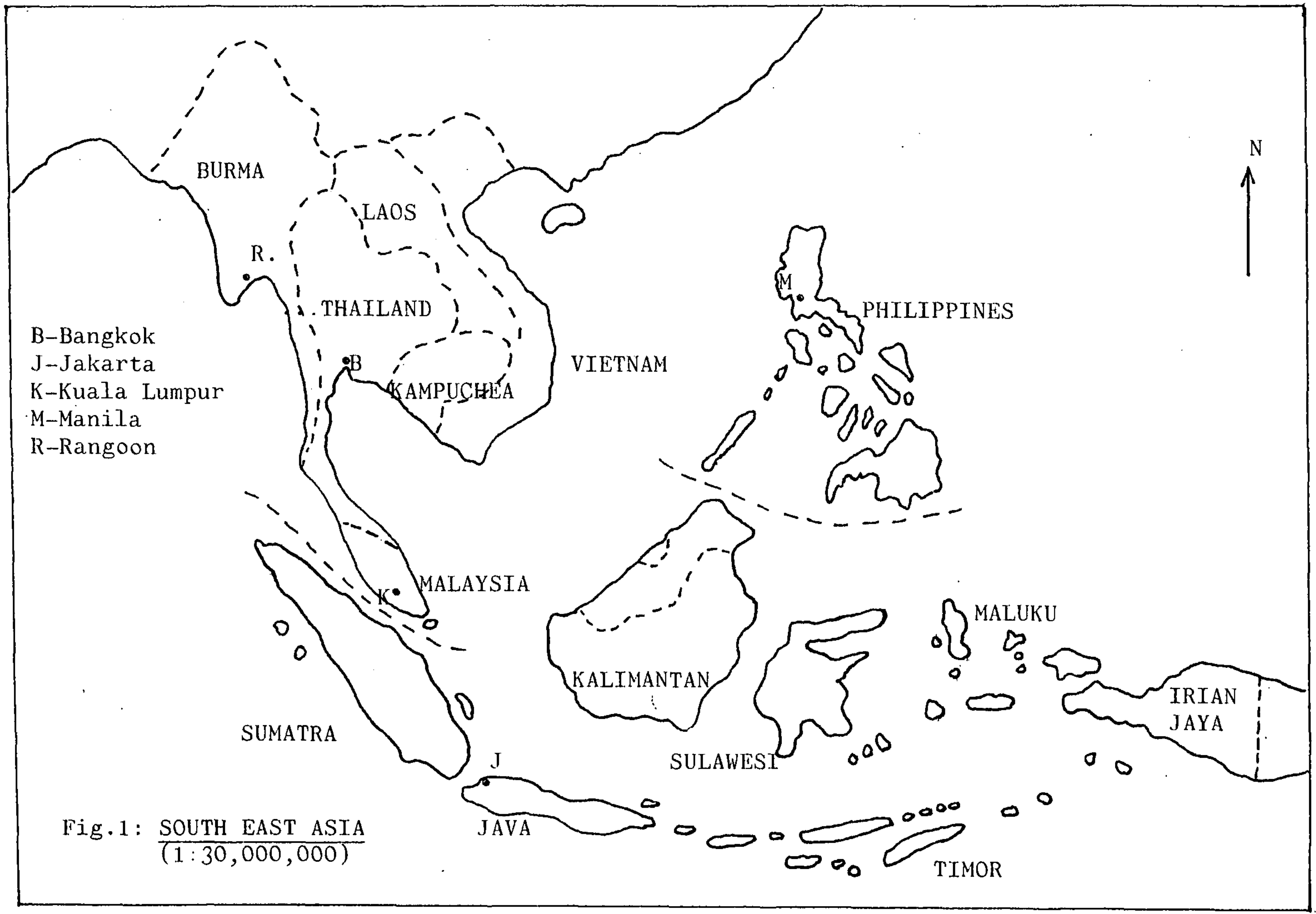


Fig.1: SOUTH EAST ASIA
(1:30,000,000)

which approximately 14% is cured by traditional salting and drying or smoking.

The major fish landing sites are at Samut Sakhon and Bangkok, with other important sites at Songkhla, Rayong and Pattani (Fig.2). Cured fish processing sites and wholesale and retail outlets were surveyed in Bangkok, Samut Sakhon, Samut Prakan, Songkhla, Khon Kaen and Nakhon Sawan. The survey findings are described below.

Samut Sakhon

Samut Sakhon province, is situated to the west of Bangkok and accounts for about 20% of the total fish landings in Thailand. It is a major centre for salted, dried and smoked fish production. A wide variety of fish, including *Saurida undosquamis* (Richardson), common name lizard fish, *Scomberoides commersonianus* (Lacepede), common name queenfish, *Megalaspis cordyla* (Linnaeus), common name hard tail scad, *Rastrelliger* spp. (Indian mackerel), *Scomberomorus* sp. (Spanish mackerel) and *Arius* sp. (marine catfish), is processed there and production rates, of different processors, range from less than 0.5 to over 5 tonnes per day.

After initial dressing, the fish are usually immersed in saturated brine, for a period of 1 to 3 days and are then sun dried, on trays supported by racks, for 1 to 4 days, depending on weather and species. Smoked fish, principally *S. undosquamis*, is sun dried for 1 day, after dressing, and is then smoked, over a saw-dust fire, for 1 night. Processing, tends to be restricted to the dry season and if wet weather interrupts drying during the dry season, the fish are smoked at a lower temperature for 2 days, without prior drying.

The standard of hygiene, at the different processors, ranged from good to poor and tended to be better at the larger processing establishments. Some sites were clean, tidy and well organised, whereas others were littered with discarded viscera, old pieces of fish, together with assorted rubbish and lacked facilities, such as hard standing and adequate drainage. Fish in the brining tanks,

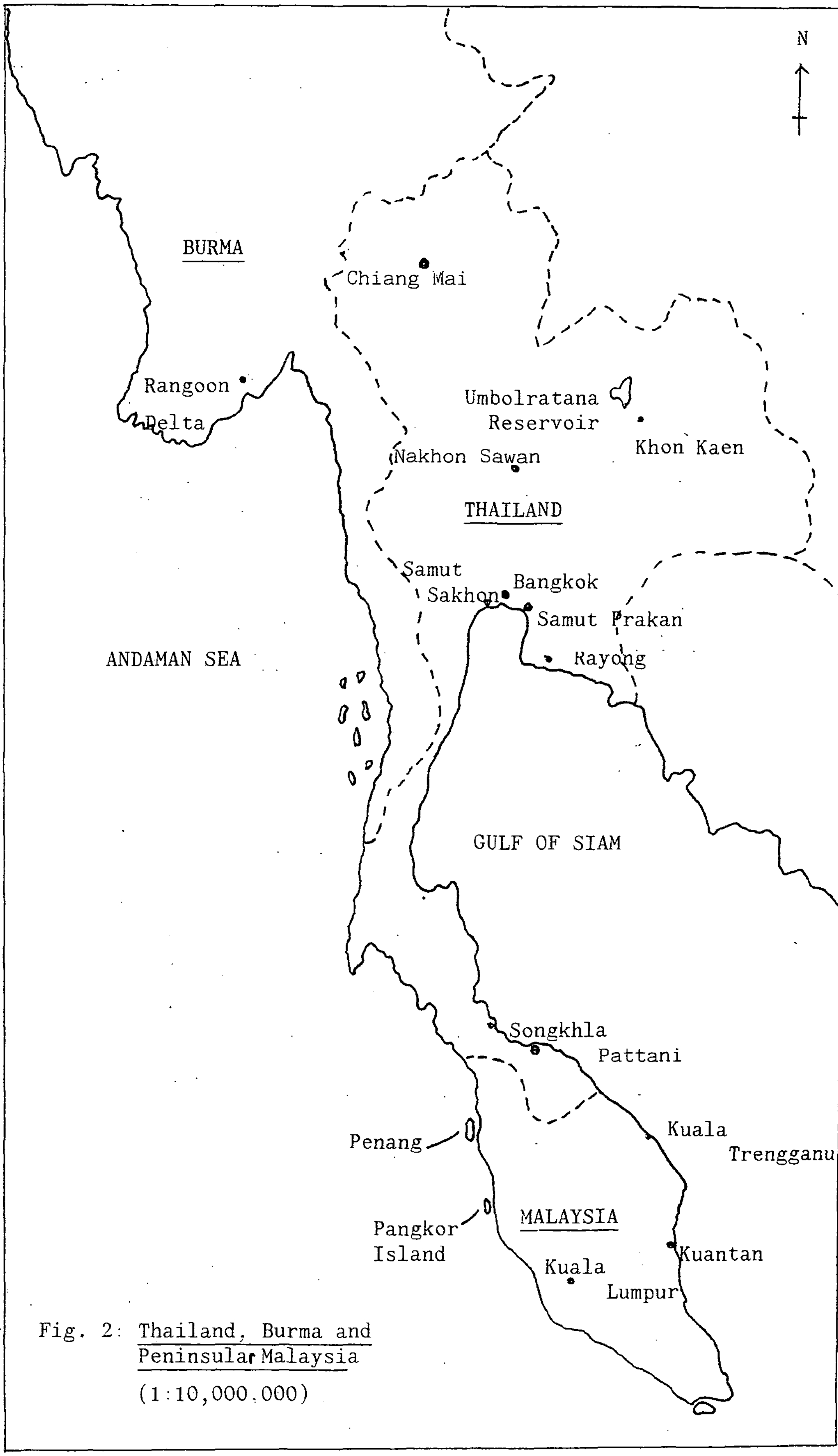


Fig. 2: Thailand, Burma and Peninsular Malaysia
(1:10,000,000)

were usually exposed to blowfly attack and frequently infested with blowfly larvae. 102 larvae, removed from fish and reared to adults, were identified as *C. megacephala* and adults of the same species were frequently observed at the processing sites. 52 adult blowfly samples, collected from 3 processing sites, were identified as *C. megacephala*.

Blowfly infestation was often cited as a major problem by processors and the illegal use of insecticides, on the fish, was frequently evident. The most popular insecticide, used by local processors, was the organophosphorus compound, Dipterex. The active ingredient of this product is trichlorphon (Dimethyl 2,2,2-trichloro-1-hydroxyethylphosphate), which has an acute LD_{50} (rats) of 560-630 mg/kg. Dipterex is approved for agricultural and horticultural use, but is not approved for use on fish. This insecticide powder, is applied to the fish, either by mixing it with the salt used during processing, or as an aqueous dip, after salting.

Infestation with dermestid beetles was stated to be a problem by smoked fish processors, particularly when market conditions forced them to store their produce for more than a few weeks. Specimens of *Dermestes maculatus* (Degeer) and *D. ater* (Degeer), were collected from heavily infested and damaged, smoked, marine catfish, that had been stored at a processor's premises for 2 months.

Samut Prakan

Samut Prakan province, is situated to the east of Bangkok and accounts for about 10% of the total fish landings in Thailand.

Only 1 of the 2 processors interviewed, was processing fish at the time of the visit. Species being processed, comprised *M. cordyla, S. undosquamis* and *Channa* spp. After initial dressing, the fish were soaked in saturated brine for a variable period, depending on species, and then sun dried for 1 to 4 days.

The processor reported losses of up to 15%, in the case of *S. undosquamis*, during the rainy season. The site was very untidy, with fish waste scattered around and large numbers of blowflies had settled on the fish waste and fish in the salting tank. Relatively few blowflies were present on the fish that had just been put out to dry. Blowfly infestation was reported to be a major problem in the rainy season, when the processor found it necessary to dip his fish in a solution of the insecticide Neguvon (another trade name for Dipterex), after salting and washing. The 9 blowfly samples, collected from 2 sites in Samut Prakan, were identified as *C. megacephala*.

Songkhla

Songkhla is a busy fishing port, situated in southern Thailand, which supports a number of canneries, freezing plants and traditional fish processors. The traditional processing community, consists of a large number of small scale operations, located in Khao Seng, on the outskirts of Songkhla.

Fish being processed, included squid, *Arius* sp., *Sardinella* sp., *S. undosquamis* and *M. cordyla*. Standards of hygiene were generally poor and maggot infested brine and fish were frequently observed. Maggot damage could result in the value of the fish dropping by 95%, according to a local processor. Occasional *C. megacephala* and an ovipositing *Lucilia cuprina* (Wied.), were observed on the fish, at 1 of the 4 processing sites visited and many of the fish were blown with blowfly eggs. The standard remedy, used by the processors, was to apply a dip containing a locally purchased, white powder, possibly Dipterex.

Bangkok

Cured fish wholesalers and retailers, were visited at the China-town, New Rd. and Tatien markets, in Bangkok. Cured fish wholesalers are concentrated in the China-town market and 5, of approximately 20 merchants based there, were interviewed.

The most common species on sale, were *Scomberomorus commerson (Lacepede), S. commersonianus, Rastrelliger* sp., *M. cordyla, Tilapia* sp. and *Eleutheronema* sp. (Threadfin). Even though most of the fish were of high moisture content, there was little apparent blowfly infestation. Dead larvae, however, were frequently present on the fish. None of the wholesalers admitted to using insecticide, but a local insecticide merchant, reported frequent sales of Dipterex to local, cured fish merchants.

Several market stalls, retailing a wide variety of cured fish, were visited in the 3 markets. Infestation with blowfly larvae was more evident during the 1982 survey than during a follow up survey in 1986, which probably reflects the use of insecticides becoming more widespread in the cured fish industry. Smoked freshwater catfish, *Kryptopterus* and *Notopterus* spp., were frequently observed to be heavily infested and damaged by dermestid beetles. Retailers estimated that about 5% of smoked fish had to be sold at a reduced price, as animal feed, because of beetle damage. Damaged fish, judged suitable for human consumption, was being sold at 50% of the price of good quality fish.

Khon Kaen

Smoked fish production sites, located close to the shore of the Ubolratana reservoir, were processing *Kryptopterus* and *Notopterus* spp., caught in the reservoir. Processing consisted of sun drying the unsalted, eviscerated fish for 3 hours and then smoking, in a kiln, for 2 days. The moisture content, of a recently processed sample of catfish, was determined to be 15%. The fish were stored for up to a week, before distribution, principally to Bangkok. The fish in store, was in excellent condition and there was no evidence of insect infestation.

Locally produced, smoked and salted, dried fish from Bangkok and Samut Sakhon, were sold at the Central market in Khon Kaen. Some of the locally produced, smoked fish was infested with beetles and the salted, dried fish, from the south, was in general, of poor quality. A basket of mackerel (*Rastrelliger* sp.), contained numerous, dead blowfly larvae, killed by the application of a white powder, by the retailer. One retailer, reported having to sell about 2% of his fish for animal feed, because of insect and mould damage. Losses were reported to have been much higher before a government owned cold store had been built in the city.

Nakhon Sawan

Nakhon Sawan is situated in central Thailand and is a centre for freshwater fish culture and processing. Snakehead (*Ophiocephalus* sp.), is the most commonly cultured species and is processed by salting and drying or smoking, by 6 local processors. A salted, dried snakehead processor was visited and reported a weekly production figure of 2 tonnes of dried fish. Production takes place throughout the year and most of the fish is sold directly to retailers in Chiang Mai or Bangkok. Processing consists of pickle curing the dressed fish and then sun drying for 2 days. Blowflies were reported to be a major problem between May and December, when the processor found it necessary to occasionally apply Dipterex to his fish. Blowfly damage results in approximately 10 tonnes of fish per year, being sold at a reduced price.

The Thailand Food and Drug Administration had constructed a large, polythene solar drier, at a cost of 100,000 baht (£2,500), at the processor's premises, in an attempt to introduce a safe remedy to the problem of blowfly infestation. The processor was not using the drier, as he found that the fish became case hardened and discoloured when processed in the drier. Case hardening occurs when the outer surface dries too quickly and forms a hard seal that prevents further water loss from the internal tissues. He preferred to dry his fish in the open and would only use the drier during prolonged periods of rain. Many attempts have been made to introduce solar driers to fish processors in the tropics, but in most cases, they have been unsuccessful (Walker and Wood, 1985). Curran *et al.*, (1985), evaluated a solar drier in The Gambia, but found that attempts to control the internal temperature, by adjusting the amount of ventilation, allowed blowflies to enter and the design offered no advantages over drying the fish outside. However, Doe *et al.*, (1977), evaluated a simple polythene tent drier in Bangladesh and found it to be an effective method of controlling fly larvae in dried fish. The Bangladesh authorities proposed to introduce similar driers to the fish drying community, but there have been no subsequent reports of their adoption by commercial fish processors.

Two smoked fish processors, both processing snakehead and catfish were visited. At the first site, processing consisted of gutting, washing and cooking over a charcoal fire, followed by smoking for 40 minutes. Production amounted to about 600 kg per week and occurs from June to November. All of the fish is sold in Chiang Mai. Beetle infestation was reported to be a problem, if the fish was not sold within 1 month and could result in losses of 100 to 200 kg per year. At the 2nd fish smoking site, the fish was processed by gutting, washing, drying for 2 to 3 hours, cooking for 24 hours and smoking for 24 hours. Snakehead and catfish were the main species processed and production averaged at 600 kg per day. All fish was processed to order and none was stored by the processor.

Summary

Traditional, cured fish processing is a popular and economically important means of preserving marine and freshwater fish in Thailand. Insect infestation, specifically blowfly infestation of salted, dried fish during processing and dermestid beetle infestation of salted, dried and smoked fish during storage, was found to be a major problem faced by the cured fish industry. In response to this, the use on cured fish, of unapproved insecticides, in particular Dipterex, has become widespread and emphasises the need for the introduction of safe, alternative infestation reduction methods.

2.3 <u>Survey of cured fish establishments in Burma</u>

Fish production in Burma has fluctuated between 519,000 and 631,000 tonnes from 1975 and 1985 and amounted to 644,000 tonnes in 1985. It can be conservatively estimated, from FAO statistics, that approximately 24% of the marine production in 1985 was processed by curing. The government owned, People's Pearl and Fishery Corporation (P.P.F.C.) is the only large scale operator in the fish curing industry and produces solely for export. The produce of small scale operations is mainly for domestic consumption, although a significant proportion, is apparently, unofficially exported.

Due to official travel restrictions in Burma, it was only possible to survey the cured fish industry in the Rangoon area.

Rangoon

The P.P.F.C. establishment was clean and well organised. *Argyrosomus* spp. (Yellow and Spotted Croaker) were being kench cured and dried at the time of the visit and there was no evidence of insect infestation, although a small amount of rodent damage was observed.

Fish being processed at a 2nd site, included *S.commerson*, *Arius* sp., *Trichiurus lepturus* (Linnaeus), common name ribbon fish and stingray. The fish were pickle cured overnight and sun dried on bamboo mats, placed on raised platforms. The reported production level was 200 tonnes per month, over a 7 to 8 month processing season. The bulk of the produce was distributed to the north. The site appeared to be efficiently managed and the processor reported no losses due to insects, rodents etc.

The quality of fish, examined in wholesalers stores, showed marked variation in quality, ranging from very good to almost decomposed. Quality did not appear to influence price. Most of the fish came from the delta region, to the south west of Rangoon (Fig.2) and was generally distributed to the north. Maggot infestation was apparent in some of the large fish eg. threadfin, but it was not possible to rear samples for identification. Preservation and disinfestation techniques included rubbing gunpowder into the fish, dipping in oil, brushing and shaking out insects. Losses, due to mould growth and rancidity, were reported to be high during the rainy season. Despite evidence to the contrary, insects and rodents were, apparently, not regarded as serious problems by the processors and wholesalers questioned. This could either reflect a lack of awareness of product quality by the consumer, or a reluctance to admit to problems, in the presence of foreigners and government officials.

Summary

A limited survey of cured fish establishments in Rangoon, revealed that a large amount of poor quality fish was being handled by local wholesalers. Quality could only deteriorate further during distribution. Although there was no evidence of insecticide use, fish were observed to be infested with blowfly larvae and retailers did admit to finding the use of "repellents," such as pepper, to be necessary. In order to ascertain more fully, the problems being faced by the cured fish industry in Burma, further survey work should be be carried out.

2.4 Survey of cured fish establishments in Malaysia

Fish production in Malaysia, fluctuated between 619,000 and 805,000 tonnes during the period 1977 to 1984 and amounted to 632,000 tonnes in 1985. Cured fish production, for the same year, is estimated to have been about 38,000 tonnes, approximately 6% of the marine production of 661,000 tonnes. The remainder is sold frozen or fresh. It must be pointed out, however, that in 1982, the Malaysian Auditor-General cast doubts on the Malaysian fishery statistics (Anon., 1982)).

Processors, wholesalers and retailers, along the east and west coasts of peninsular Malaysia (Fig.2), were surveyed.

West coast of peninsular Malaysia

Processors, visited on Pangkor Island, reported no problems, although rats, at one processing site, were observed consuming fish meal, adjacent to drying fish. Some drying *S. commersonianus* was infested with unidentified blowfly larvae. Production on the island was dominated by small species, such as *Stolephorus* sp. (anchovy) and

Leiognathus sp.(pony fish), both of which dry quickly and rarely suffer from insect infestation.

A processor, visited in Penang, was processing salted, dried *Scomberomorus* and *Lutjanus* spp. (red snapper). The site was clean and well organised and consequently, losses were negligible. The processor was experiencing problems with Spanish mackerel, imported from Thailand, which had to be immersed in brine, to remove maggots. The presence of a cold store and freezer was also significant in reducing losses.

Retail outlets, in Penang, appeared to be generally free of infestation problems and losses. Salted, dried Spanish mackerel from Thailand was again seen to be infested with blowfly larvae and one retailer reported losses of 20%, due to insect infestation, before installing a cold store.

East coast of peninsular Malaysia

Several salted, dried fish processors were visited in the vicinity of Kuala Trengannu. Fish being processed, included *Arius, Stolephorus, Sardinella, Parupeneus* spp., (goatfish) and stingray. Facilities and standards of hygiene, were lower than observed on the west coast and maggot infestation was frequently both observed and cited as a problem by processors. Dusting the fish with black pepper was given as a method of disinfestation.

A wholesaler in Kuala Trengannu, estimated losses of 30% due to blowfly infestation, despite having access to a cold store. The problem was reported to be worse during the rainy season, when badly infested fish frequently had to be sold as fertilizer.

Maggot infestation was also observed to be a problem at retail stalls visited in Kuala Trengannu, Sungai Petani, Marang and Kuantan. Retailers reported losses of 10 to 50% during the rainy season, when spoiled fish is often sold for animal feed or fertilizer. Disinfestation methods included applying pepper, resalting and drying, brushing/trimming the fish or storing it in a cold store.

Summary

The survey revealed that processors, wholesalers and retailers, located along the east coast of Malaysia, were experiencing losses of up to 50%, due mainly to blowfly infestation. No effective, remedial measures were evident and, at the time of the survey, there were no observations of insecticide use. In view of the extent of the blowfly problem and observations, elsewhere in South East Asia, this situation might well have subsequently changed since the survey, conducted in 1982.

2.5 Survey of cured fish establishments in Indonesia

Indonesia is the largest of the countries visited, with a population in excess of 150,000,000. The most densely populated island is Java , which, although comprising only about 7% of the land area, has over 65% of the population.

During the period 1977 to 1984, total fish production has fluctuated between about 1.57 million and 2.2 million tonnes. The production for 1985 was 2.07 million tonnes, of which the marine catch accounted for 87%. Approximately 37% of the marine catch was cured by salting, drying or smoking, during the same year. Salted, dried fish, accounts for about 92% of cured fish production, with Java and Sumatra, being the most important production areas. The average fish consumption (1981-1982) was reported, in the government statistics, to be 14.5 kg/capita/annum (although this is probably an underestimate) and accounted for about 70% of the total, dietary, animal protein intake.

West Java

The salted, dried fish processing sites and retail outlets, visited in West Java, are shown in Figure 3 .

Jakarta

The main traditional processing areas are located at Kali Baru and Muara Angke, on the north side of the city. Facilities have recently been improved at Muara Angke and the government is encouraging processors to move there from

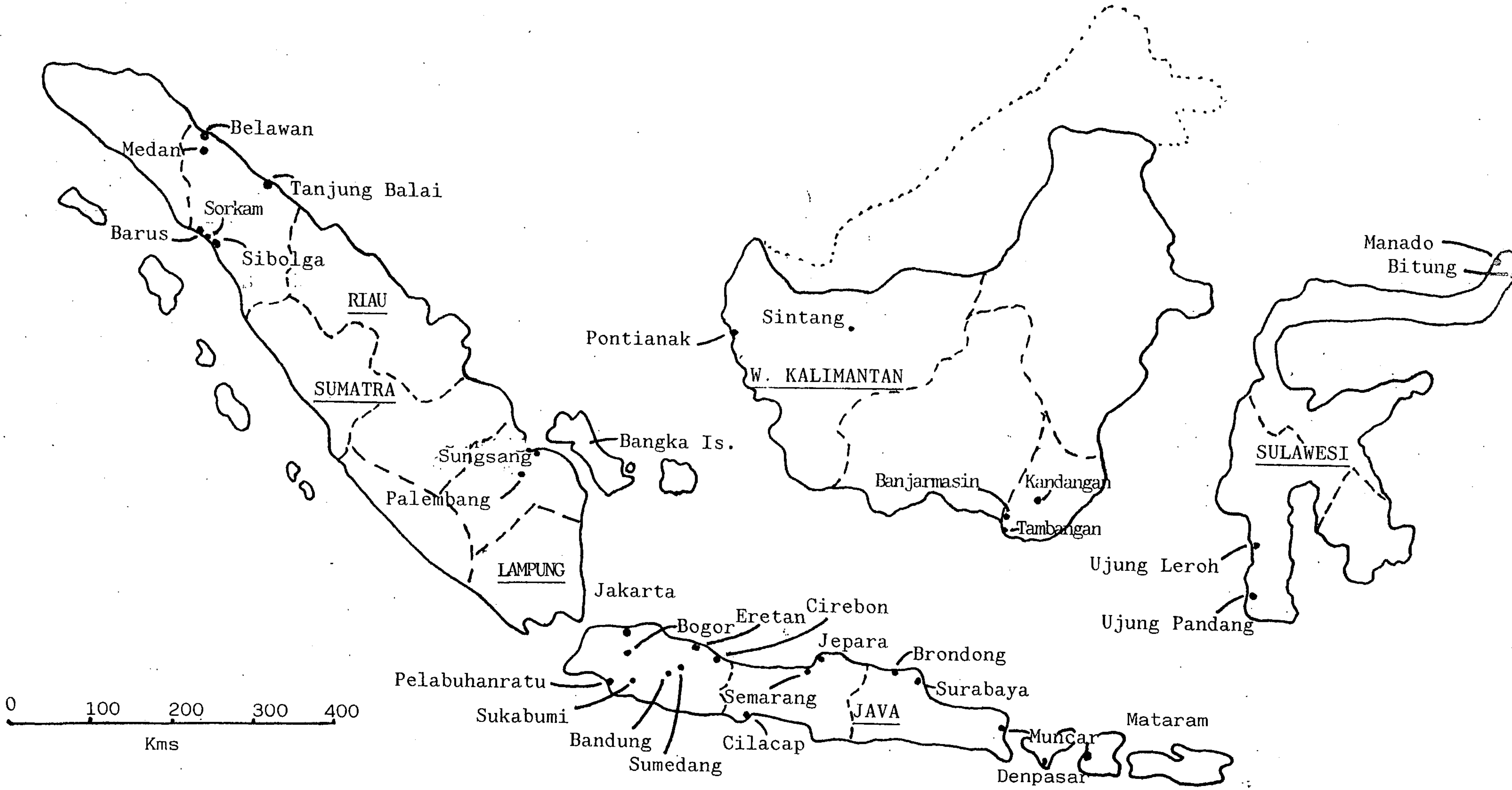


Fig.3 Locations visited during surveys of cured fish establishments in Indonesia.

Kali Baru, where basic facilities, such as running water are lacking.

Fish being processed at Kali Baru, included *Arius, Scomberomorus* and *Lutjanus* spp. After evisceration, the fish were usually pickle cured overnight and then sun dried, on trays, for 3-6 days. Blowfly activity, in particular *C. megacephala*, over the drying fish, was very apparent and processors reported that blowfly infestation was their major problem, especially during the rainy season. Processors have resorted to spraying their fish with easily available, household insecticides, such as Baygon and Startox , in order to control infestation. A similar situation exists at Muara Angke, despite the presence of improved facilities.

The active ingredients of Baygon are dichlorvos and propoxur. Dichlorvos (dichlorovinyl-dimethyl-phosphate) has a high acute oral toxicity (LD_{50} rats, 56-80 mg/kg). However, mammals can rapidly degrade and detoxify sub-acute dosages to O,O-dimethyl- phosphate (Sayeed Quaraishi, 1977). Propoxur (O-isopropoxy-phenyl-N-methyl carbamate), also has a high acute oral toxicity (LD_{50}, rats 70-200 mg/kg).

Startox contains dichlorvos and bioallethrin (LD_{50} 425-575mg/kg) and like Baygon, is unsuitable for use on food commodities. Bioallethrin is a synthetic pyrethroid insecticide with an LD_{50} rats of 860 mg/kg. In addition to containing highly toxic active ingredients, both products use kerosene, which is known to be carcinogenic, as the carrying agent. Similar products, such as Swallow (bioallethrin, propoxur and dichlorvos) and Mafu (dichlorvos, permethrin and piperonyl butoxide), are also available in local stores.

A local wholesaler, listed fragmentation, insect infestation and mould spoilage as being responsible for about 10% of his stock having to be sold as animal feed. Local retailers reported having to sell between 15 and 30% of their stock as animal feed, for the same reasons.

Eretan

Marine catfish (*Arius* sp.), was the main species being processed at the time of the visit. Treatment consisted of pickle curing overnight, followed by sun drying for 4 to 6 days. Again, blowfly infestation was described as a major problem. Dead blowflies were observed on some of the drying fish and the processor reported finding it necessary to control infestation by applying Baygon.

Cirebon

Cirebon is an important fishing and traditional processing centre, where several species, including *Arius, Scomberomorus, Sardinella, Rastrelliger* and *Leiognathus* spp. are salted and dried. Blowfly activity was very heavy over drying *Arius* sp., at one processing site and many of the fish were blown with eggs. Losses, due to blowfly infestation, were estimated by the processor, to be about 15%. During the initial survey in 1982, there was no evidence of insecticide use by the local processors. However, during subsequent visits between 1983 and 1988, it became apparent, that use of insecticides, such as Baygon, Startox and Swallow, was widespread. The most popular methods of application were either spraying or smearing neat insecticide onto the fish.

Beetle samples, collected from infested *Arius thalassinus* (Ruppell), were later identified as *Dermestes carnivorus (Fab.), D. maculatus* and *D.* ater.

Gebang

The main species, processed by the cooperative at Gebang, were *Arius* sp., *M. cordyla, Pampus argentus* (Euphrasen), common name pomfret, *Rastrelliger spp., S. commersonianus, Sardinella* sp., *T.lepturus* and a mixture of small species. Processing usually consisted of a pickle cure, followed by sun drying, on mats placed on tables, or the ground.

Although the complex was generally well organised, basic facilities were absent and the fish were typically of poor quality. Many fish were maggot infested and it was reported that heavily infested fish were thrown away. There was no evidence of insecticide use at the time of the survey in 1982, or during a later visit in 1988.

Bandung

Bandung is the major distribution centre for cured fish in West Java and handles fish from all over Indonesia. Several wholesalers were visited and reported from 10 to 50% of stock having to be sold for animal feed, at low prices. Principal causes of losses, were given as insect damage, rancidity and microbial spoilage. Losses, primarily due to beetle infestation, tended to occur when the fish had been in store for over 2 months and preventative measures included occasional spraying with Baygon.

Fish on sale at local retail markets, showed evidence of dermestid beetle and blowfly infestation. Some retailers placed Baygon powder on a piece of paper, left on top of the fish, to act as "bait". Other than occasional brushing, no other loss prevention methods were observed.

Sukabumi

Some of the fish, on sale at local retailers, was of low quality, due to fragmentation, blowfly larvae, mould and discolouration. It was reported, that during the rainy season, between 10 to 40% of stock was sold for animal feed, at 10 to 20% of the normal price.

Pelabuhanratu

Rancid, discoloured and mouldy fish were evident at the retail market, but no insect infestation was apparent. Spoilage was reported to occur during the rainy season, when about 10% of stock was sold at reduced prices.

Bogor

Local wholesalers handle fish from throughout Indonesia, mainly from Sumatra and East Java. Some stock was in poor condition, due to fragmentation, insect infestation, mould and rancidity. One wholesaler estimated about 10% of fish having to be sold as animal feed.

Central Java

Processing sites were visited at Jepara and Cilacap (Fig. 3). Processors at Jepara, were mainly handling small species, belonging to the families Leiognathidae, Engraulidae and Trichiuridae. These fish dry quickly and no infestation was apparent. Infestation was reported to be a problem in the rainy season, when approximately 10% of fish are damaged.

Several species of fish, including large species, such as marine catfish, are processed at Cilacap. Most processors cited blowfly infestation as being their main problem and salting tanks, infested with maggots, were frequently observed. *C. megacephala*, *L. cuprina* and *Musca domestica vicina* were reared from larvae collected from infested *Arius* sp., during processing trials in Cilacap. *M. domestica* has been reported to occasionally infest cured fish in Indonesia, by Indriati *et al.*(1985), although had not previously been reared from larvae infesting fish. Petrol and Baygon were used by some processors, to protect their fish against infestation.

East Java

Fish processing and retail outlets were visited in Brondong, Surabaya and Muncar (Fig. 3).

Brondong

Brondong is an important fishing centre, that produced 16,000 tonnes of fish in 1985. The catch is dominated by *Decapterus russelli* (Ruppell), which accounted for 86% of the production and *Rastrelliger kanagurta* (Cuvier), which amounted to 5%. Large fish, eg.*Lutjanus*, *Arius*, *Scomberomorus* and *Coryphaena* spp.(dolphin fish) are also landed. Almost the entire catch is processed locally, by salting and drying or salting and boiling.

Processors reported that blowfly infestation was a major problem in the rainy season. Fish were observed to be infested in the salting tanks and on the drying racks. Larvae from infested fish were reared to the adult stage and identified as *C. megacephala* and ovipositing *L. cuprina* were observed on drying fish. The standard of hygiene ranged from good to very poor and there was evidence that applying Baygon or Startox were the standard methods of reducing infestation.

Surabaya

Wholesalers in Surabaya, reported that blowfly larvae were often present on inadequately dried fish and could cause a drop in value of between 50 and 75%. Browning, rancidity and belly burst were also given as problems.

Both dead and alive blowfly larvae were present on fish for sale at the local market. Most of the retailers interviewed, admitted to controlling the problem by spraying the fish with neat Baygon or Startox. Blowfly infestation was obviously the most serious problem being faced by the retailers and, in one case, was stated to cause a 45% drop in value.

Muncar

Muncar ranks 2nd in importance to Brondong, as a fishing centre and approximately 60 salted, dried fish processors are located there. Unfortunately, no processing was taking place at the time of the visit. A previous visit had revealed blowfly infestation of *Arius* sp. and losses of 30% in large fish, due to blowfly infestation were reported (Hanson S.W. *pers. comm.*).

2 local retailers were visited and were observed to apply the insecticides Baygon and Mafu to their fish.

Sulawesi

Sulawesi produces approximately 16% of fish in Indonesia, of which, about 11% is landed in the province of South Sulawesi. Ujung Pandang and Ujung Leroh, in South Sulawesi and Manado and Bitung, in North Sulawesi were visited (Fig. 3).

Ujung Pandang

Mixed, small fish, predominantly *Sardinella, Stolephorus* and *Leiognathus* spp. were being processed at one site in Ujung Pandang. None of the fish were infested with blowfly larvae and there was no adult blowfly activity. A 2nd processor, visited in 1988, was observed to treat his fish with the agricultural insecticide, Sevin 85 S. The insecticide was added to the brine, before the fish were salted. The active ingredient of this insecticide is carbaryl (1-naphthyl methylcarbamate), which has an acute LD_{50} (rats) of 4000 mg/kg and acts as a cholinesterase inhibitor. It is principally used to protect grain products during storage.

3 local wholesalers, reported experiencing problems with blowfly infestation, particularly when business was slow. Poor quality, infested fish, suffered a 60% reduction in retail value. Heavy mite infestation was observed at the premises of 1 wholesaler. The species was identified as *Lardoglyphus konoi* (Sasa and Asanuma) and was stated to cause a drop in retail value of 30-35%.

Ujung Leroh

Most of the fish landed at Ujung Leroh, is processed at sea and further dried on land, when necessary. Blowfly infestation was reported to cause problems, particularly to *Decapterus* sp.

Manado and Bitung

Traditional processing in Manado and Bitung, consists solely of smoking, using either a hot or cool smoking technique. Tuna is the most commonly processed fish and none of the processors interviewed, were experiencing losses or quality problems. Salted, dried fish, on sale in Manado, was in good condition and free of insect infestation. There was no evidence of insecticide use and none of the retailers reported losses.

Nusa Tengarra Barat

Nusa Tengarra Barat comprises 2 islands, Lombok and Sumbaya and is one of the smallest provinces in Indonesia. The total annual fish landings, of about 45,000 tonnes, represent approximately 2% of the total Indonesian landings.

Lombok

Large quantities of cured fish pass through Lombok, on the way to East Java. It was reported that much of this fish is in poor condition and typically 10 to 15% is sold as animal feed.

Samples of salted, dried tuna, *Euthynnus affinis* (Cantor), purchased at the fish market in Mataram, were found to contain potentially dangerous levels of histamine, a sign of prolonged storage at high temperatures. Hanson and McGuire (1982).

South Sumatra

South Sumatra is the 5th largest province of Indonesia, with fish landings totalling about 100,000 tonnes. Bangka Island is the main fish landing and processing region in the province.

Bangka Island

A variety of fish, including *Scomberomorus, Lutjanus, Scomberoides, Sardinella* and *Stolephorus* sp., were being processed. *C. megacephala* activity over the fish was low, although processors reported that infestation could be a problem and that petrol was applied to fish to reduce insect damage.

Very poor quality fish, including some infested with blowfly larvae, were on sale at a local wholesaler. Approximately 20% of stock was reportedly sold as animal feed.

Palembang

Palembang is an important fish distribution centre, both for marine species processed in Sungsang, Bangka Island, Padang and Bititung and freshwater species, processed in Inderalayu. Numerous cured fish retailers and approximately 25 wholesalers are located next to the fish landing site and reportedly handle about 450 tonnes of dried fish per month. The standard of hygiene at the cured fish market was found to be very poor and much of the fish, in particular the large species, was heavily infested with *P. casei* and blowfly larvae. Both *C. megacephala* and *L. cuprina* adults were observed on the fish.

Sungsang

Sungsang is a fishing community of approximately 3,000 inhabitants, located at the mouth of the Musi river. Most of the houses are built on stilts, over the littoral zone and are connected by raised, wooden paths. Basic services, such as piped water and sewerage are absent, the community is dependent on rainwater runoff, from the roofs of the houses and waste is simply dropped onto the mud, beneath the houses. The visiting doctor, accompanying us, reported that gastric ailments were very common and that cholera was present in the community.

Approximately 50 small scale fish processors operate in Sungsang and produce about 8 tonnes of fish per month, during the peak season, from March to May. One processor reported blowflies as being his main problem and estimated infestation levels of about 25%. Infested fish were devalued by 25%. Some processors were reported to spray their fish with gasoline, as a remedial measure. Blowfly larvae were observed in the salting tanks of 2 processors and *C. megacephala* and *L. cuprina* were present on drying *Arius* sp.

North Sumatra

Annual fish landings total about 160,000 tonnes in North Sumatra and major landing areas include Belawan, Tanjung Balai, Sibolga and Barus (Fig.3). Salting and drying are the most widely used traditional processing methods in the province, although some salted, boiled and smoked fish are produced.

Belawan

12, privately owned, landing sites operate in Belawan and principally handle fresh fish. Drying is simply regarded as a means of handling surplus fish, especially small pelagic species eg. *Decapterus, Rastrelliger* spp. The sites visited, were clean and very well organised. No problems were observed or reported.

Tanjung Balai

Small pelagic fish were again the main species processed and no problems were evident. Marine catfish, *Arius* sp., were being dried and a processor reported occasional problems with blowfly infestation. Larvae were removed by hand.

Fish, on sale at the local retail market, was generally of low quality and blowfly infestation was evident. Large fish were sprayed with petrol to reduce infestation. Severe spoilage, resulted in fish being sold for animal feed.

Medan

Discolouration, infestation and fragmentation were given as problems by wholesalers. Infestation of large fish was evident and several bags of small fish were being sold as animal feed at reduced prices.

Sibolga

A variety of fish, including *Decapterus, Sardinella, Megalaspis, Lutjanus* and *Stolephorus* spp. are salted and dried by local processors. Blowfly infestation was reported as an occasional problem and one processor found it necessary to apply Baygon to his fish, on occasions.

Approximately 25 wholesalers operate in Sibolga and most reported having no problems with their fish. One wholesaler was observed to spray his fish with Lebaycid 550EC, the active ingredient of which is fenthion (0,0-dimethyl-0-(4-methylmercapto-3-methylphenyl)-triphosphate). This compound has an acute LD_{50} (rats) of 190-615 mg/kg and is a cholinesterase inhibitor. It is produced for pre-harvest use on crops.

Only 1 of the retailers interviewed, reported occasional blowfly infestation and there was no evidence of insecticide use in the market.

Sorkam

Mixed, small species of fish, being pickle cured, were observed to be heavily infested with blowfly larvae. The processor applied petrol to his fish as a remedy to infestation.

Barus

Approximately 20 dried fish processors operate in Barus and process a variety of fish, including *Stolephorus, Leiognathus, Arius, Lutjanus* and *Scomberomorus* spp. Approximately 40% of large fish were reported to become infested with blowfly larvae during periods of prolonged, wet weather. Some heavily infested tuna were observed on the drying racks. Remedial measures included applying Baygon or petrol to the fish.

<u>West Sumatra</u>

West Sumatra produces approximately 1.5% of fish in Indonesia and Padang is a principal fish landing port. Although no cured fish is produced in Padang, it is an important distribution centre of cured fish.

Padang

Approximately 25 retailers, handling fish from Palembang, Medan and Kalimantan, sell dried fish at the central market. The fish on sale was, without exception, of good quality and showed no sign of insect infestation. Some blowfly infested queenfish and *Dermestes* infested snapper were seen at 1 wholesaler's premises, apparently destined for animal consumption.

West Kalimantan

Approximately 3% of Indonesia's fish is landed in West Kalimantan and marine fish account for about 80% of the local production. Pontianak is the principal distribution centre for both marine and freshwater fish.

Pontianak

Two salted, dried fish processors operate in Pontianak and process a variety of marine fish species. The larger of the 2 operations, produces in the region of 30 tonnes per month, although production drops to 10 tonnes per month in the rainy season. Blowfly activity, over the fish in the salting tanks, was high and numerous fish were infested with blowfly larvae. Dead blowfly larvae, were present on some fish and the processor reported finding it necessary to spray infested fish with Baygon, dissolved in paraffin oil, when blowfly activity was high. Both *L. cuprina* and *C. megacephala* were present on the fish and 35 adult *C. megacephala* were reared from larvae infesting *Arius* sp. during drying.

Several salted, dried fish wholesalers, handling a variety of fish, including *Scomberomorus, Arius, Sardinella, Decapterus, Ophiocephalus* and *Notopterus* spp., were visited and the fish in stock, was of variable quality. Infestation with *P. casei* and *D. maculatus* was evident and live blowfly larvae were frequently present. Dead, 3rd instar, blowfly larvae, probably killed by insecticide, were observed on salted, dried *S. commerson*, at one wholesaler's premises.

Sintang

Sintang is situated along the Kapuas river, approximately 400 km inland from Pontianak. Traditional fish processors in the district, have formed an association and reportedly produce in the region of 5 tonnes of salted, dried fish a year. *Ophiocephalus* is the most commonly processed species and processing is achieved by descaling, gutting and splitting, followed by a pickle cure for 24 hours and then sun drying, on racks, for 3 to 4 days. During poor weather, the fish is apparently smoked. Unfortunately, no processing was taking place at the time of the visit, as the river level was too high for fishing.

Sintang is a distribution centre for salted, dried fish that is processed upriver, in the Kapuas Sulu district. *Ophiocephalus* and *Heliostoma* are the most commonly processed species and peak production occurs from June to September. Several retailers and a fish collecting boat were visited and the fish examined, was in good condition, showing no signs of insect infestation or damage. There was no visible evidence of insecticide use, although it was reported by the proprietor of the collecting boat, that some fish traders spray Baygon on their fish.

Kuala Karang

Kuala Karang is a small fishing community, situated on 1 of the islands, south of the mouth of the Kapuas river and is approximately 6 hours, by launch, from Pontianak. The fish used for salting and drying, consists predominantly of mixed, small species, that form the by-catch of the local, shrimp fishing fleet. Trammel and gill nets are the most commonly used gear and the fleet consists of approximately 100 vessels. 3 relatively large scale processors, each producing about 1-5 tonnes of dried fish per month and numerous small scale processors, operate in the village and peak production occurs between July and October. Many of the processors visited, were observed to be having problems with blowfly infestation and there were numerous instances of fish suffering heavy infestation in the salting tanks and infestation during drying. Apart from 1 processor, who made a point of completely submerging his fish in brine, no effort was made to close the the salting tanks, to protect the fish from blowfly infestation. While there was no evidence of insecticide use by the processors at the time of the visit, the widespread occurrence of blowfly infestation, makes it likely that insecticides, such as Baygon and Startox will eventually be adopted by the processing community, unless safe alternatives are introduced.

South Kalimantan

Approximately 5% of Indonesia's fish production comes from South Kalimantan and about 60% of production is provided by inland capture fisheries.

Banjarmasin

Banjarmasin is the principal centre of cured fish distribution in South Kalimantan. Several wholesalers, handling a variety of fish, were visited at the central, wholesale market. Several of the larger fish species on sale, were heavily infested with blowfly larvae and some marine catfish, which had been in store for 3 months, was badly damaged by *Dermestes* larvae and was being sold, at a reduced price, as animal feed.

K. Tambangan

Approximately 100 boats, using gill nets and beach seines, operate from this small fishing village of approximately 600 inhabitants. About 75% of the catch is processed by salting and drying. The village head reported that local processors apply Sevin to their fish, to remedy blowfly infestation, which is particularly severe in the rainy season. Before 1987, Baygon was the most commonly used insecticide in the village, but the processors now prefer to use Sevin, as it does not impart a taint to the fish.

Kandangan

The large swamp fishery, comprising some 900 hectares and supporting some 15 fishing villages, principally produces *Trichogaster, Ophiocephalus* and *Heliostoma* spp. The fish is either sold live to collectors or is lightly salted and dried. The dried fish is purchased by wholesalers in Banjarmasin and distributed to Java. The peak processing season occurs between July and September, when up to 100% of the catch is dried. Blowflies were reported to cause problems in September.

Summary

Salting and drying is one of the most important fish preservation methods used in Indonesia, accounting for approximately 37% of the marine catch and is likely to remain important for the foreseeable future. The survey revealed, that losses due to blowfly, principally *C. megacephala*, infestation, during processing and dermestid infestation during storage, were widespread and high. A variety of household insecticides were being used on the fish, to reduce insect infestation. This dangerous practice was particularly prevalent in Java, but is also common in other parts of Indonesia. The use of kerosene based insecticides on fish must increase the risk of bowel cancer developing in consumers and the careless manner in which these insecticides are handled by processors, must present a serious hazard to the processors and their families.

2.6 Survey of cured fish establishments in the Philippines.

The total catch for the Philippines was 1.87 million tonnes in 1985, of which approximately 22% was processed by salting and drying or smoking.

Processors were visited in Navotas, Metro Manila, which is the most important landing centre in the Philippines. Local retail markets were also surveyed.

Salted, dried fish, being processed at Navotas, included *Rastrelliger, Decapterus, Nemipterus* and *Sardinella* spp. Processing consisted of brining, followed by sun drying on tables. Production fluctuated between 32 and 80 tonnes per month and the processor estimated his losses to be 30% due to blowfly infestation during the rainy season and 5% over the rest of the year.

Fish on sale in local markets, was of variable quality, ranging from very good to decomposed. Fragmentation and mould were regarded as problems and even though some fish were seen to be infested with *D. carnivorus*, insect infestation was not regarded to be a cause of losses by retailers.

2.7 Summary of survey findings

The survey revealed traditionally processed fish to be an economically important commodity in South East Asia and an important source of animal protein for the population, particularly in rural areas. Losses of cured fish, both physical and financial, result from a range of causes and are particularly high in small-scale processing operations, where facilities and standards of hygiene are poor. Blowfly infestation was found to be one of the most important causes of losses and in excess of 3,000+ blowfly samples, collected from fish at 20 processing sites, at 10 locations in Indonesia and Thailand, showed that 1 species, *C. megacephala*, was overwhelmingly involved. Poor standards of hygiene and lack of care to prevent infestation, also contribute to the high losses (see later sections). Losses during storage were found to occur in all countries visited and were principally due to infestation with *Dermestes* spp. and *P. casei*. Fragmentation, rancidity, mould spoilage and rodent attack are also important causes of losses.

The use of insecticides, such as Baygon, Startox, Mafu, Swallow, Lebaycid, Dipterex and Neguvon, all of which are both potentially dangerous to man and unapproved for use on fish, appeared to be an increasing problem and illustrated the urgent need for the further investigation and development of safe, practical methods of reducing infestation.

+ Appendix 1

3. FIELD INVESTIGATIONS INTO THE NATURE OF INSECT INFESTATION OF CURED FISH IN INDONESIA

3.1 Introduction

As a result of the survey finding, that insect infestation was a major problem during the processing and storage of cured fish, it was decided to concentrate the research effort on finding methods of reducing this important cause of post-harvest losses. Before commencing loss reduction trials, it was necessary to investigate, in detail, the nature of insect infestation. The aims of this investigation were to determine at which stages of processing and storage the fish were most susceptible to infestation, to acquire information on the biology of the insects concerned and to develop field methods of infestation monitoring and assessment. Most of the initial infestation data was collected during fieldwork in Cirebon, W. Java in 1983 and 1984. Further information was assembled during subsequent loss reduction trials, between 1984 and 1988, at Cirebon and other locations in Indonesia and Thailand. Detailed biological information on the most important blowfly species, *C. megacephala*, was obtained from laboratory investigations in Grimsby, which are described in later sections.

3.2 Processing site and fish processing techniques used during the initial investigation into insect infestation.

The initial investigation and subsequent loss reduction trials, were carried out at a commercial fish processing site, because this was the only way to obtain results under realistic processing conditions. Although it would have been possible to hire a site and process the fish ourselves, possibly under more controlled conditions, this would not have resulted in an insight into the subtleties of the processing operation, or the problems faced by the processor. A detailed insight into a cured fish operation can only be obtained by working closely with a processor, over a lengthy period of time and is essential, if effective loss reduction methods, that are acceptable to the processor, are to be successfully developed and introduced.

The processing site, which had been visited the previous year, was selected because the processor was processing large species of fish, that were susceptible to blowfly infestation and was observed to be experiencing serious blowfly infestation problems with his fish. The site was located at 59 Gg. Cucut, Pesisir, Cirebon, W. Java and owned by Hadji Sonja, whose trust and full cooperation were essential to the success of the field trials.

The processing premises were situated within 0.5 km. of the fish landing site and were typical of the small scale processing operations found in the area. They consisted of a one storey building, containing storage rooms and a small, concreted processing area, accommodating two concrete salting tanks. The fish were sun dried on interwoven bamboo mats, supported by racks, along the dirt road at the front of the premises.

The 2 species of fish, *A. thalassinus* and *S. commerson*, used for the trials, were purchased at the local landing site. Freshness of the raw material was variable, from a few hours to about 14 days old. In most cases the fish had been well iced, with anal temperatures of less than 5°C. The fish were labelled with a code number, weighed, transported by becak (tricycle taxi) to the processor and usually processed immediately on arrival.

A. thalassinus was processed by beheading and gutting on a rough concrete floor, placed in 1 of 2 adjacent, concrete tanks, submerged in fresh water and allowed to soak overnight. This allowed the fish to ferment, which imparted a desirable soft texture to the flesh. The fish were next removed from the water, drained and then salted, by placing solar salt in the abdominal cavity, before being returned to the salting tank. Each layer of fish, in the salting tank, received a sprinkling of salt. The dimensions of the 2 salting tanks were 71 x 46 x 53 cm and 64 x 55 x 53 cm respectively. Both tanks were in a state of poor repair, with rough, damaged surfaces and edges. The plastic sheeting, used by the processor to cover the fish during salting, could not be closely fitted to the top of the tanks and hence allowed easy access to blowflies. Each tank could accommodate approximately 7 layers of 8 large fish. The quantity of fish processed, depended on availability and was usually below full capacity. On occasions, however, two salt storage tanks would also be used to process unusually large quantities of fish. The salt used, was of poor quality and discoloured by the presence of sand and silt. During salting, a pickle formed, that completely submerged the lower layers of fish. The upper layer of fish was partially exposed to the air and hence blowfly attack. The fish were allowed to pickle for approximately 24 hours and then removed from the tank, split and returned, for a few hours, to the pickle, which was diluted with a few scoopfuls of fresh water. They were then, again removed from the pickle, washed in fresh water and placed on interwoven bamboo mats, the dimensions of which were 94 x 62 cm. Care was taken, at this stage, to smooth down the cut surfaces of the fish, before placing them out to dry. This was believed to reduce the possibility of blowfly infestation. The drying period varied with fish size and environmental conditions. A good drying day would typically last for 8 hours, whereas a poor day might be as short as 3 hours. The fish received a 2nd split on the 2nd drying day and were occasionally turned over until judged sufficiently dry, usually after 4 to 6 days, in the case of large fish. When necessary, the processor sprayed Baygon onto the drying fish. During the drying period, the fish were placed indoors overnight. At the end of drying, they were stacked in piles and stored indoors, before being sold.

S.commerson was split "head on", gutted and completely submerged in brine for 24 hours. After removal from the brine, the fish were washed and placed out to dry on bamboo trays. They were occasionally turned over during drying, which took from 2 to 3 days.

Generally little attention was paid to hygiene during processing. The fabric of the building and the salting tanks were in a poor state of repair and difficult to clean. The interwoven mats, used for drying, were seldom washed and provided convenient refuges for blowfly larvae during drying. The fish were exposed to damage by blowflies, beetles, rodents and cats throughout processing and storage.

In order to obtain representative results, the standard processing methods were used throughout the initial investigation and subsequent loss reduction trials (refer to Section 4 for details), except where indicated otherwise.

3.3 Environmental data and changes to the fish during processing.

Materials and methods

Temperature and humidity were continuously monitored, using a Casella standard thermohygrograph. The instrument was calibrated each morning and placed outside with the drying fish*. After each day's drying, it was returned to the store, with the fish.

Pickle temperature was recorded, at intervals, using a mercury thermometer and samples of pickle were collected, at the end of salting, for analysis of salt content. Salt concentration was determined as % NaCl (weight/volume), by titration with silver nitrate.

Weight loss during drying, was used as an indicator of change in fish moisture content. The fish were weighed at hourly intervals after salting and the recorded weights expressed as a percentage of the weight recorded immediately after salting. In addition, samples of fish were analysed for salt and moisture content, at the end of drying. The standard, analytical methods, described in FAO (1981) were used.

* For 1983 trials, the thermohygrograph was placed in the sun. For all subsequent trials, it was placed in the shade.

Results

During the daytime, the fish were exposed to temperatures ranging from 30°C. to 48°C. in the open (Figs. 4 and 5). Relative humidity fluctuated between 85 and 40% during drying. Daily changes in relative humidity were generally characterised by a decrease during the morning and early afternoon, followed by an increase through the late afternoon and evening. Overnight, both temperature and relative humidity remained relatively constant. Overnight temperature fluctuated between 28 and 30oC. and relative humidity remained in excess of 70%.

The salting and drying process resulted in flesh moisture content being reduced to between 38 and 51% in *A. thalassinus* (Table 3) and 49% in *S. commerson*. By the end of

Table 3. Changes in moisture and salt content of *A. thalassinus* and *S. commerson* during processing.

Moisture and salt content of flesh	TRIALS+ 1 Fresh	Dry	2 Fresh	Dry	3 Fresh	Dry	4 Fresh	Dry
Moisture (% wwb)								
Arius	73	38	80	47	79	43	79	51
Scomberomorus	75	49	--	--	--	--	--	--
Salt (% dwb)								
Arius	--	30	--	23	--	22	--	15
Scomberomorus	--	21	--	--	--	--	--	--

Fresh = fish samples taken before processing, on arrival at processor's premises.
Dry = fish samples taken at completion of drying process.

the drying period, both species had dried down to between approximately 50 and 60% of their freshly salted weight. As expected, the smaller fish showed higher drying rates than the larger fish, especially during the first 12 hours of drying.

Salt content, of the flesh, increased to between 30% and 15% dry weight basis (dwb) in *A. thalassinus*, after pickle curing (Table 3) and 21% in *S. commerson*, after brining. Salt concentrations of pickle samples, collected at the end of salting, ranged from as low as 15% (w/v) to saturated values as high as 39% (w/v).

+ Trials conducted Feb.-March 1983.

3.4 Insect species associated with the fish during processing and storage.

Materials and methods

Adult blowflies, visiting the fish during processing, were captured using a small, transparent, polythene bag. This technique proved more successful than either nets or water traps. Specimens were killed in a bottle containing ethyl acetate and then pinned, or preserved in 5% formalin, which was replaced with 70% alcohol, after shipment to the United Kingdom. Blowfly larvae, were collected by hand, relaxed by immersion in boiling water and preserved in 5% formalin. Adult beetles were also hand collected and either preserved in 5% formalin or pinned.

In 1984, eggs and larvae were reared by being placed on a small piece of fresh catfish, which was then introduced to a jar (approximate dimensions 6 x 12 cm), containing a layer of peat, which provided the pupation medium. The jar entrance was covered with fine mesh. The blowflies were reared at ambient temperature and relative humidity and examined each morning.

Results

Tables 4 and 5, show that of the 4 species of *Diptera*, associated with the fish during processing, only 2 species, *C. megacephala* and *L. cuprina*, actually infested the fish.

C. megacephala was the most common blowfly present on the fish during processing, although *L. cuprina* was frequently observed. Ratios of *C. megacephala* : *L. cuprina* , determined from blowfly counts, recorded over a series of 10 trials, conducted between February and March and during July 1984, ranged from a minimum of 11 : 1 to a maximum of 163 : 1, with a mean ratio of 46 : 1.* Although *L. cuprina* was present in smaller numbers than *C. megacephala*, those observed, were invariably, either searching for an oviposition site, or actually ovipositing on the fish. All

*Appendix 2

Table 4. Insect species associated with cured fish in Cirebon, W.Java

Insect species	Sampling location
C. megacephala and *L. cuprina*	Fish landing site. Salting tank containing *A. thalassinus*. Drying *A. thalassinus* and *S. commerson*. Stored *A. thalassinus* and *S. commerson*.
Parasarcophaga misera (Sarcophagidae)	Drying *A. thalassinus* and *S. commerson*.
Musca domestica vicina	Fish landing site. Drying *A. thalassinus* and *S. commerson*. Stored *A. thalassinus* and *S. commerson*.
D. maculatus, *D. carnivorus*, *D. ater*	Stored *A. thalassinus* and *S. commerson*.
Necrobia rufipes (Cleridae)	Stored *A. thalassinus*.

of the *M. domestica* and *P. misera* and most of the *C.megacephala*, present on the drying fish, were feeding and not ovipositing.

The egg and larvae rearing results (Table 5), suggest that *C. megacephala* is the more frequent cause of infestation, although infestation with *L. cuprina* can reach high levels in individual fish (Table 6).

Table 5. Blowfly adults reared from eggs and larvae infesting cured fish at the Cirebon processing site, between February and March 1984.

Culture number	*C. megacephala* Males	*C. megacephala* Females	*L. cuprina*	Sample origin
1.	10	10	0	Larvae on salted *A. thalassinus*.
2.	35	32	0	Eggs on drying *A. thalassinus*.
3.	13	22	0	Eggs on salted *A. thalassinus*.
4.	12	11	0	Larvae on drying *S. commerson*.
5.	15	16	0	Larvae on drying *A. thalassinus*.
6.	15	18	0	"
7.	8	8	0	"
8.	8	7	2	"
9.	9	15	0	"
10.	18	11	0	"
11.	18	11	3	"
12.	2	11	8	"
13.	14	12	0	"
14.	15	13	1	"
15.	6	5	1	"
16.	62	43	0	"
17.	65	31	0	"
18.	27	38	4	"
19.	10	9	0	"
20.	1	2	3	*S. commerson* in store.

Table 6. Infestation details of two individual, whole fish samples of *A. thalassinus*.

Fish weight (g)	No. pupae present	% pupal cases occupied	No. *C. megacephala* reared	No. *L. cuprina* reared
1,022	1,256	13.6*	408	532
836	277	1.4+	7	179

* 171 occupied cases, comprising 46 *C. megacephala* and 125 *L. cuprina*.
+ 40 occupied cases, comprising 1 *C. megacephala* and 39 *L. cuprina*

During the first 2 days of storage, both of the above species were, on occasions, observed to continue to oviposit on the fish. Dermestid beetles (*D. maculatus, D. carnivorus* and *D. ater*) were, however, found to be the most serious cause of infestation during storage.

3.5 Blowfly settling activity during fish processing

Materials and methods

During the initial investigation into the nature of insect infestation, in 1983, 1 tray of fish, from each batch, was photographed at hourly intervals during drying and blowfly counts were taken from the projected slides, on return to the United Kingdom. For subsequent trials, it was found to be more efficient and informative, to make direct, instantaneous counts, of the blowflies that had settled on all of the drying fish of each treatment. Also, during the initial investigation, the total numbers of *C. megacephala* and *L. cuprina* were recorded. During subsequent trials, a record of the sexes of *C. megacephala* present, was also made and counts were taken at more frequent 15 or 30 minute intervals. In addition, the level of blowfly activity on fish in the salting tank was monitored and is discussed in Sections 4 and 7.

Results

C. megacephala activity fluctuated during the day and tended to be higher during the early morning and late afternoon (Figs.4 and 5)*. No correlations between blowfly activity and temperature and relative humidity were apparent. The fish were seen to be most attractive to settling blowflies during the first 2 days of drying. Blowfly activity on the drying *A. thalassinus*, was noticeably higher than over the *S. commerson*, which appeared to be less attractive to blowflies. Blowflies were only occasionally observed to settle on *S. commerson* during drying.

* Heavy fish sampling, for loss assessment purposes, precluded insect infestation monitoring during Trial 2. ∴ results only obtained from Arius Trials 1 and 3.

3.6 Blowfly oviposition on fish during processing.

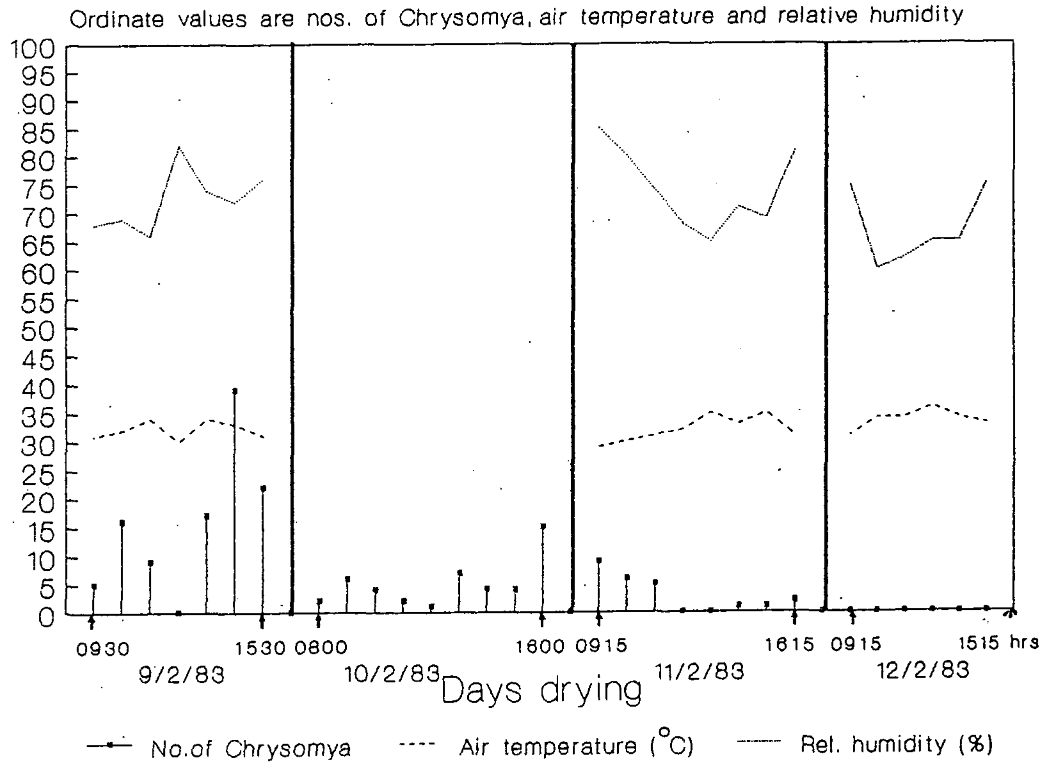
Materials and methods

The nature of the salting process, resulted in the quantitative part of this investigation, being confined to the drying stage of processing. A sample of 5 fish was randomly selected from each batch and at hourly intervals during drying, each fish was closely inspected for the presence of egg batches. To prevent recounting the same eggs, the position of each egg batch was marked with a tag.

Results

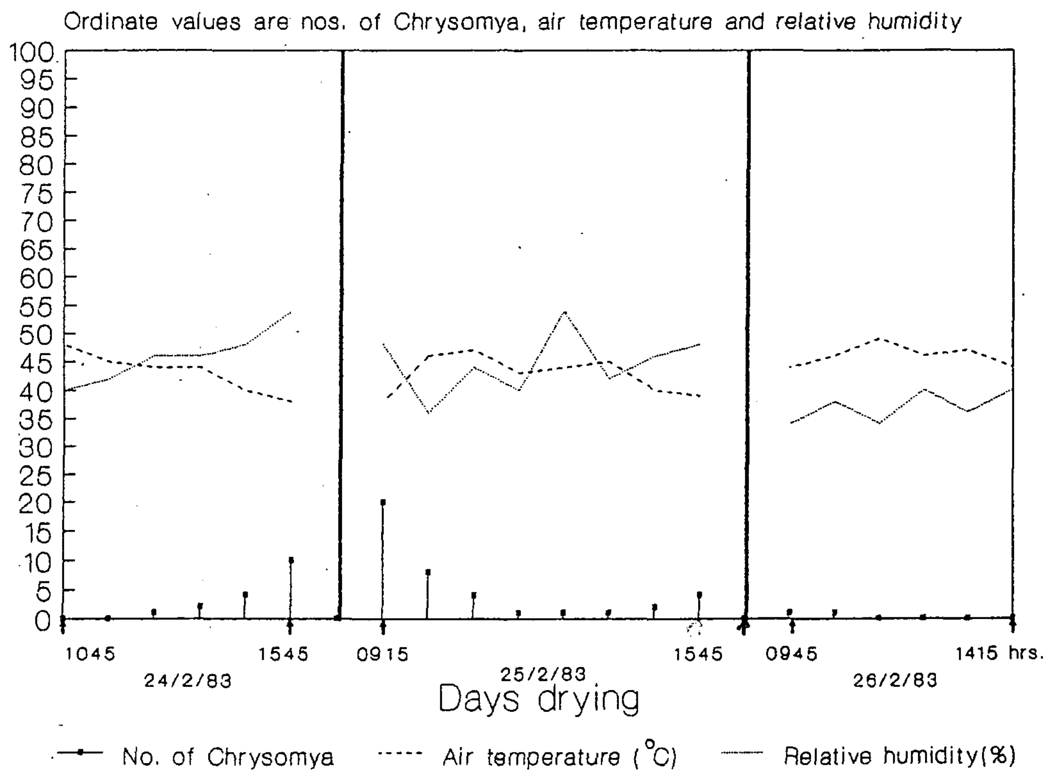
Although *A. thalassinus* was exposed to blowfly attack, both prior to and during, the initial stages of processing, there was no evidence of blowfly oviposition until the fish were being salted. This was the situation in every trial. As the fish were immediately gutted and beheaded on arrival at the processing site and then put straight into water, there was little opportunity for infestation during this stage of the process. However, during the 24 hour immersion period in water, some of the fish were exposed to blowflies, but did not become infested. *C. megacephala* activity, on the fish, became intense during salting, when large numbers of eggs were deposited on the

Fig. 4: Blowfly activity on drying
A. thalassinus, Trial 1, Cirebon, 1983



Instantaneous counts of flies on 5 fish

Fig. 5: Blowfly activity on drying
A. thalassinus, Trial 3, Cirebon, 1983



Instantaneous counts of flies on 5 fish

fish, the salt and the salting tank walls. This suggests that a chemical change occurs to the fish during salting, which makes it highly attractive to ovipositing blowflies. The *S. commerson* was completely submerged in brine and did not attract blowflies during salting, in this particular trial.

Blowfly oviposition continued during the first 2 days of drying (Fig. 6). After this period, during which the fish had dried down to about 70% of their salted weight, the fish became less attractive to ovipositing blowflies, although occasional feeding blowflies were still attracted. The *S. commerson* was relatively unattractive to ovipositing blowfly and only 2 egg batches were deposited on this species during drying. The oviposition behaviour of *C. megacephala* and *L. cuprina* differed, in that *C. megacephala* tended to oviposit on the under surface of the fish, or in the boney orifices at the cut, anterior end, whereas *L. cuprina* inserted the ovipositor into small cavities in the flesh.

The method of overnight storage influenced the susceptibility of the fish to blowfly attack. When the fish were stored down an open side passage, instead of indoors, they were subjected to continued, heavy blowfly attack during the early evening, even though they were covered with a tarpaulin. This was particularly apparent with the Trial 3 *Arius* (Fig. 6), when the morning inspection, on the 2nd drying day, revealed that 25 egg batches had been laid on the fish whilst they were stored along the side passage, overnight.

Under ambient environmental conditions, the eggs of *C. megacephala* and *L. cuprina* hatched within 15 and 12 hours respectively.

3.7 Larval infestation of fish during processing.

Materials and methods

The fish were examined for larvae before gutting, after soaking in freshwater, after salting, at hourly intervals during drying, at the end of drying and at intervals during storage. Larvae were simply recorded as being present or absent, when the fish were individually inspected during processing, before the drying stage. During drying, the fish were individually inspected for blowfly larvae and the level of infestation of each fish was graded as follows:

Grade 1 (zero to light)---larvae absent or very occasional.

Grade 2 (moderate to heavy)---numerous larvae crawling over the fish, or present as feeding groups.

Results

Larval infestation of *A. thalassinus* first became apparent during salting and resulted in between 76 and 100% of the fish becoming infested (Table 7). The salt concentration of the pickles, in which the fish were immersed, ranged from 21.4 to 26.1% (w/v) and contained numerous, active blowfly larvae. Daily monitoring of the fish during drying, demonstrated that infestation became most apparent on the 3rd drying day, when the larvae were large and relatively easy to find (Fig. 7). The larvae remained active within the fish throughout drying, although there was evidence of larvae starting to leave the fish, to pupate, from the 4th drying day onwards. Larval activity, in infested fish, continued during the first few days of storage.

Fig 6: Blowfly oviposition on fish
during drying, Cirebon, Feb.'83

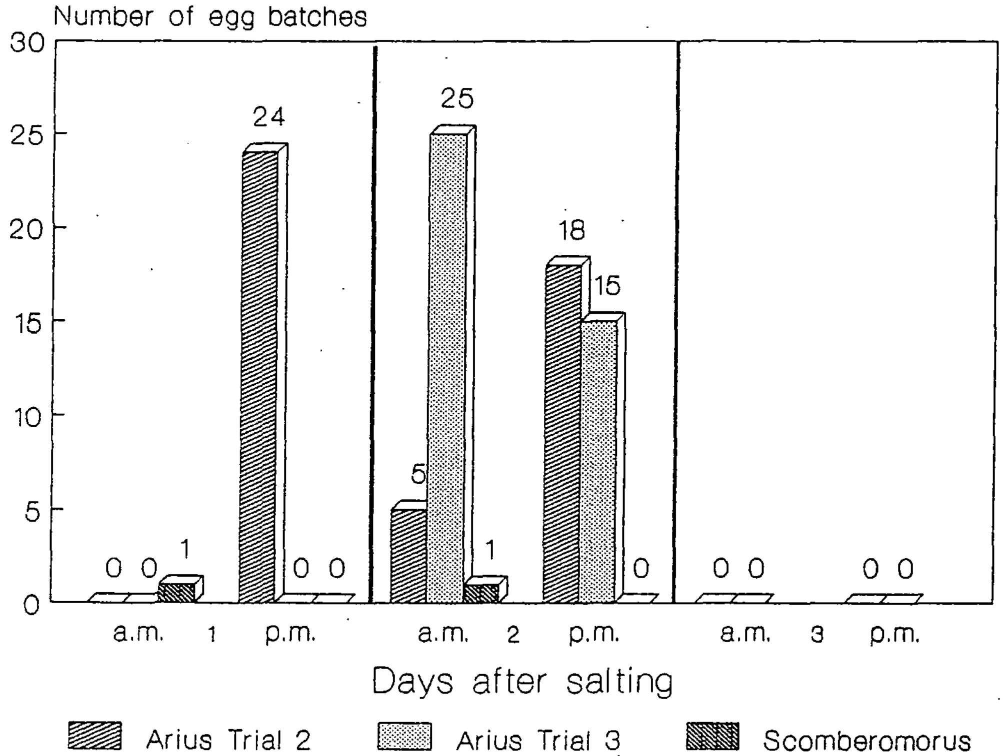
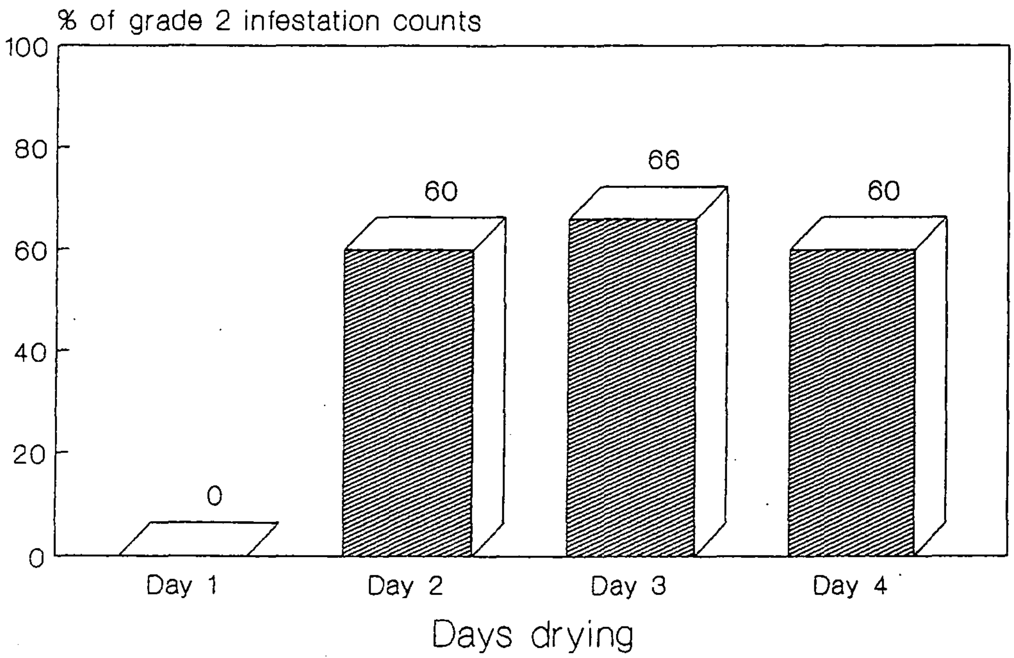


Fig.7: Infestation of *A. thalassinus*
with blowfly larvae during drying.
Cirebon, February, 1983.



Trial 3

Table 7. Timetable of blowfly infestation (% fish infested with larvae) during processing and storage.

Trial	Species	Before processing	After 24 hrs in water	After salting	During storage
1.	*Arius*	0 (40)	0	100	0*
2.	"	0 (44)	0	76	0+
3.	"	0 (45)	0	96	9**
6.	*Scomberomorus*	0 (49)	-	0	31++

* After 10 days storage
\+ After 10 days storage
** After 3 days storage
++ After 5 days storage

Numbers of fish, used in trials, are given in parentheses.

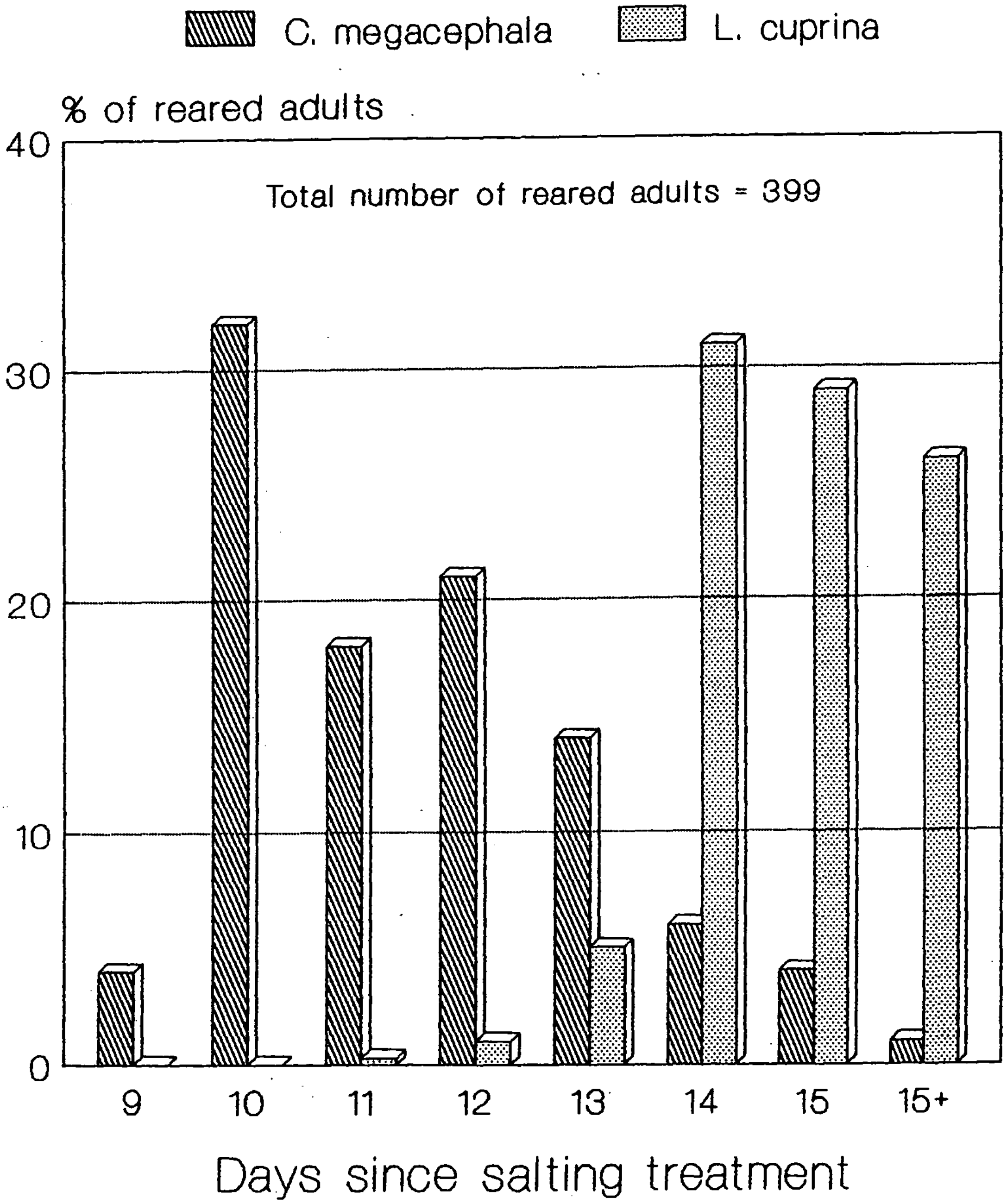
The initial emergences of adults, reared from egg and larvae samples (Fig. 8), indicated that the egg to adult period of *C. megacephala* was 9-10 days, later emergences were probably the result of subsequent oviposition during drying, and that of *L. cuprina* was 11-12 days. These results and field observations, also suggested that infestation with *C. megacephala* was most common during salting and early drying whereas *L. cuprina* frequently continued to infest the fish during early storage.

3.8 Discussion and conclusions

A number of species of *Diptera*, infesting fish during curing, have been identified and most belong to the families Calliphoridae and Sarcophagidae (Walker and Donegan, 1984). Prior to the fieldwork described above, the list of Calliphorid species, identified as pests of cured fish, comprised *Chrysomya albiceps* Wied., *Chrysomya chloropyga putoria* Wied., *Chrysomya regalis* Desvosdy (Meynell, 1978a, McLellen, 1963, Proctor, 1972), *Lucilia* sp. (Ahmed *et al.* 1978) and *Wohlfartia* spp.* (Cole, 1963, Green, 1967). The fieldwork, carried out in Cirebon, identified for the first time, *C. megacephala* and *L. cuprina* as important pests of

*Sarcophagidae

Fig. 8. Emergence periods of adult blow-flies reared from larvae infesting fish during processing and early storage.



Consolidated results of 56 batches of eggs and larvae reared in Cirebon, 1984 salted, dried fish (Esser and Warren, 1983). Subsequent to these findings, *C. megacephala* has been reported to infest cured fish in W. Java (Indriati *et al.*, 1985) and in the PDR Yemen (Sachithananthan *et al.*, 1986). *L. cuprina* has since been reported to infest dried fish in Malawi (Walker and Donegan, 1984).

The dermestid beetles, identified infesting *A. thalassinus* during early storage, are well documented as pests of cured fish (FAO. 1981) and are widespread throughout the tropics.

Settling activity of adult blowflies, could be influenced by a number of factors, including season, climatic conditions, availability of feeding and breeding media and spatial and temporal occurrence of adult emergences. The relatively high blowfly activity, during the first 2 days of drying, is almost certainly due to the high moisture content of the fish, making the fish more attractive to blowflies. The marked preference of the blowflies for *A. thalassinus* over *S. commerson*, could be due to either chemical or physical differences eg texture, or a combination of both. The strong attraction of blowflies to the salting tank and the intense activity of ovipositing blowfly on *A. thalassinus* during salting, suggests that the fish produce a powerful olfactory stimulus during the salting process. The observations of settling activity, on the drying fish (Figs. 4 and 5), indicated a bimodal cycle, activity being higher during the early morning and late afternoon. The hypothesis, that activity increases during the afternoon, was supported by the processor's remarks that he finds it necessary to put his fish away by about 4pm, because of increased blowfly numbers. Additional, more detailed observations, of blowfly settling activity were made during subsequent trials and are discussed further in Section 7.

The observations, that blowfly will readily oviposit on fish, both during and after salting, and that blowfly larvae appeared able to tolerate the relatively high salt concentrations in the pickle and fish, were at variance with previously published opinion (FAO, 1981). Sidaway and Balasingham (1971) reported that blowfly larvae only infested cured fish in Malaysia, before salt penetration. This was clearly not the case in Indonesia. In view of the widespread belief that salted fish were unattractive to blowflies and that salting protected fish against blowfly and beetle infestation, this important observation was further investigated during subsequent field trials and laboratory investigations, which are detailed in later sections of this thesis.

The oviposition sites on the fish, chosen by both species of blowfly, would have reduced the considerable risk of the eggs being desiccated during fish drying.

The field observations of egg incubation period and larval duration of *C. megacephala* corresponded with Wijesundara (1957a) and the findings for *L. cuprina*, corresponded with Mackerras (1933). Egg incubation and larval duration are discussed further in Section 7.

Conclusions

1. Blowfly infestation is a major cause of damage to salted, dried fish in Indonesia.

2. *C. megacephala* and *L. cuprina* infest fish during salting and drying and the eggs and larvae of these species appear to be tolerant of high salt concentrations.

3. Dermestid beetles infest and damage the fish during storage.

4. Processors have resorted to applying unapproved insecticides to their fish, in order to reduce blowfly infestation.

4. FIELD INFESTATION AND LOSS REDUCTION STUDIES CONDUCTED IN WEST JAVA DURING 1984

4.1 Introduction

The fieldwork, described in Sections 2 and 3, identified blowfly infestation as a major cause of damage and losses in salted, dried fish and revealed that fish processors had resorted to applying household insecticides to their fish, in order to control the problem. The need to develop safe, effective techniques of reducing infestation was clearly urgent.

In addition to investigating the nature of insect infestation, during the trials described in Section 3, an attempt was made to obtain a quantitative assessment of the losses of edible, salt free, dry matter, resulting from blowfly infestation, by using the standard, quantitative loss assessment method described in FAO (1981). The results, obtained by this method, indicated losses of 11 to 34%, during processing. This method, however, was subject to a number of constraints, all relating to the large size of the fish, that made the reliable determination of physical losses extremely difficult, under field conditions.

A more practical method of assessing losses of edible solids, during processing, was to compare the weight yield of fish, that had suffered visible blowfly damage during processing, with the yield of fish that had suffered no visible damage (as was usually the case with insecticide treated fish). The fish were weighed after gutting and ranked in order of weight. They were then stratified according to weight and divided into the various groups subjected to the different treatments being assessed. This method resulted in each group of fish having a similar range of sizes and assumed that fish suffering damage, dry at the same rate as undamaged fish. At the end of drying, a visual, subjective assessment was made of the blowfly damage caused to each fish. As it was the level of blowfly infestation and damage and not the generally small, and difficult to determine, physical losses, that influenced the processor's decision to apply household insecticide to his fish, it was felt that these were more appropriate indicators of the efficacy of the loss/infestation reduction methods being evaluated, than the FAO method.

3 potential loss/infestation reduction methods were evaluated, during the course of 2 visits to Indonesia in 1984. These were (i) subjecting the fish to extended salting time, (ii) repairing the walls of the salting tank and fitting a lid that sealed the entrance, followed by using flyscreens during drying, (iii) application of a newly developed, synthetic pyrethroid insecticide, Fastac (active ingredient alphacypermethrin). The trials were carried out under both rainy and dry season conditions, at the Cirebon processing site, described in Section 3.

4.2 Field trials conducted under rainy season conditions, during February and March 1984.

4.2.1 Effects, on blowfly infestation and losses, of subjecting *A. thalassinus*, to an extended salting period during processing.

Materials and methods

39 fish were processed, by the method described in Section 3.2.1. After 24 hours in salt, they were removed from the pickle, split and checked for the presence of blowfly eggs and larvae. The fish were then individually weighed and divided into 2 groups, each group containing a similar weight distribution. The group of fish receiving the normal, 24 hours salting treatment, was returned to the pickle and left for 1 hour, before once more being removed from the pickle, washed and placed out to dry. This group consisted of 19 fish with a mean, gutted weight of 1.96 ±* 0.49 kg. The group receiving extended salting, was returned to the pickle and left for a further 24 hours, before being washed, placed on trays and put out to dry. The trays, containing the different treatments, were placed on the drying racks in random order. This group consisted of 20

* ± s.d. throughout thesis

fish with a mean weight of 1.88 ± 0.59 kg. Pickle samples were taken after 24 and 48 hours, for determination of salt concentration.

Counts of blowflies, present on each group of fish, were taken at 15 minute intervals and temperature and relative humidity were continuously monitored, using a thermohygrograph.

Internal temperatures of 2 fish, were recorded at hourly intervals during drying, by inserting a temperature probe, to a depth of 1 cm, into the flesh of the fish.

At the end of each day's drying, the fish were examined for the presence of blowfly eggs and larvae. The number of egg batches, present on each fish, was recorded and the level of infestation was graded as follows:- Grade 1 (zero to light) = larvae absent or only occasional. Grade 2 (moderate to heavy) = numerous larvae crawling over the fish, or present as feeding groups.

On completion of drying, the fish were weighed and examined for infestation and damage. Damage was graded as follows:- Zero (0) = no damage. Light (L) = occasional, small lesions. Moderate (M) = occasional, large lesions. Heavy (H) = numerous, large lesions.

The design of this trial would have been improved, had the fish, being subjected to the different salting treatments, been purchased on consecutive days. This was the original intention and would have allowed simultaneous drying of both treatments. However, the very erratic fish landings, at the time of trial, dictated the experimental design that was used. Subsequent, extended salting period trials, in which both treatments were allowed to dry simultaneously, are described in Section 5.

Results and discussion

During processing, the fish lost weight, due to dehydration and increased in salt content (Table 8).

Table 8. Changes in fish weight and salt content during processing.

Salting time (hrs)	Mean weight after salt (g)	Mean weight after drying (g)	% weight loss	Salt conc (% dwb)
24	1,960 ± 490	912 ± 270	53	19
48	1,880 ± 560	969 ± 350	48	22

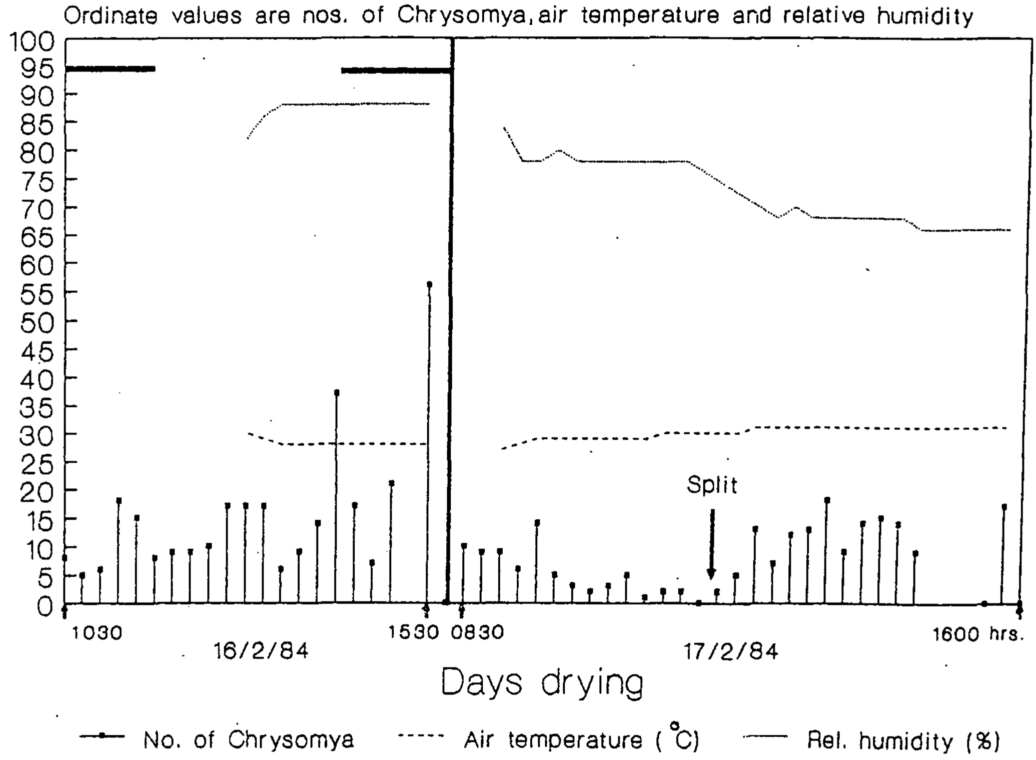
Extended salting treatment, resulted in an average increase in salt concentration of 3% and both figures fall within the range of salt concentrations determined during the trials described in Section 3.

Both groups of fish, dried down to approximately 50% of their salted weight during the drying period, which lasted for 5 days in the case of the 24 hrs salted fish and 4 days with the 48 hrs salted fish. The 1 day difference between the drying periods, was due to the poor drying conditions that prevailed on the day the 24 hrs salted fish were first placed out to dry.

Both *C. megacephala* and *L. cuprina* were attracted to the fish during salting. The mean blowfly counts* for the 1st and 2nd afternoons of salting, were 5.2 and 3.2 respectively. The lower count, during the 2nd afternoon, was probably due to some blowflies being attracted to the drying, 24 hrs. salted fish. Figure 9, shows that blowfly activity, over the 24 hrs. salted fish, was relatively high during the 1st drying day and increased during the course of the afternoon. Drying conditions were poor throughout the day, the relative humidity remained above 80% and the fish lost little moisture. On the 2nd drying day (17/2/84), there was reduced activity over the 24 hrs. salted fish. This can be explained, by some of the flies being attracted onto the 48 hrs. salted fish, when it was put out to dry (Fig. 10). Similar levels of blowfly activity were observed over the 2 groups of fish on February 17th, indicating that extended salting did not impart a repellent effect against blowflies. In order to obtain a better comparison of blowfly activity

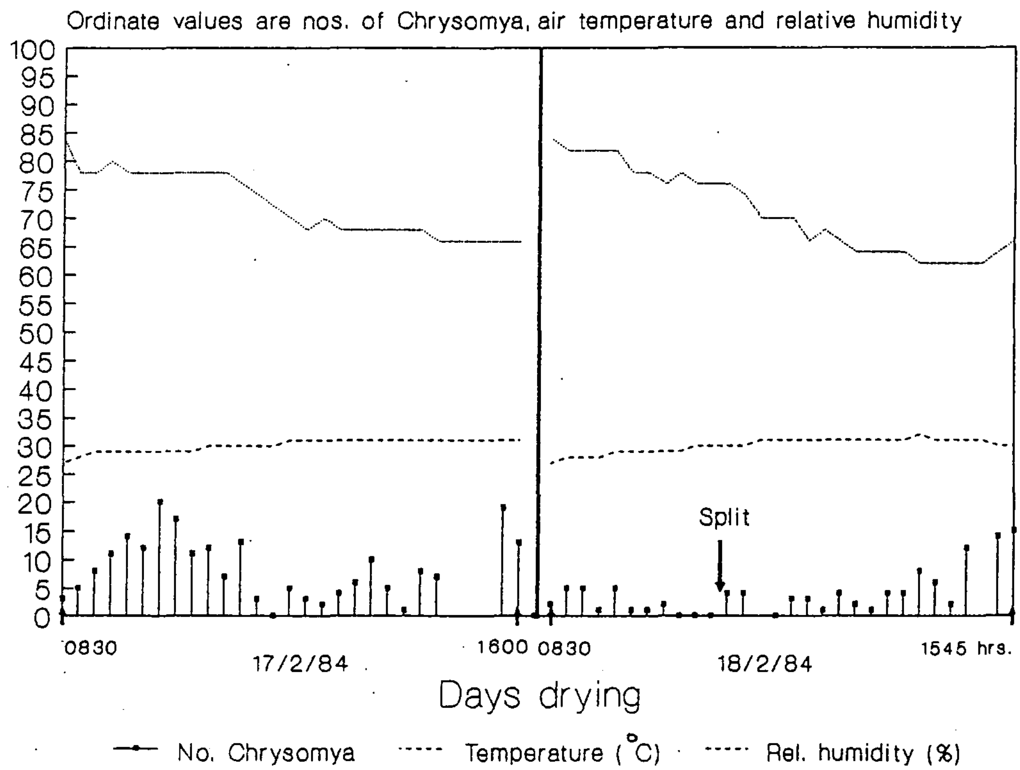
*Mean counts of total number of blowflies on each treatment.

Fig. 9: Blowfly counts on drying *Arius* that had been salted for 24 hours.



Trial at Cirebon site (19 fish)

Fig. 10: Blowfly counts on drying *Arius* that had been salted for 48 hours.



Trial at Cirebon site (20 fish)

on fish subjected to normal and extended salting, the experiment was later modified so that both groups of fish were first placed out to dry on the same day (see Section 4.3.1. for further details).

Similar levels of oviposition occurred, on both groups of fish, during the 1st drying day (Fig. 11). The reduced level of oviposition on the 48 hrs. salted fish, during its 2nd day of drying, could be due to an increase in the proportion of "spent" female flies, resulting from previous exposure to both groups of fish in the salting tank and the 24 hrs. salted fish during drying. It could also be a result of some gravid flies being attracted to another batch of fish, being salted on the same day.

Larval infestation, first became apparent during salting, and the larvae present, were able to tolerate the salt content of the pickle (Table 9).

Table 9. Effect of salting time on salt concentration of pickle and larval infestation during salting.

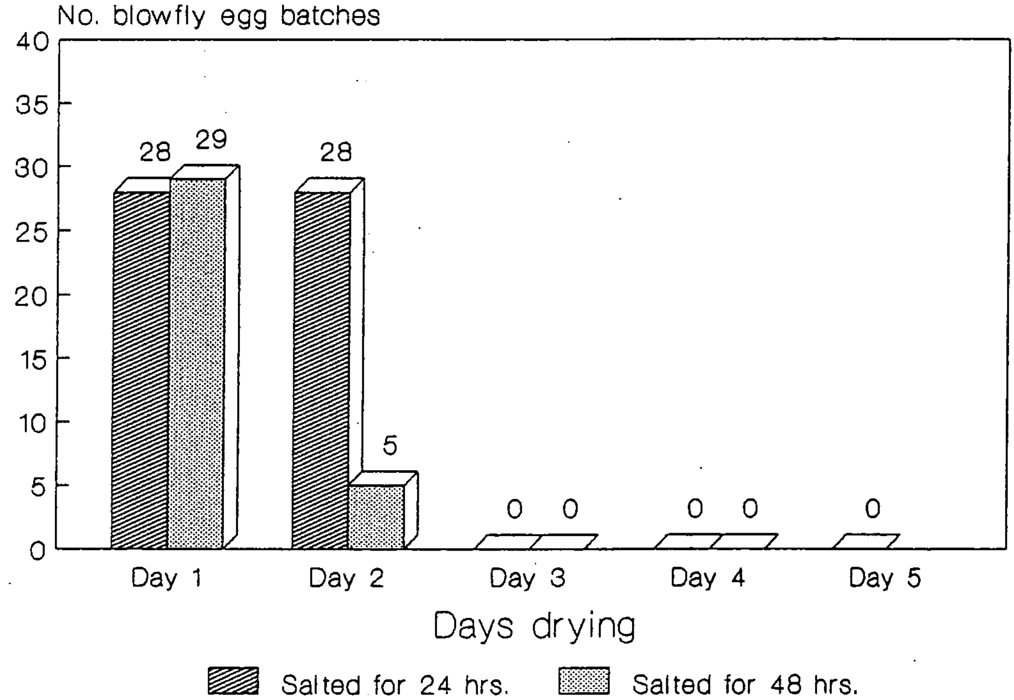
Salting time (hrs.)	Salt content of pickle (%w/v)	% of fish infested
24	24	24*
48	19	48

* 19% of the infested fish were completely submerged in the pickle.

The longer period spent in the salting tank by the 48 hrs. salted fish, resulted in infestation becoming more widespread and, by the end of salting, the incidence of infestation in the 48 hrs. salted fish, was double that of the 24 hrs. salted fish. During the extended salting period, the salt concentration of the pickle decreased by 5%, due to a combination of continued salt uptake and water loss by the fish.

Levels of larval infestation, during drying, were high and similar in both groups of fish (Fig. 12). The

Fig. 11: Effect of salting treatment on blowfly oviposition on *A. thalassinus* during drying. Cirebon site, Feb., 1984

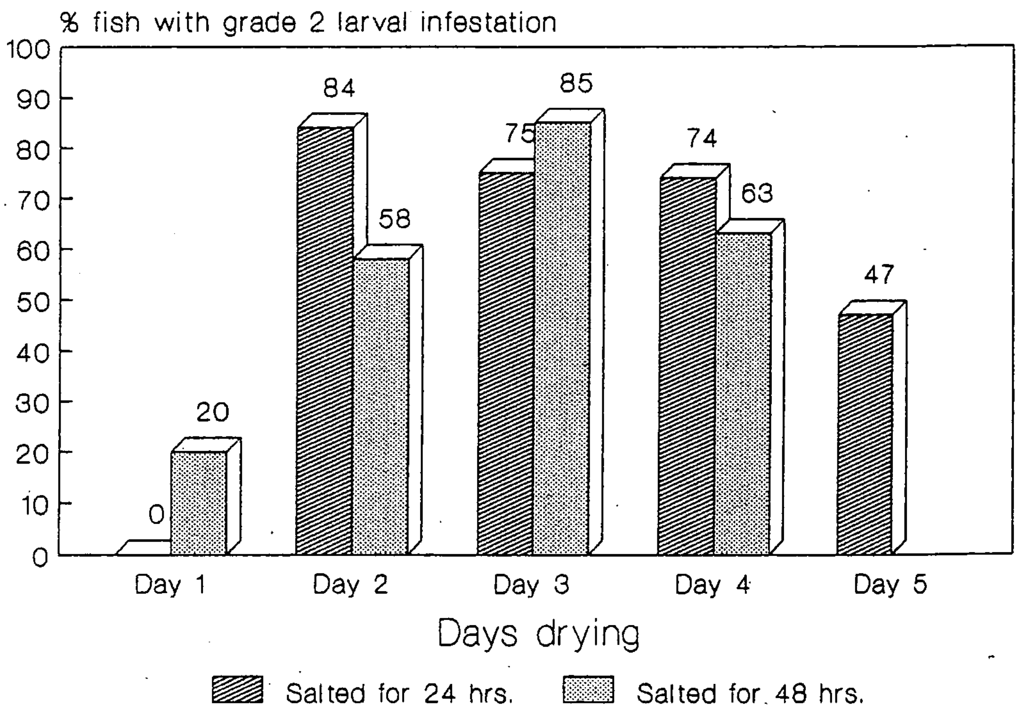


24hrs group=19 fish 48hrs group=20 fish

χ^2 day 1 : 0.02, $p > 0.05$

χ^2 day 2 : 16, $p < 0.01$

Fig. 12: Effect of salting treatment on larval infestation of *A. thalassinus* during drying. Cirebon site, Feb., 1984



24hrs group=19 fish 48hrs group=20 fish

slightly elevated salt concentration of the 48 hrs. salted fish, appeared to offer no protection against infestation. Infestation levels decreased on the last drying day, as the larvae left the fish in order to pupate. The larvae were able to tolerate the elevated internal temperatures of the fish during drying. Mean internal temperatures ranged from 33 to 42°C (Fig.13) during the drying period, and a maximum temperature of 47°C was recorded in one fish (Fig.14).

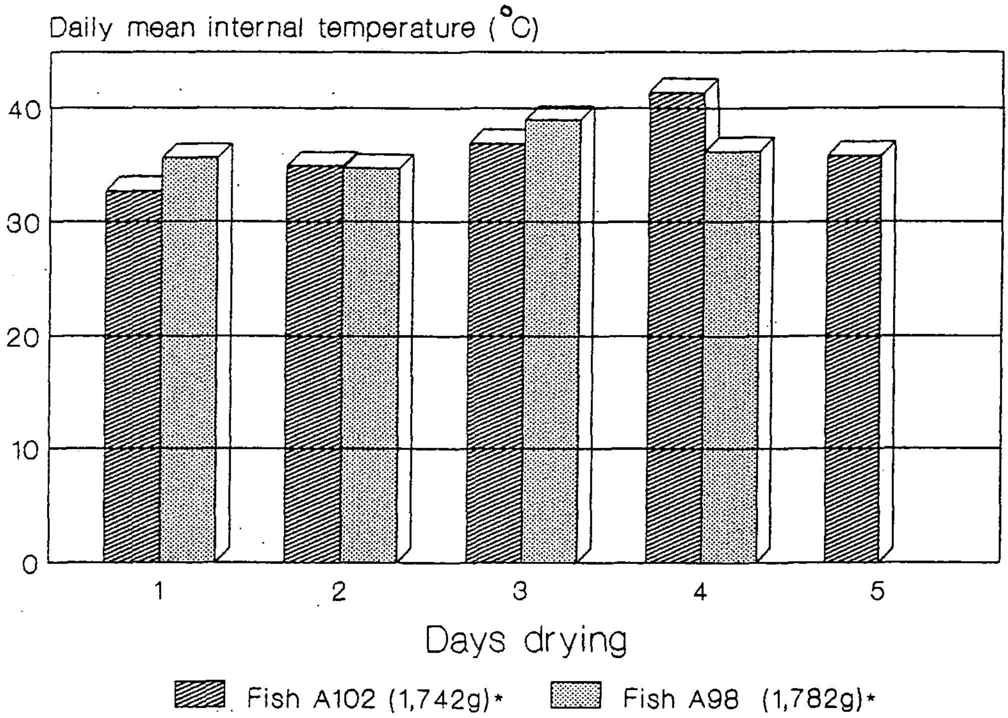
Visual assessment of damage (Table 10), showed that, although the 24 hrs. salted fish exhibited greater damage at the end of drying, by the time the fish had been in store for 3 days, continued consumption of the fish by the larvae present during drying, resulted in a relatively small difference in damage between the 2 groups.

Table 10. Effect of salting treatment on larval damage (% of damaged fish) during processing and early storage.

Salting time (hrs)	Time fish examined	Level of damage			
		Zero	Light	Moderate	High
24	After drying	0	37	21	42
	After 3 days storage	0	11	21	63
48	After drying	30	55	10	5
	After 3 days storage	11	17	56	17

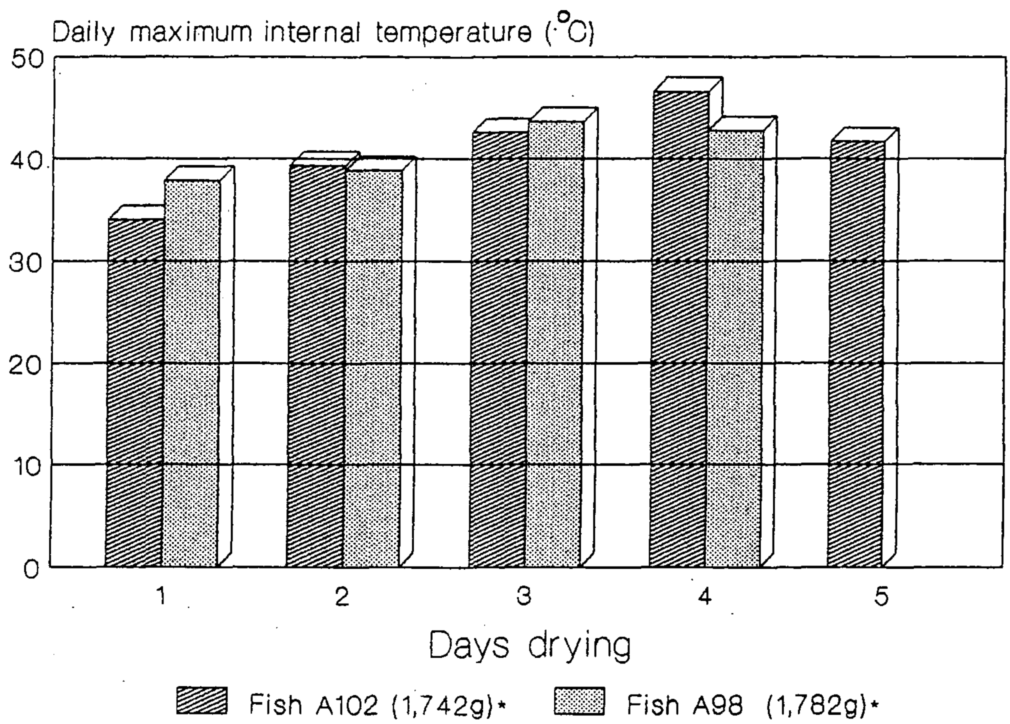
There was no significant difference between the weight losses of the 2 groups of fish (t-test, $p > 0.05$).

Fig.13: Mean internal temperature of *A. thalassinus* on each drying day. Cirebon site, February 1984



* Weight after salting

Fig.14: Maximum internal temperature of *A. thalassinus* on each drying day. Cirebon site, February 1984.



* Weight after salting

4.2.2 Effects of screening on blowfly infestation of *A. thalassinus* during processing.

Materials and methods

One of the 2 salting tanks was modified to prevent blowfly access to the fish during salting. The rough, uneven tank walls were made up with mortar, to give a smooth, level surface. A sturdy, wooden lid was constructed with insteps, so that it fitted the top of the tank (Fig. 15). Strips of foam rubber, with a minimum width of 4 cm, were glued around the edge of the lid, to provide a seal when placed on the tank. The seal was improved by placing heavy stones on the lid, when it was in position. To screen the fish during drying, 3 cages were constructed of locally available materials (Fig. 16).

Three trials*, designed to evaluate the effectiveness of screening during different stages of processing, were carried out and are detailed as follows:

Trial 1

Twenty seven marine catfish were purchased and each fish was weighed, after beheading and gutting. The fish were divided into 2 groups of similar size ranges. 1 group, consisting of 13 fish, was placed into the the modified tank and immersed in fresh water. The entrance to the tank was covered by the lid. The other group, consisting of 14 fish was submerged in water in the open tank. After 24 hours, both groups were salted in the usual way, the control group in the open tank and the experimental group in the sealed tank.

On completion of salting, the experimental group was removed from the sealed tank and each fish examined for the presence of blowfly larvae. The fish were again divided into 2 groups. 1 group, consisting of 7 fish, was placed on trays and received no protection during drying. The other group, consisting of 6 fish was enclosed within the fly cages for the first 2 days of drying. The fish that had been salted in the open salting tank were also inspected for blowfly larvae and divided into 2 groups. 1 group of 10 fish, received no protection during drying and the other group, of 4 fish, was

*For Trial 3, insect infestation was monitored only during salting.

Fig.15. Modifications to salting tank

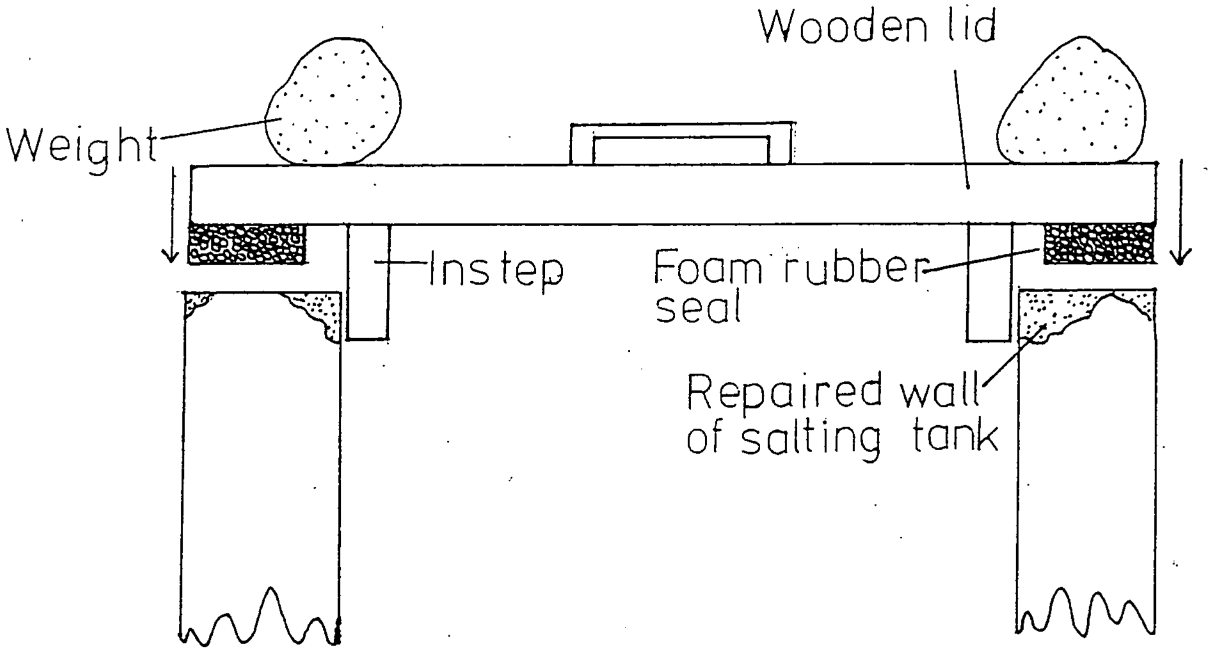
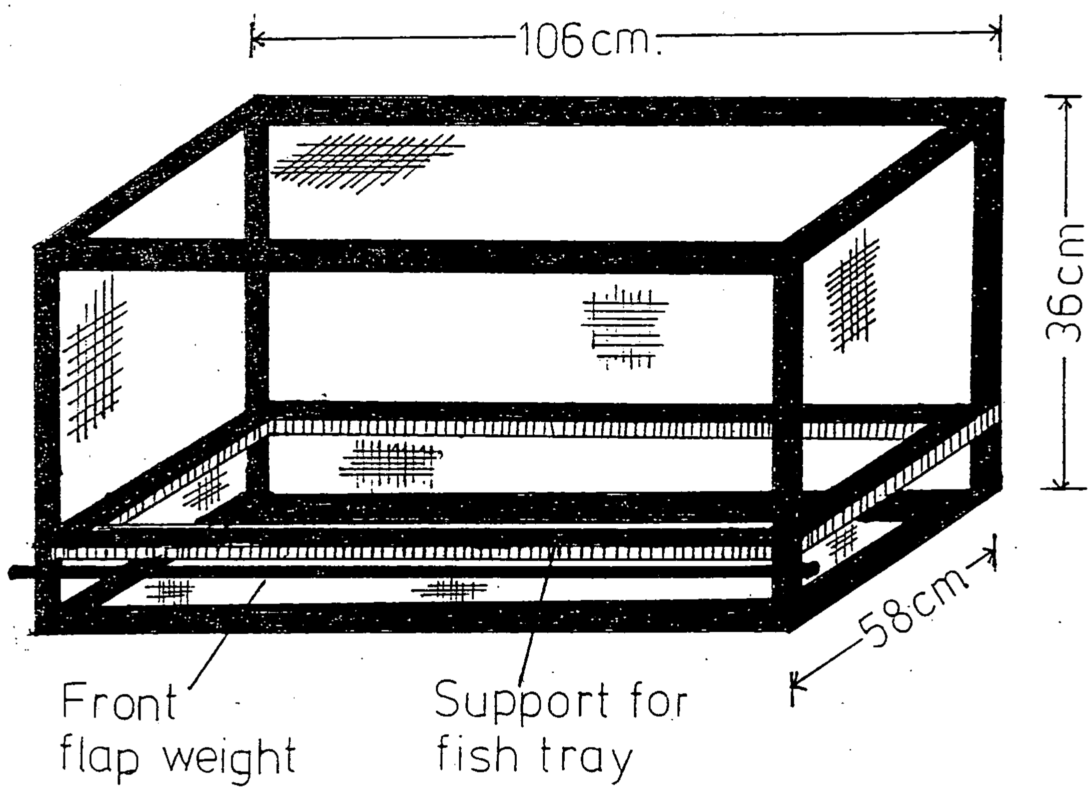


Fig.16. Drying rack screen



placed in the remaining cage, for the first 2 days of drying.

During drying, all fish were monitored for egg batches and larval infestation and at the end of drying the fish were weighed and assessed for damage by blowfly larvae.

Drying rates were monitored by recording the weights of 3 unscreened and 3 screened fish at the start and end of each drying day.

Trial 2

In this trial, 12 marine catfish were beheaded, gutted, weighed and divided into 2 groups of 6 fish, each group having a similar weight range. The control group was processed in the open tank and the experimental group was processed in the sealed tank. After salting, each fish was individually examined for blowfly infestation and 3 of the fish, from the open tank, were then placed on trays and received no protection during drying. The remaining 3 fish were placed in a flyscreen before drying. The fish from the sealed tank were also divided into 2 equal groups, 1 group received no protection during drying and the other group was enclosed in a flyscreen.

During drying, all fish were monitored for egg batches and larval infestation and at the end of drying the fish were weighed and assessed for damage by blowfly larvae.

Trial 3

This trial was carried out to further evaluate the effectiveness of the modified tank and lid in protecting the fish against infestation during salting. 44 marine catfish were dressed, weighed and divided into 2 equal groups of similar size ranges. One group was processed in the open tank and the other in the sealed tank. After salting, each fish was examined for the presence of blowfly larvae.

Results and discussion

It can be seen from Table 11, that infestation during salting, was effectively reduced, by the simple technique of repairing the walls of the salting tank and fitting a lid that provided a good seal.

Table 11: Percentage of fish infested with larvae during salting.

Trial	Fish salted in sealed tank	Fish salted in open tank	χ^2
1.	15	100	6.25**
2.	0	25	0.5
3.	0	100	2.2**

** $p<0.01$

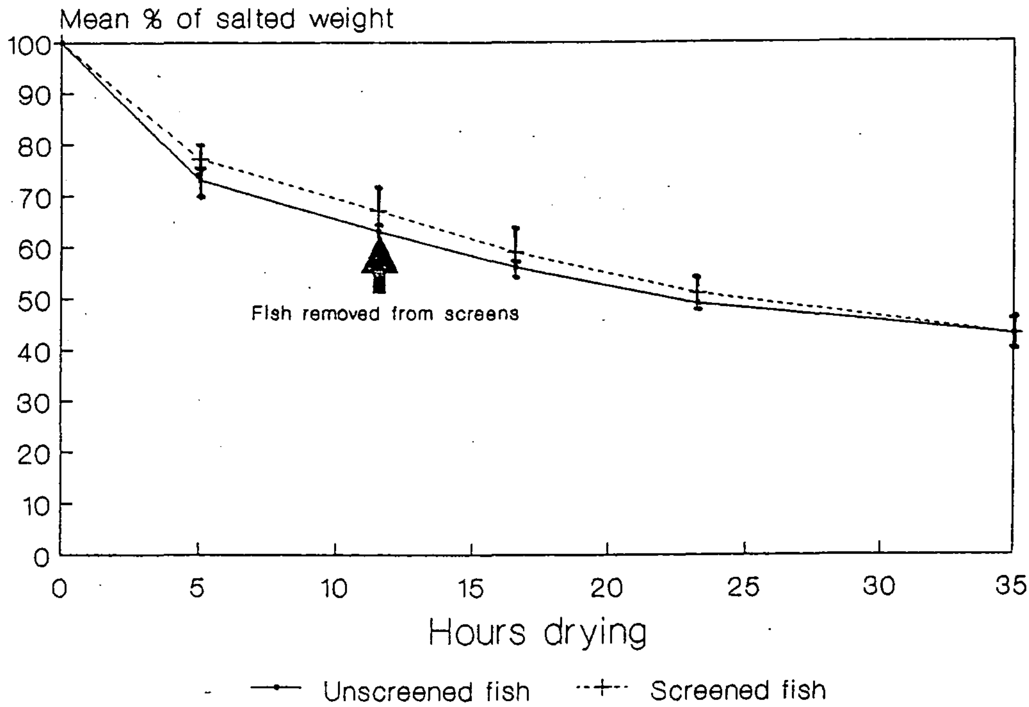
The small amount of infestation of the screened fish in Trial 1, was probably due to larvae having gained access to the fish, through a previously unnoticed, small hole in the bottom of the tank. Sealing this hole prevented any further infestation during salting.

The screens used for the first 2 days of drying, caused a small reduction in drying rates, in both trials (Figs. 17 and 18) and resulted in the screened fish losing 3-4% less weight than the unscreened fish. Removal of the screened fish from the screens, resulted in an increase in their drying rates and, by the end of drying, there was no significant difference between the weight losses of the screened and unscreened fish. Popham (1980), also found that screening, in this case mosquito netting, had no appreciable effect on fish drying rates.

During drying, egg batches were deposited on the unscreened fish in both trials (Figs.19a and 19b). Screening was completely effective in protecting the fish against ovipositing blowfly for the first 2 days of drying, when they were most attractive to blowflies. A single batch of eggs was laid on a fish in Trial 1, after the fish had been removed from the protection of the flyscreens.

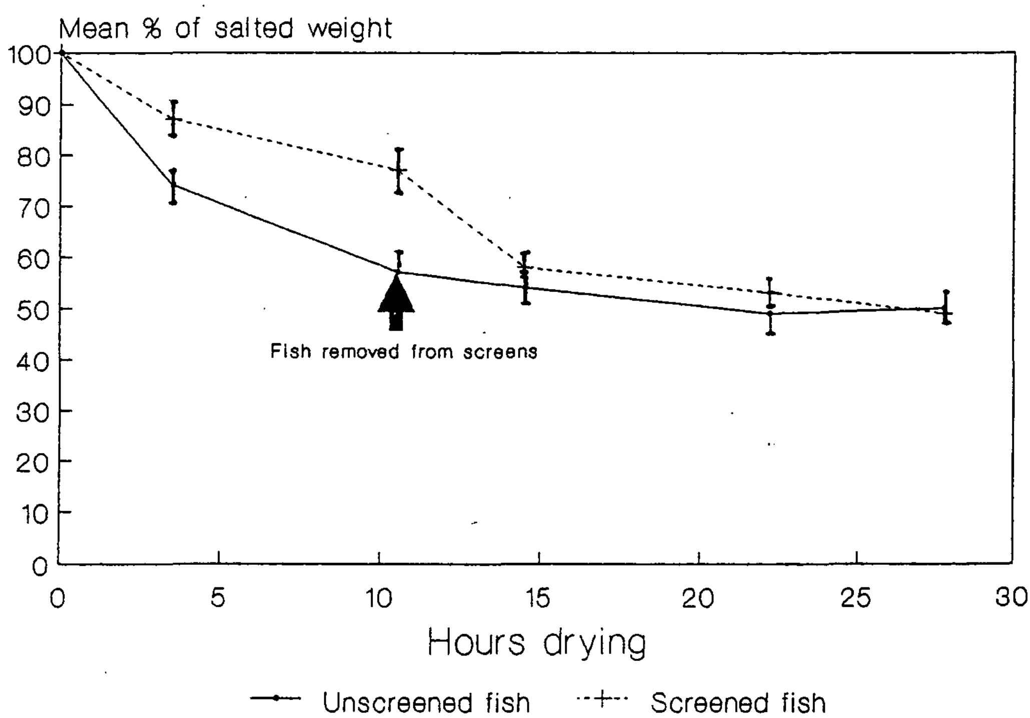
Grade 2 infestation, was apparent on the fish that had been unprotected during salting, by the end of the 1st drying day (Figs.20a and 20c). The larvae had obviously developed from eggs that were deposited on the fish during salting.

Fig.17: Effect of screening on drying rates of *A. thalassinus*. Trial 1, Cirebon, February, 1984



3 fish

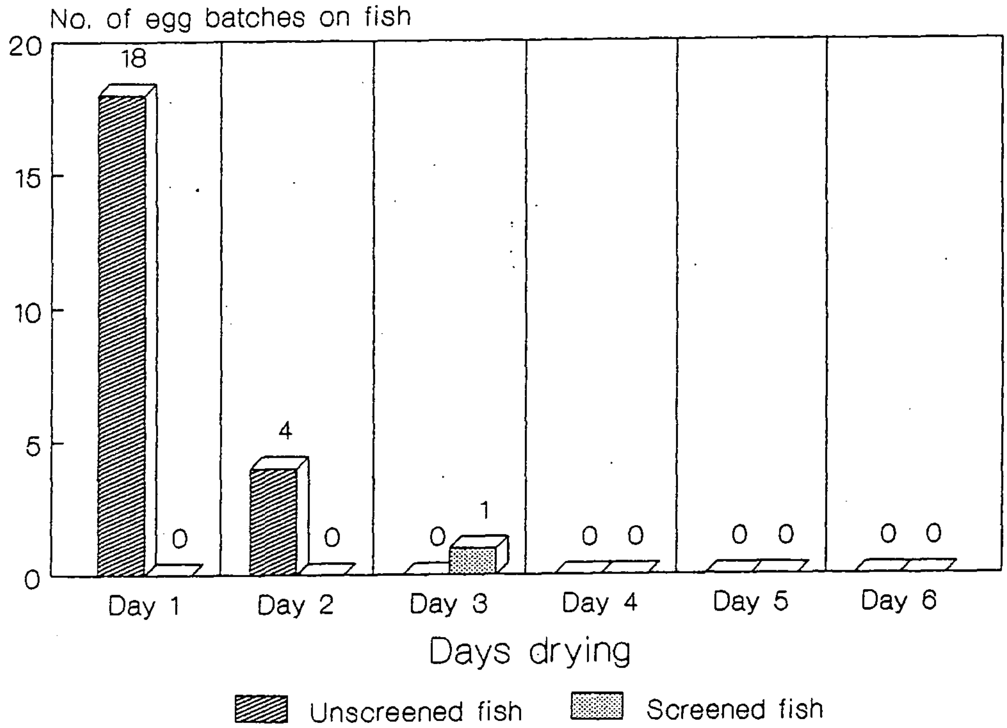
Fig.18: Effect of screening on drying rates of *A. thalassinus*. Trial 2, Cirebon, February, 1984



3 fish

Fig. 19: Effect of screening on blowfly oviposition on *A. thalassinus*, during drying. Cirebon, February, 1984

(a) Trial 1



(b) Trial 2

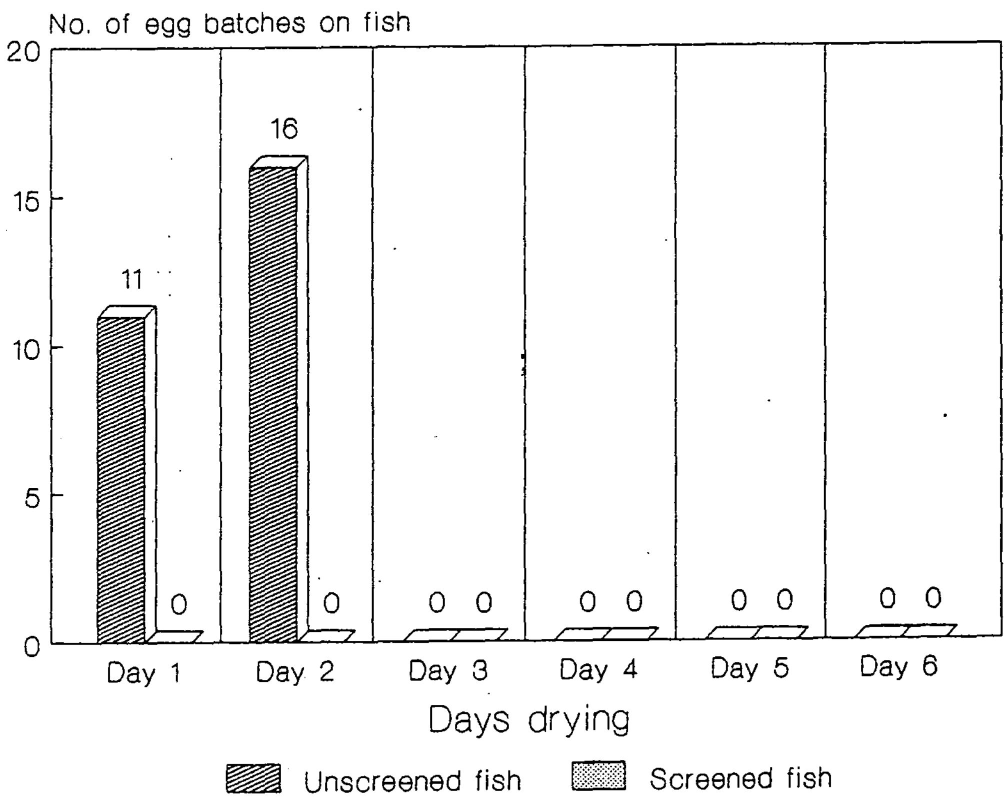
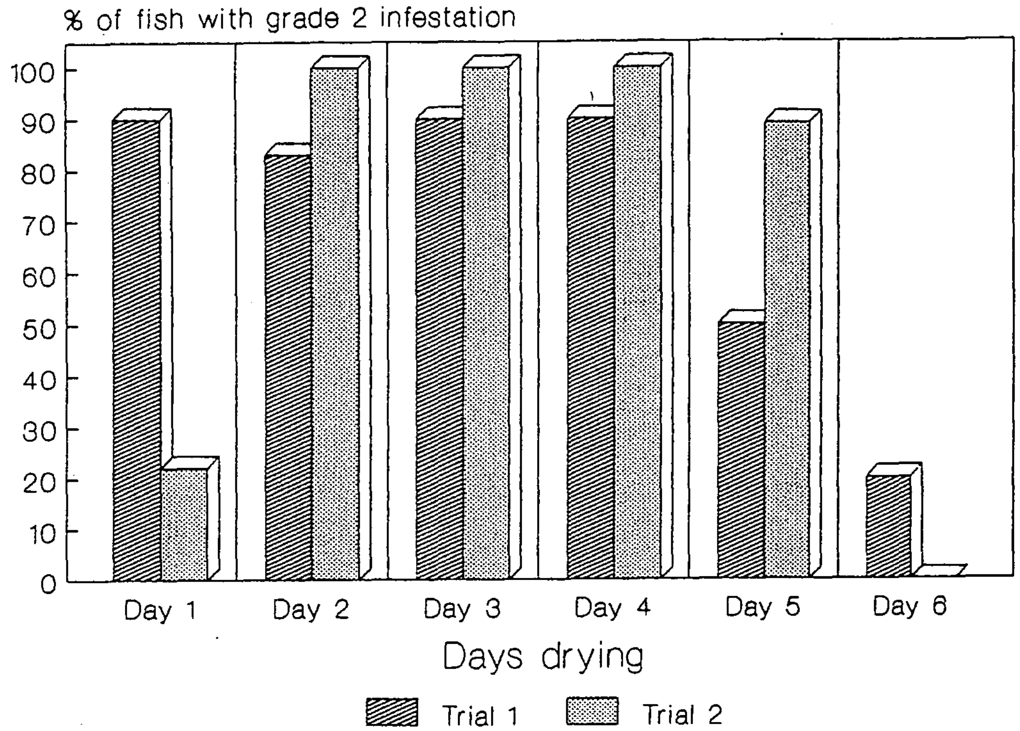
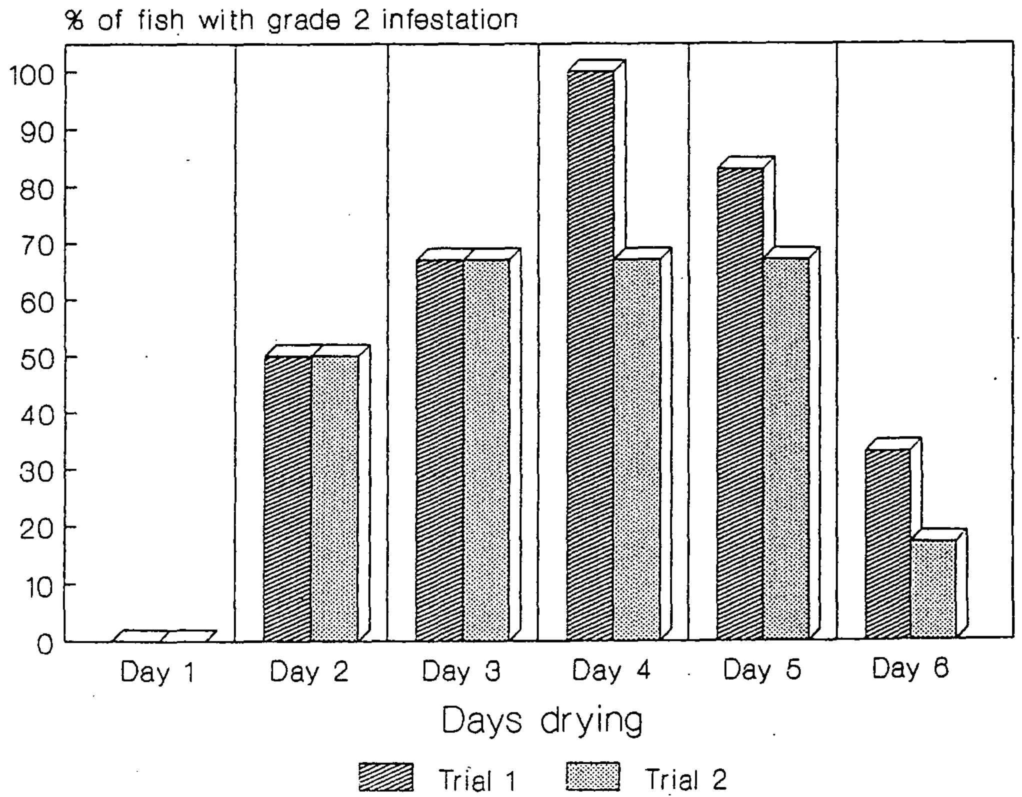


Fig. 20: Effect of screening on larval infestation of *A. thalassinus* during drying. Cirebon, February, 1984

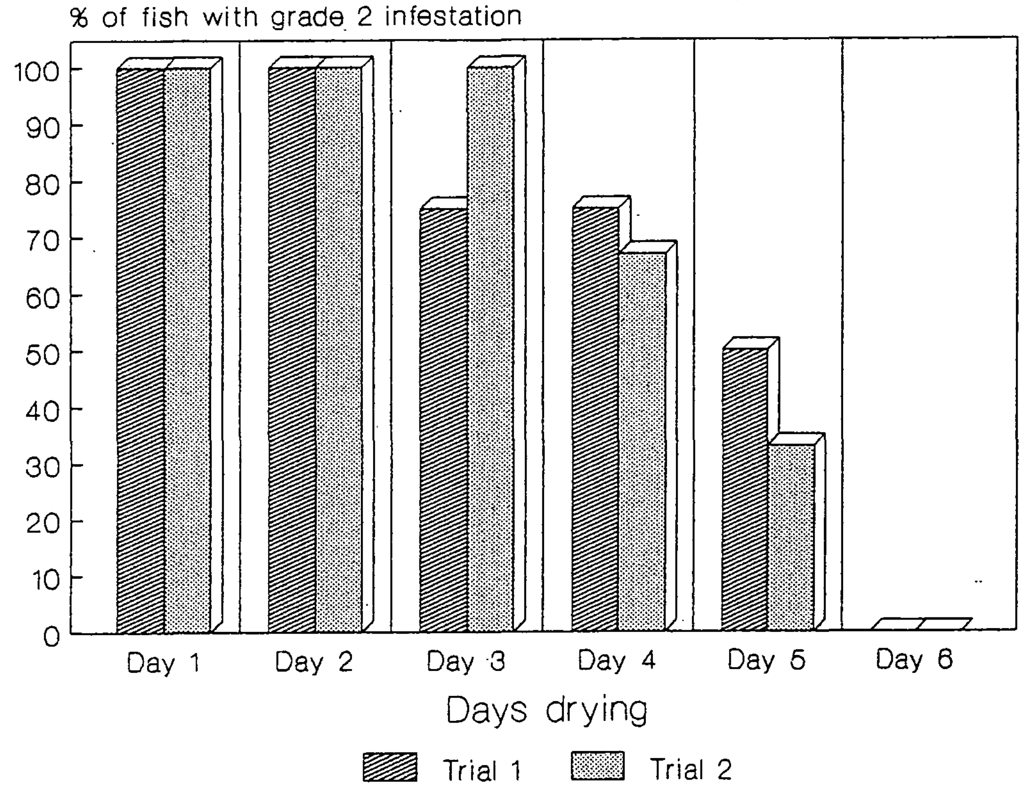
(a) Fish unscreened throughout process



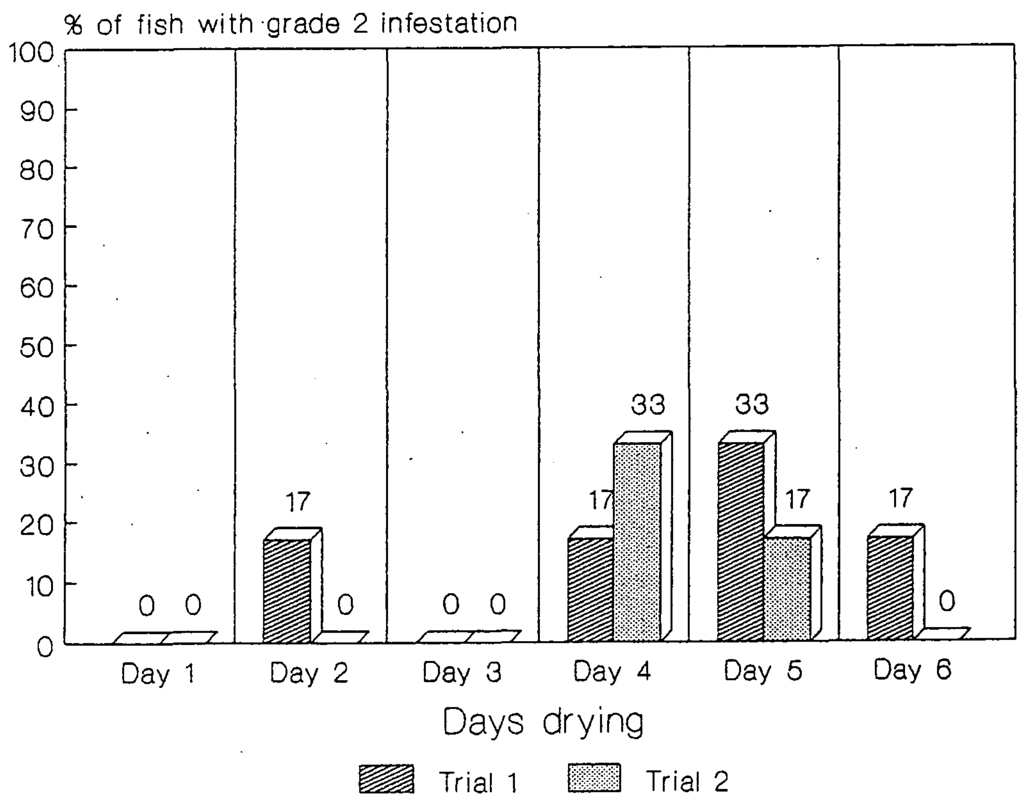
(b) Fish screened during salting only



(c) Fish screened during drying only



(d) Fish screened throughout process



Grade 2 infestation, of the fish protected during salting only, became apparent on the second drying day (Fig.20b). The larvae, in this case, will have developed from eggs deposited during the first drying day.

Infestation became most apparent 3 days after salting and decreased by the fifth day, as the larvae left the fish to pupate. Screening during salting and drying, effectively reduced infestation during processing, although blowflies did manage to obtain access to 1 of the screened fish, resulting in some infestation (Fig.20d).

The results of these trials, clearly demonstrated, that screening for only part of the process, was ineffective in controlling infestation.

Damage resulting from blowfly larvae, reflected the infestation results (Figs.21a to 21d) and demonstrated that the fish must be screened both during salting and for, at least, the first 2 days of drying, for the method to be effective in reducing infestation and damage.

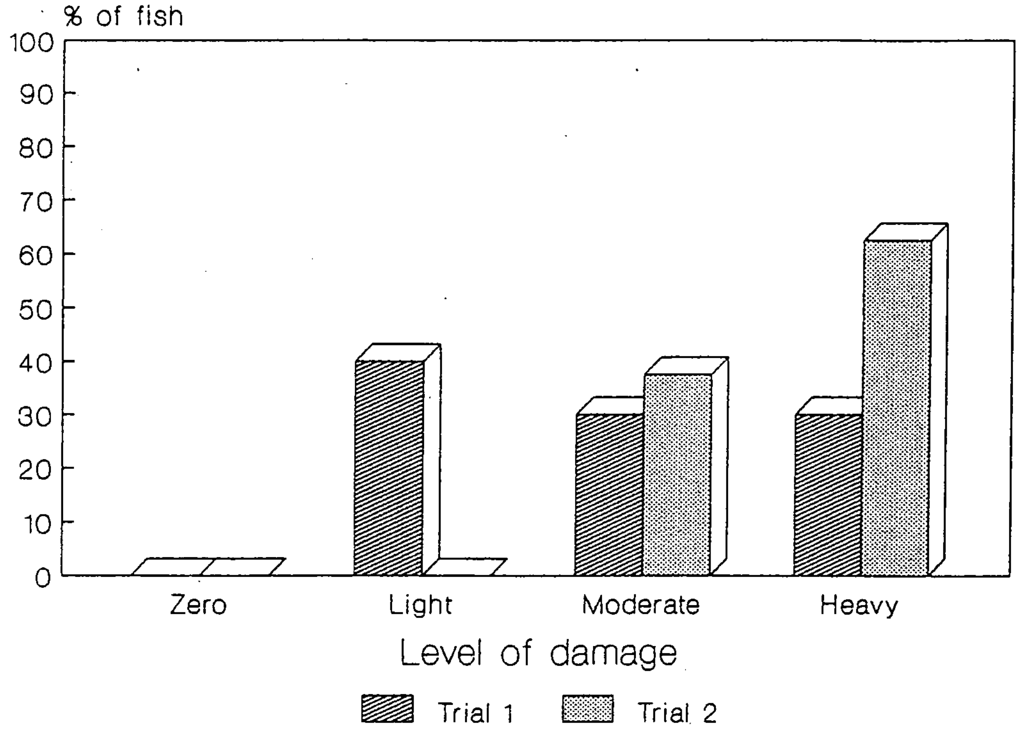
Although screening was demonstrated to be an effective method of reducing infestation and damage, there were some practical problems associated with the method. Good hygiene had to be continually maintained and despite careful washing of the floor, salting tanks and drying trays, there was still some infestation and damage to the fully screened fish. A 2nd and major problem, was associated with the design of the screens, which could only accommodate 1 tray of fish each. The size of the screens led to handling and overnight storage difficulties, which the processor found unacceptable.

The processor did, however, readily adopt the salting tank lid and fabricated a second lid, at his own expense, for use on the other salting tank. During a subsequent visit to Cirebon in 1988, it was found that the processor was continuing to use the same lids.

Screening does work and it is a potentially useful method of controlling insect infestation. Further evaluation, using different flyscreen designs, particularly at sites where permanent screens could be installed, should be carried out.

Fig. 21: The effect of screening on damage caused to *A. thalassinus* by blow-fly larvae. Cirebon, February, 1984

(a) Fish unscreened throughout process



(b) Fish screened during salting only

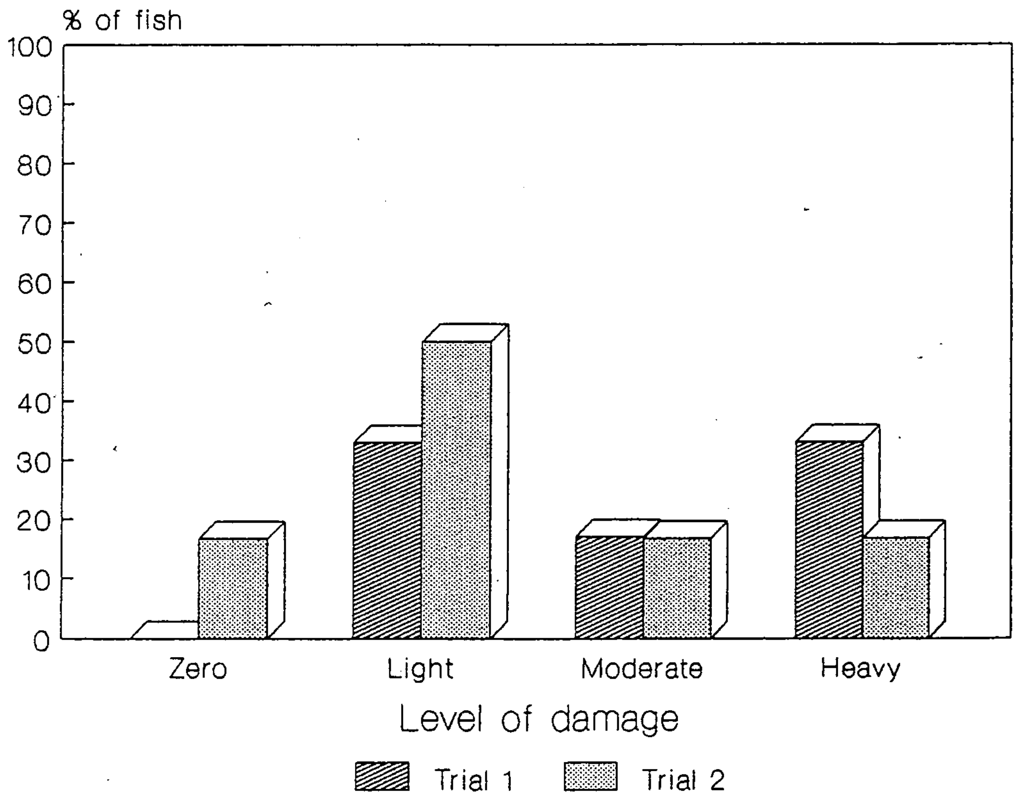
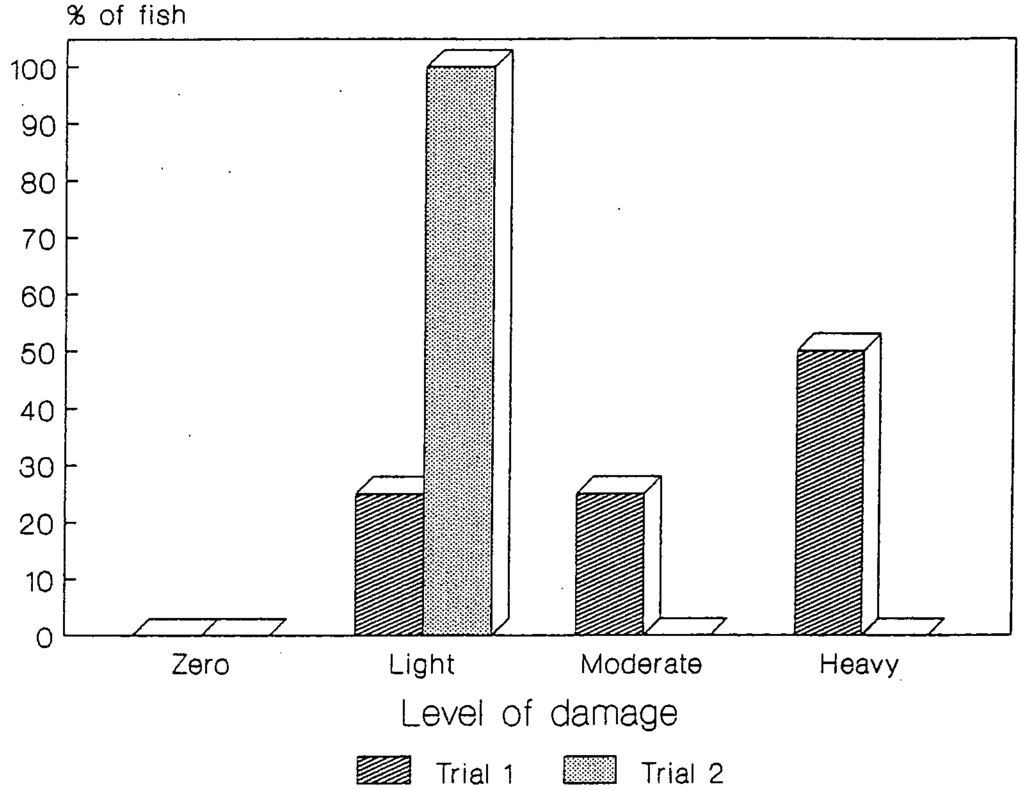
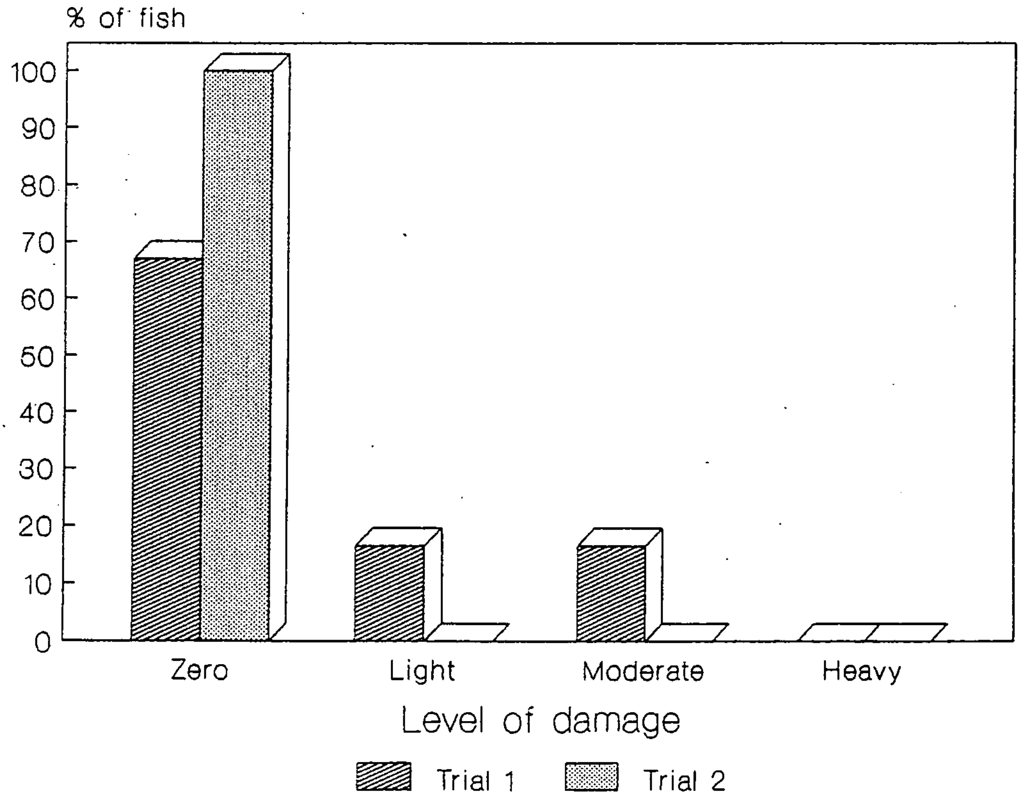


Fig. 21 (contd.)

(c) Fish screened during drying only



(d) Fish screened throughout process



4.2.3. Effects on insect infestation during processing and storage, of treating *A. thalassinus* with the pyrethroid insecticide, Fastac.

Materials and methods

A series of 5 trials, evaluating the effectiveness of a range of concentrations of Fastac, in controlling insect infestation of *A. thalassinus* during processing and storage was carried out at the processing site in Cirebon. All of the fish, used in the trials, were processed using the method described in Section 3.2, and divided into groups of similar size ranges. During processing, environmental conditions, blowfly activity, oviposition and, larval infestation and damage, were monitored using the methods described in Section 4.2.1.

The insecticide used, was the recently developed synthetic pyrethroid insecticide, Fastac, (1Rcis)S and (1Scis)R alpha-cyano-3-phenoxybenzyl 3-(2,2-dichlorovinyl)-2,2-dimethyl-cyclopropanecarboxylate, supplied as a suspension in xylene, by P.T. Shell Indonesia.

Treatment consisted of dipping each fish in a 6 litre, aqueous emulsion of diluted Fastac, at a range of different dilutions, after salting and splitting. To ensure complete coverage, the fish were completely submerged in the insecticide, for 2 periods, each of 3 seconds duration. The control fish were dipped, for the same period, in water. There was 1 exception to this method of insecticide application. In Trial 5, 1 group of fish was subjected to a spray treatment of Fastac, instead of being dipped.

The fish, subjected to different treatments, were placed on separate trays and put out to dry. The trays were arranged in random blocks on the racks, each block of trays containing fish of the same treatment. To prevent the strong repellent effect of Fastac from interfering with blowfly activity on the control fish, a distance of at least 0.5m separated the different treatments.

When the fish were split, usually on the 2nd drying day, the newly exposed surfaces were sprayed with Fastac, of the same concentration as the earlier dip. The 2 squirts of insecticide spray, applied to each fish, amounted to approximately 4ml. Details of concentrations tested and the numbers and weights of fish used in each trial, are given in Table 12.

Table 12: Summary of treatments used in Fastac evaluation trials.

Trial	Treatment	Mean gutted weight (g)	Number of fish
1	0.05% dip	2,983	8
	Control	2,756	6
2	0.025% dip	1598	10
	Control	1604	10
3	0.0125% dip	1915	10
	Control	1903	10
4	0.00625% dip	1575	10
	Control	1571	10
5	0.003% dip	-	6
	0.002% dip	-	6
	0.001% dip	-	6
	0.002% spray	-	6
	Control	-	6

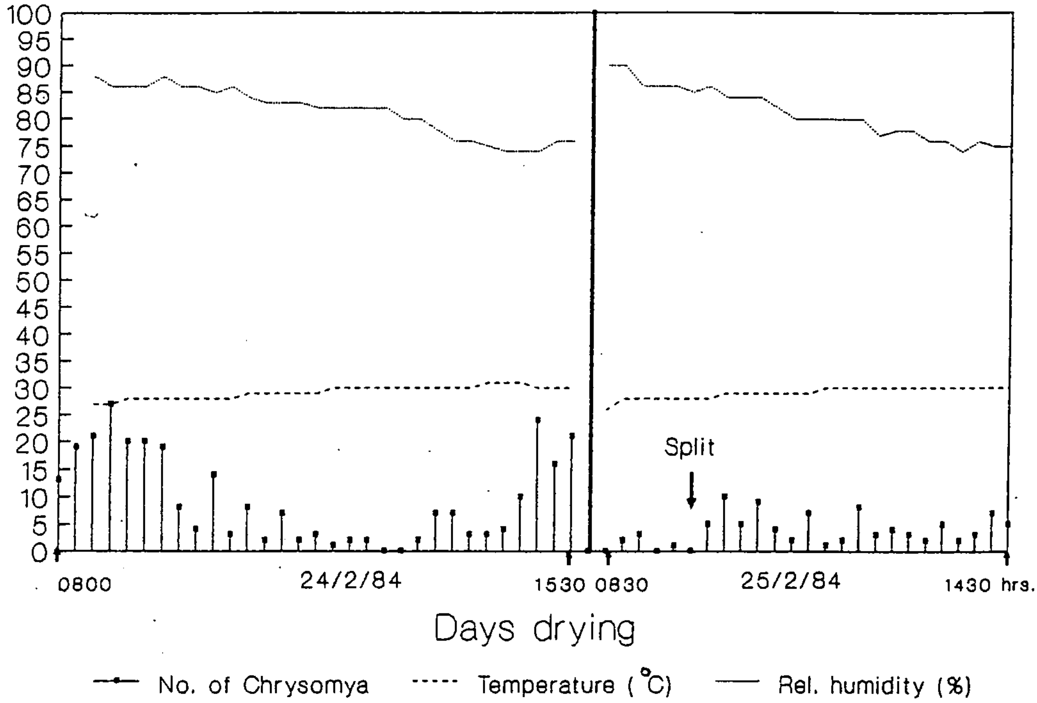
Results and discussion

Figures 22-25, show that Fastac had a strong repellent effect against blowfly adults, over the concentration range used. This effect was most apparent during Trial 2, when activity over the control fish was very high (Fig. 23). No blowfly activity figures are given for Trial 5, as activity was very low over the fish. Only occasional blowflies were observed on both the control and

Fig. 22: Effect of insecticide on blowfly activity (instantaneous counts of flies on fish, at 15 min. intervals)

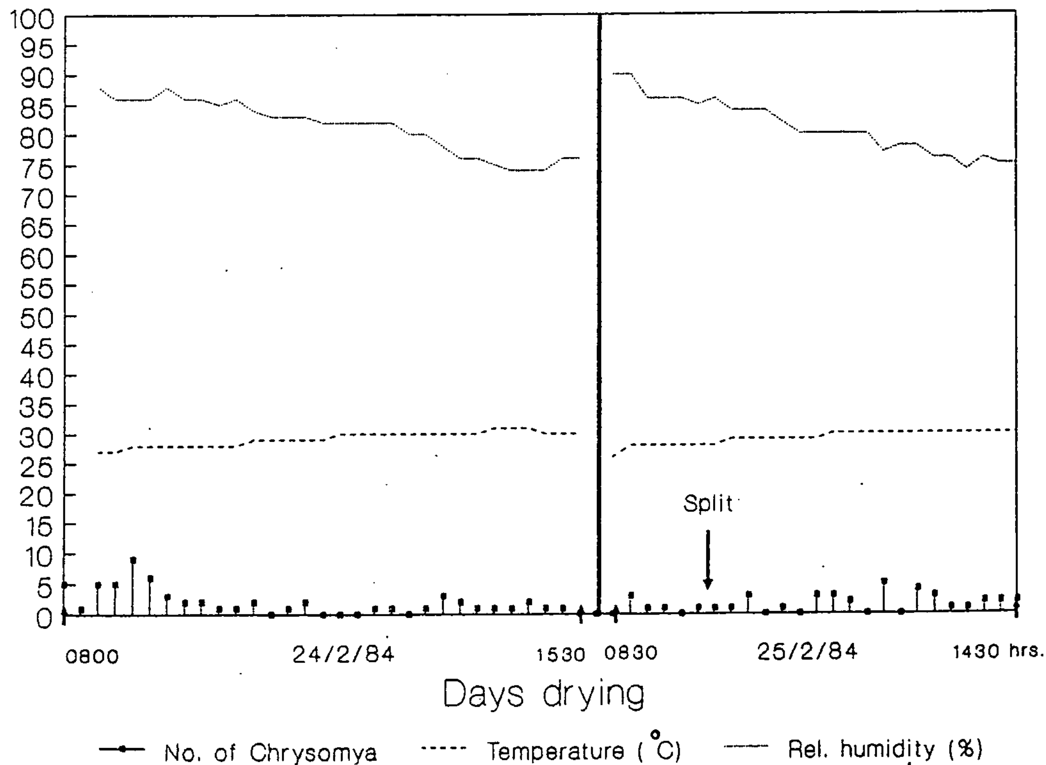
(a) Control fish(**TRIAL1**)

Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Trial at Cirebon site, February, 1984

(b) Fish dipped in 0.05% Fastac



<u>mean count</u>

(a) 6·5

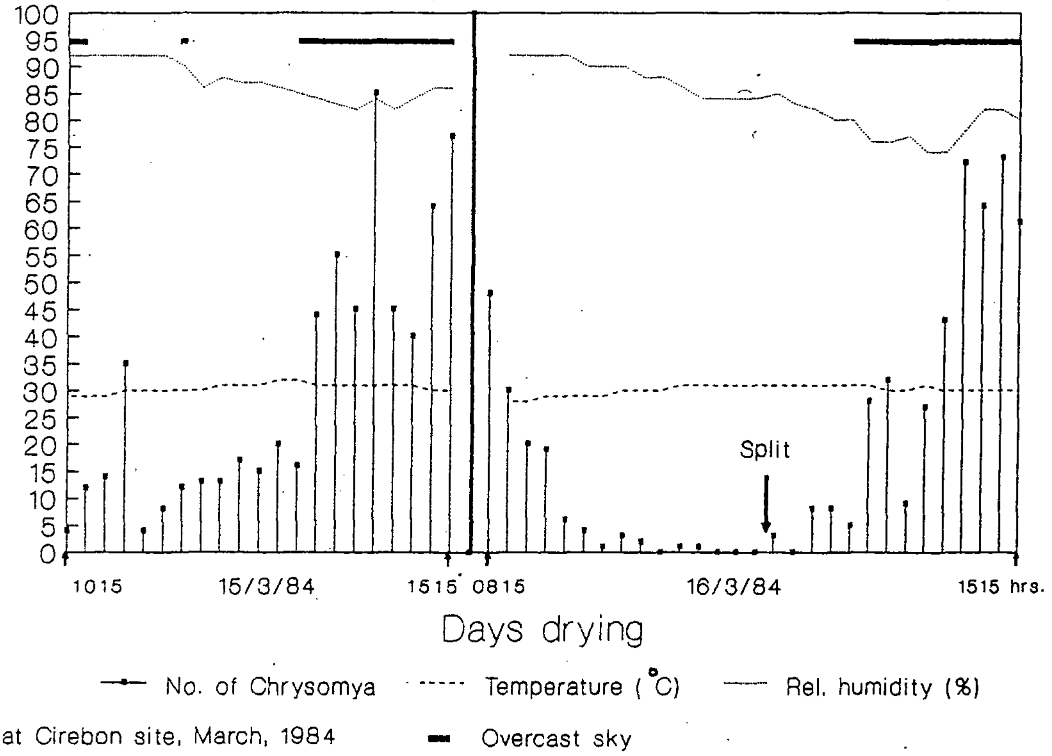
(b) 1·8

$t = 5{\cdot}9$, $p < 0{\cdot}001$

Fig. 23: Effect of insecticide on blowfly activity (instantaneous counts of flies on fish, at 15 min. intervals)

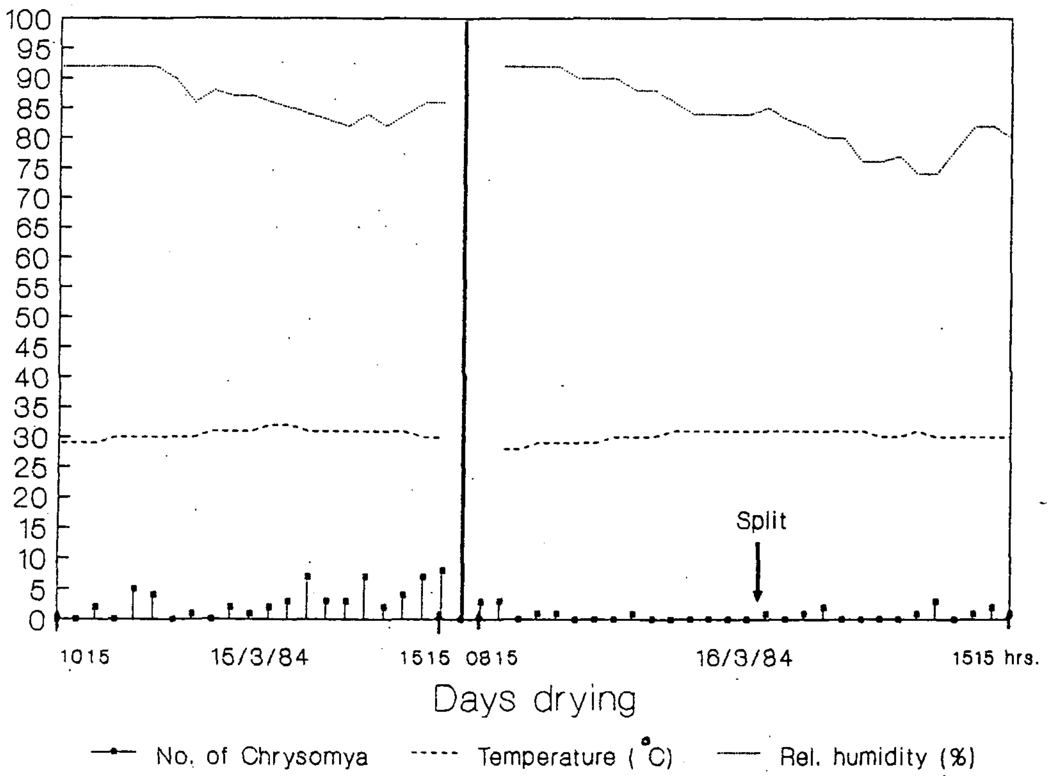
(a) Control fish **(TRIAL 2)**

Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Trial at Cirebon site, March, 1984

(b) Fish dipped in 0.025% Fastac



<u>Mean count</u>

(a) 23.6

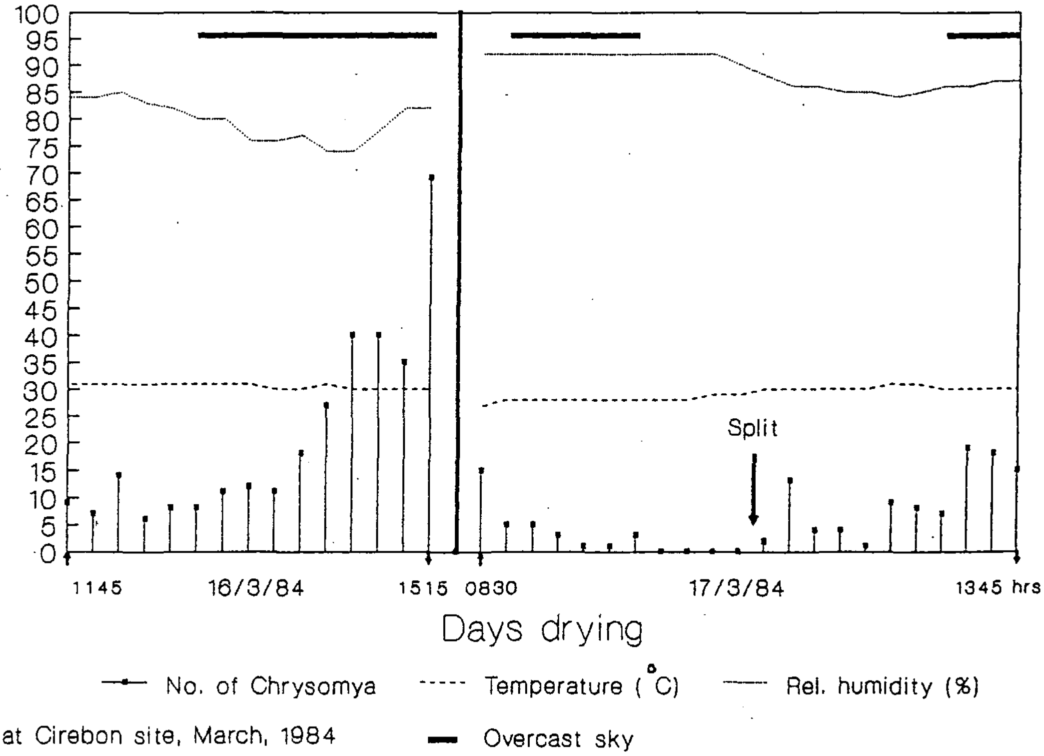
(b) 1.6

$t = 6.7, p < 0.001$

Fig. 24: Effect of insecticide on blowfly activity (instantaneous counts of flies on fish, at 15 min. intervals)

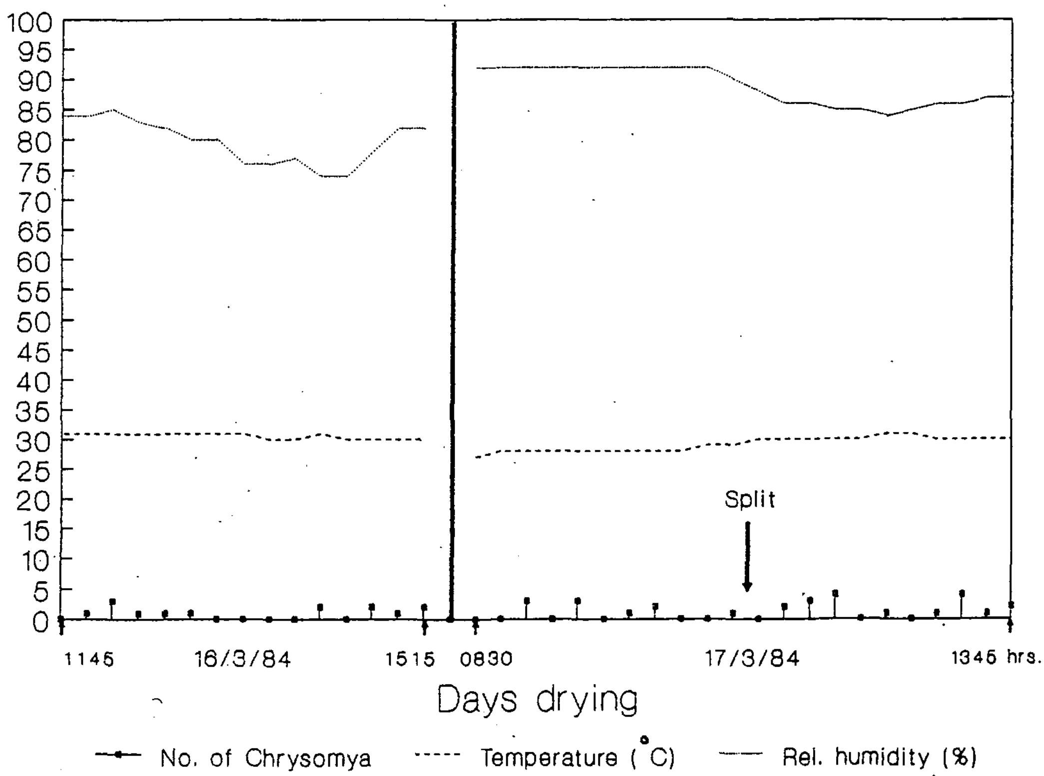
(a) Control fish **(TRIAL 3)**

Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Trial at Cirebon site, March, 1984

(b) Fish dipped in 0.0125% Fastac



<u>Mean count</u>

(a) 11·8

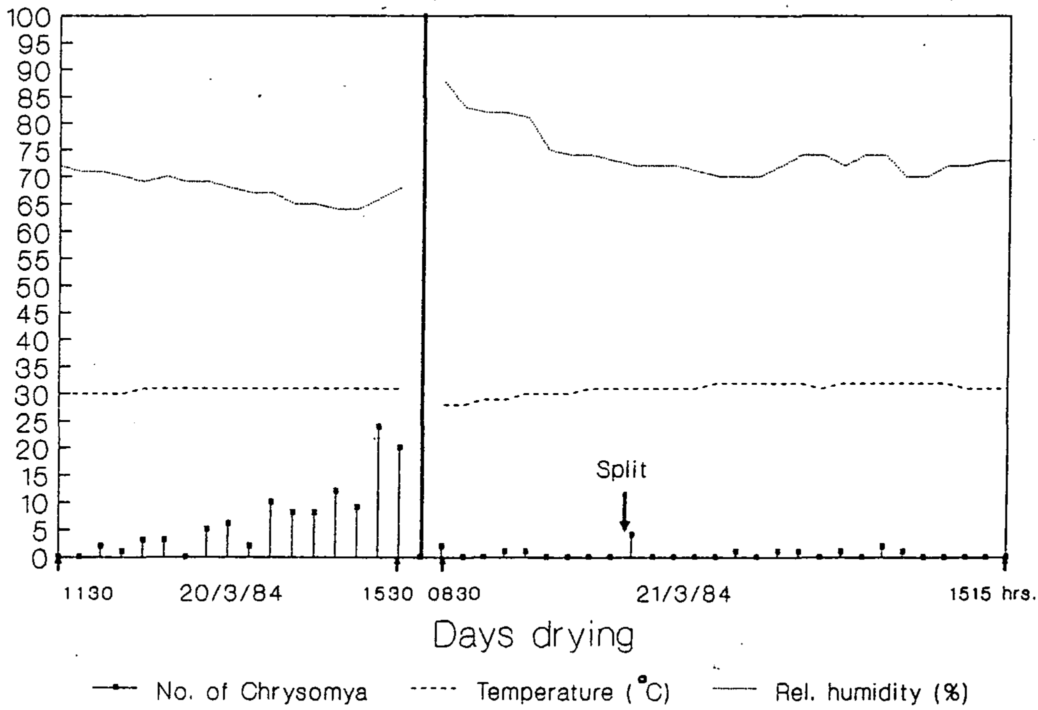
(b) 1·1

t = 4·7, p<0·001

Fig. 25: Effect of insecticide on blowfly activity (instantaneous counts of flies on fish, at 15 min. intervals)

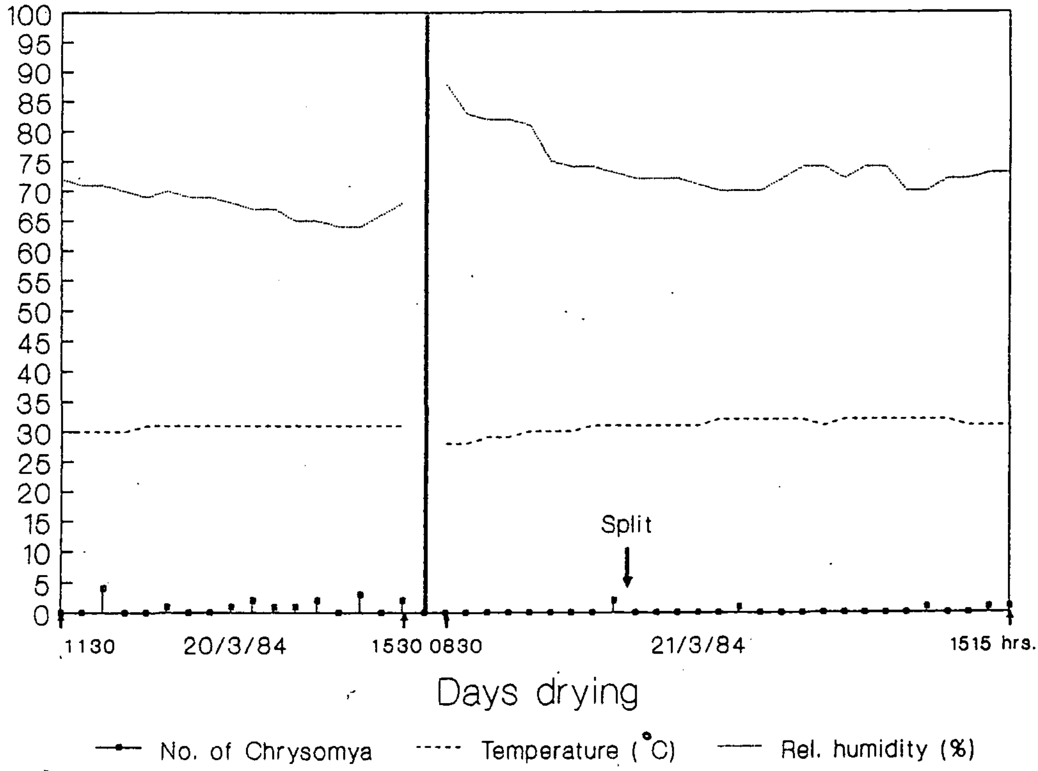
(a) Control fish **(TRIAL 4)**

Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Trial at Cirebon site, March, 1984

(b) Fish dipped in 0.00625% Fastac



Mean count
(a) 2·78
(b) 0·006
$t = 3.1$, $p < 0.05$

insecticide treated fish and numbers were too low to demonstrate any repellent effect of the Fastac treatment.

The effects of treatment with Fastac, on oviposition and larval infestation and damage, are summarised in Table 13. Fastac was effective in reducing oviposition on the fish, when applied, as a dip, at concentrations of 0.05% to 0.00625%. No grade 2 larval infestation was present in fish treated with Fastac, at concentrations of between 0.05% and 0.003%. Grade 2 infestation was, however, apparent in fish treated with concentrations of 0.002% and 0.001%. Fastac was clearly demonstrated to be larvicidal. Fish, which were heavily infested during salting, showed no further larval activity after being dipped in Fastac concentrations of 0.05% to 0.003%. Dead, 1st instar larvae were observed to be present on the fish after treatment with Fastac. Visual assessment, demonstrated that dipping the fish in Fastac prevented damage and losses during processing, at concentrations as low as 0.001%. Losses of edible solids in the untreated fish ranged from 5-11%. Application of Fastac, as a spray, prevented infestation during salting, but did not give continued protection during drying, presumably because of uneven application and lower uptake by the fish.

At the time of these trials, the only, listed, maximum residue levels for fish, were for pyrethrum at 3 mg/kg and piperonyl butoxide at 20 mg/kg (FAO, 1981). The pyrethrum residues, resulting from dipping wet and dry fish, had shown that adequate blowfly infestation control was not possible unless treatments were used, which resulted in residues in excess of the maximum residue levels (FAO, 1981). Although residue levels in the fish treated with Fastac during these trials, were not, at the time, available, the concentrations of the Fastac treatments, were much lower than had previously been reported for any other, effective, insecticide treatment of drying fish.

Table 13: The effect of insecticide treatment on blowfly infestation of *A. thalassinus* during processing- summary of field trial results obtained during February/March, 1984.

Trial	Treatment	No. egg batches after 2 days	χ^2	% fish with grade 2 infestation after 3 days	χ^2	% fish with each level of damage LEVEL 0	L	M	H	%£ loss	χ^2 (damage)
1.	0.05% dip	0		0		100	0	0	0	0	
	Control	5	5^{+}	38	1·3	0	33	33	33	5	
2.	0.025% dip	5		0		100	0	0	0	-	
	Control	45	30^{++}	100	2·0	0	0	80	20	-	$8·1^{++}$
3.	0.0125% dip	1		0		100	0	0	0	0	
	Control	31	26^{++}	54	3·2	20	10	70	0	11	$5·1^{+}$
4.	0.00625% dip	0		0		100	0	0	0	0	
	Control	21	19^{++}	70	$5·1^{+}$	20	10	50	20	10	$5·1^{+}$
5.	0.003% dip	4	0	0/0*	1·5	100	0	0	0	0	0·5
	0.002% dip	4	0	17/0*	1·6	100	0	0	0	0	0·5
	0.001% dip	1	0·8	33/0*	0·3	100	0	0	0	0	0·5
	0.002% spray	12	3·1	67/67*	0	33	0	67	0	13	0·2
	Control	4		67/100*		33	33	33	0	5	

* First figure is after 1.5 days drying, second figure after 2 days drying.

£ Loss of edible solids estimated from difference between weight yields after drying, of undamaged insecticide treated fish and damaged control fish.

$^{+}p<0.05$

$^{++}p<0.01$

0 = No damage

L = Light damage

M = Moderate damage

H = Heavy damage

χ^2(damage) conducted on number of fish with moderate-heavy damage

Significance tests conducted on fish numbers (not % values) throughout thesis.

4.3 Field trials conducted under dry season conditions, during July 1984.

4.3.1 Effects of extended salting on insect infestation and damage of *A. thalassinus* during processing and storage.

Materials and methods

In order to allow the 24 hrs. and 48 hrs. salting treatment fish to dry simultaneously, it was necessary to commence processing the fish, subjected to the 48 hrs. salting treatment, 1 day in advance of the fish subjected to the 24 hrs. treatment. Because there were no cold storage facilities available, the fish used for the 48 hrs salting treatment, were purchased 1 day before the fish that were subjected to the 24 hrs salting treatment. Both batches of fish were purchased from the local landing place. As a result of this procedure, it was not possible to stratify the fish into 2 groups of similar size distribution. The 24 hrs. treatment, consisted of 9 fish with a mean, gutted weight of 1,886g and the 48 hrs. treatment consisted of 9 fish with a mean, gutted weight of 1,456g.

The fish were processed by the standard method, the only variation being the extended salting time of the fish salted for 48 hrs. The fish were examined for blowfly larvae after salting and then placed out to dry. The trays, containing the different treatments, were arranged in a random order. During drying, environmental data, blowfly activity, oviposition, larval infestation and damage to the fish were monitored, using the methods described in Section 4.2.1.

Drying rates were determined by weighing individual fish on an electronic, top-pan balance, at the end of each day.

At the end of drying, 3 fish samples from each treatment, were individually packed in high density, polythene bags and frozen, before being analysed for salt content in the U.K.

Results and discussion

Environmental conditions were favourable for drying and by the end of drying on day 4, the fish given the 24 hrs. salting treatment, showed a weight reduction of 35% and the fish salted for 48 hrs., a reduction of 39%. The difference in drying rates, between the 2 groups of fish (Fig. 26), was probably due to the larger size of the fish, subjected to the 24 hrs. salting treatment.

During salting, the salt contents of the 24 hrs. and 48 hrs. treatments, increased to 14.9 and 15.4% (dwb) respectively. The very small increase in salt content, imparted by the extended salting treatment, was consistent with the results obtained during the previous investigation into extended salting, described in Section 4.2.1.

Blowfly activity was high during the first 2 days of drying (Figs. 27a and 27b) and tended to peak in the early morning and during the afternoon. The slightly elevated salt content, of the fish that had been salted for 48 hours, did not have a repellent effect against settling blowflies.

As would be expected from the salt analysis results, the extended salting treatment had no repellent effect against ovipositing blowfly (Fig. 28) and subsequent grade 2 infestation patterns were similar for both groups of fish (Fig. 29).

Table 14: Effect of salting treatment on larval damage (% of salted fish) during salting and drying.

Salting time (hrs.)	Level of damage			
	Zero	Light	Moderate	Heavy
24	44	33	22	0
48	67	33	0	0

χ^2 on number with moderate-heavy damage = 0.5, $p > 0.05$

Damage to the fish during processing (Table 14), appeared to be lower than that recorded during the rainy season, (Table 10, p 54) when the fish took 1 day longer to dry. The level of

Fig. 26: Drying rates of *A. thalassinus* subjected to normal and extended salting periods. Cirebon, July, 1984

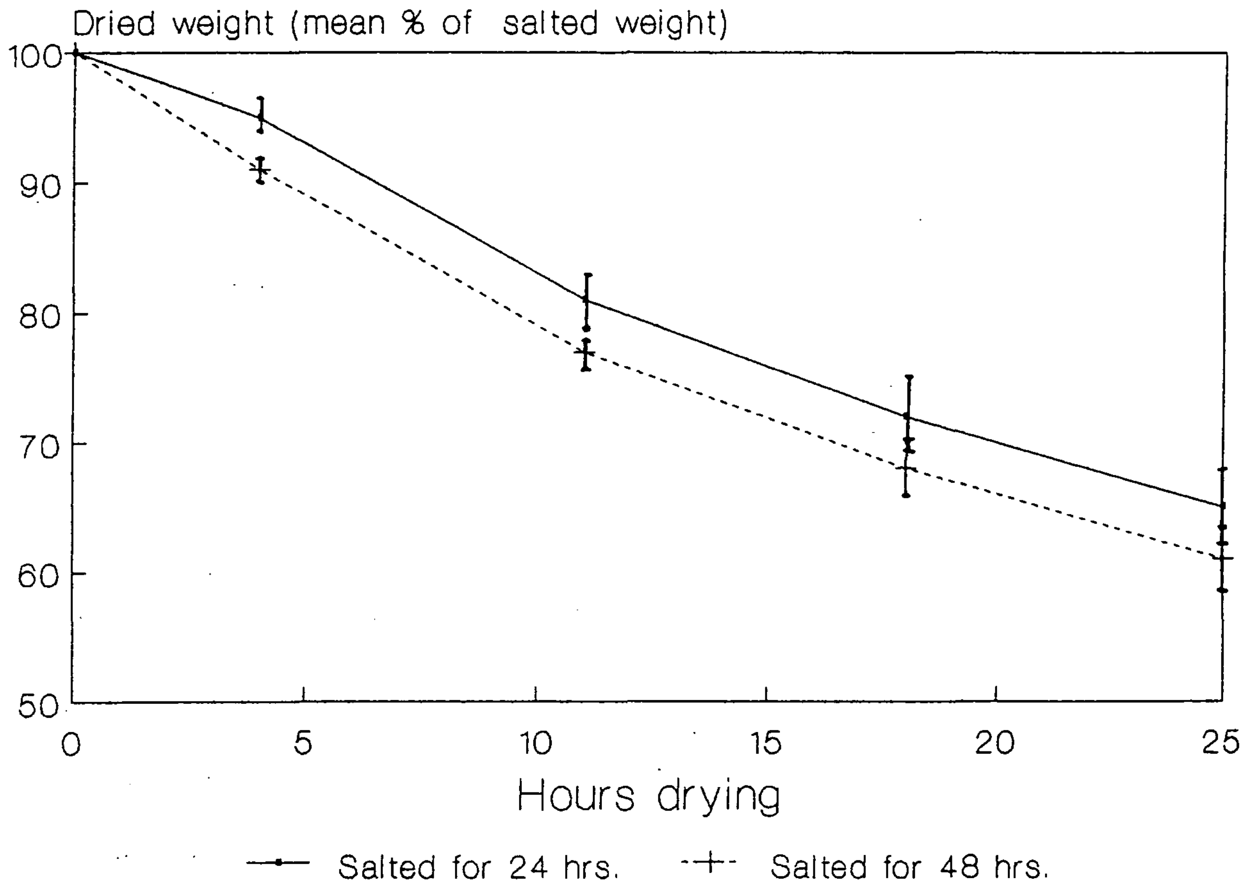
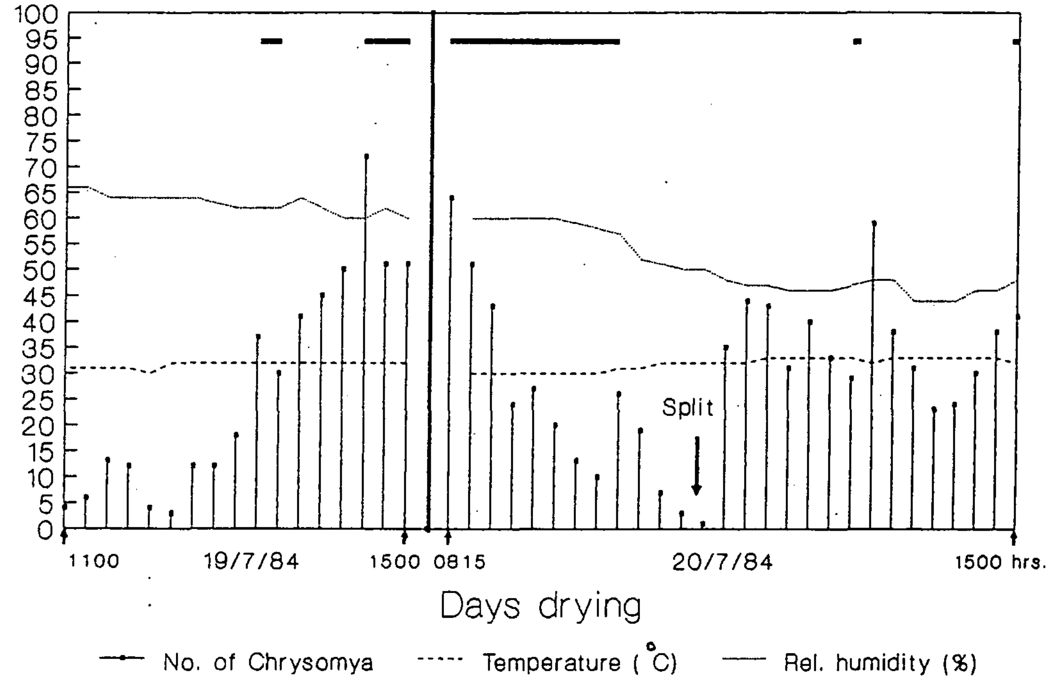


Fig. 27: Effect of extended salting period on blowfly settling activity on A. *thalassinus*. Cirebon, July 1984

(a) Fish salted 24 hrs.

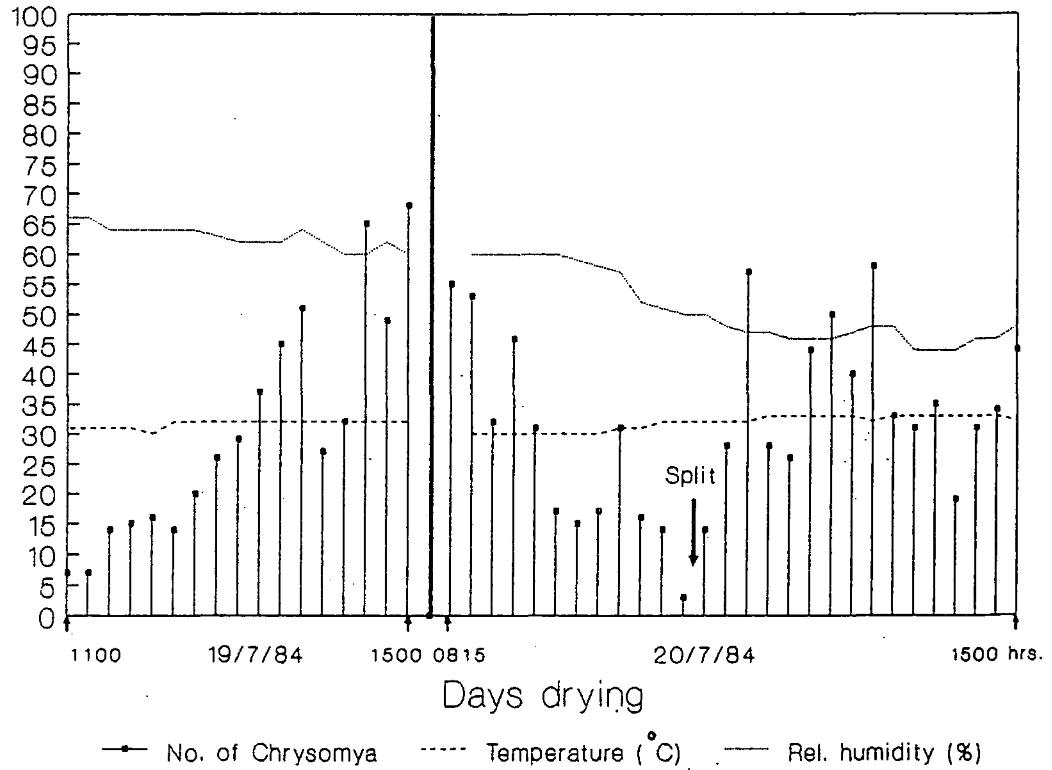
Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Instantaneous counts taken every 15 minutes

▬ Overcast sky

(b) Fish salted 48 hrs.



Mean count

(a) 28·4

(b) 30·9

t = 1·85, p > 0·05

Fig. 28: Effect of salting treatment on blowfly oviposition on *A. thalassinus* during drying. Cirebon, July, 1984

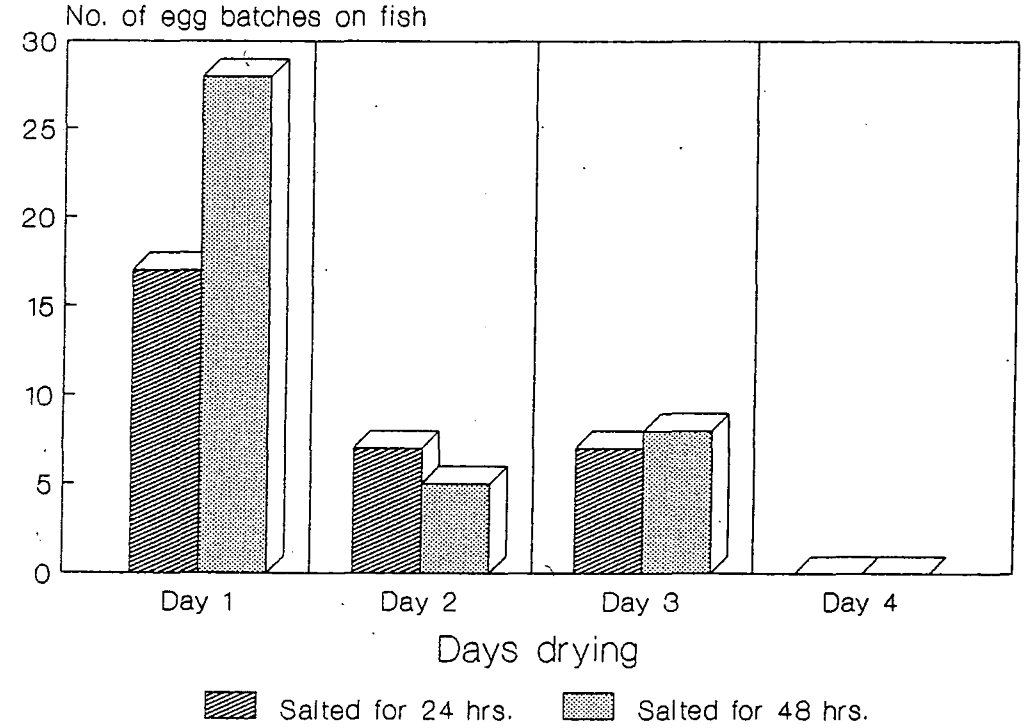
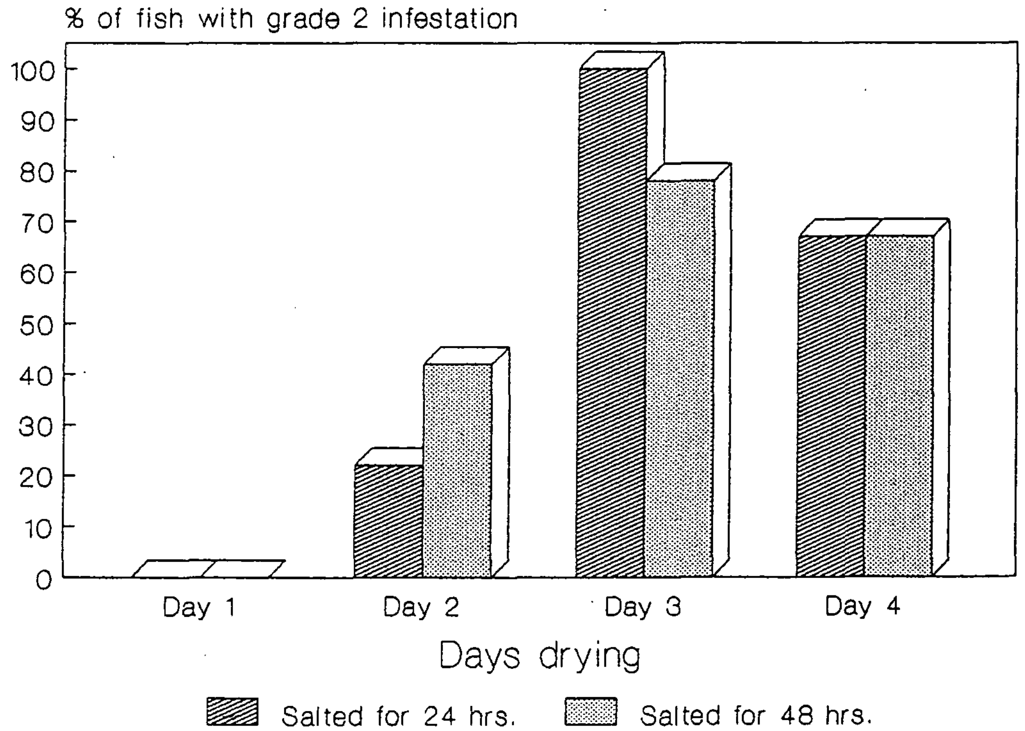


Fig. 29: Effect of salting treatment on larval infestation of *A. thalassinus* during drying. Cirebon, July, 1984



damage, suffered by the fish salted for 24 hrs, was only slightly higher than that of the fish salted for 48 hrs and it can be seen from Fig. 26, that there was only a small difference between the weight yields of the 2 groups of fish, with the 48 hours salted fish showing the higher weight loss.

4.3.2 Effects on insect infestation of *A. thalassinus* during processing and storage, of a combination of screening during salting and treatment with an insecticide dip before drying.

Materials and methods

Two trials, evaluating the effectiveness of a combination of screening during salting and treatment with Fastac before drying, were carried out under dry season conditions in Cirebon.

The fish were purchased at the local landing site and processed by the usual method. During processing, environmental conditions, blowfly activity, oviposition and larval infestation and damage were monitored, using the methods described in Section 4.2.1. In addition, the repellent action of Fastac was measured by timing the periods that blowflies which had landed, remained on the untreated and treated fish.

For Trial 1, 46 fish were divided into 4 groups, each group containing a similar weight distribution. The fish, used for this trial, were salted in the tank protected by a closely fitting lid. In Trial 2, 24 fish were divided into 4 groups of 6. For this trial, the fish were salted in the open tank. The numbers and mean, gutted weights of the fish used in the trials are given in Table 15.

Table 15: Summary of treatments used in Trials 1 and 2.

Trial	Treatment	Mean, gutted weight (g)	t	Number of fish
1. Closed tank.	Control	1230 ± 431		10
	0.001% Fastac	1164 ± 435	0.5	12
	0.006% Fastac	1288 ± 572	0.3	12
	0.05% Fastac	1236 ± 527	0.03	12
2. Open tank.	Control	1515 ± 475		6
	0.001% Fastac	1554 ± 500	0.14	6
	0.006% Fastac	1559 ± 407	0.17	6
	0.05% Fastac	1592 ± 570	0.25	6

Insecticide treatment consisted of dipping each fish into a 6 litre volume of appropriately diluted Fastac for 2 x 3 second periods. The control fish were dipped in water for the same period. The fish were then placed out to dry on trays. When the fish were split, on the 2nd drying day, the newly exposed flesh was sprayed with Fastac of the same concentration as the dip.

On completion of drying, the fish were weighed, assessed for damage, placed in woven, plastic sacks and stored in a room at the processor's premises for 15 weeks.

After storage, the fish were again weighed and examined for the presence of Diptera larvae and the larvae and adults of *Necrobia* and *Dermestes* spp. Fish samples, for insecticide residue analysis, were taken after 1 week's storage, enclosed in polythene bags and stored at -20°C, before being analysed at the Shell Research Centre laboratory.

Results and discussion

During salting, the screened fish remained free of infestation, whereas the fish salted in the open tank, suffered 87% infestation, once more demonstrating the effectiveness of this simple infestation prevention technique.

Fastac had a marked repellent effect against adult blowflies. Figures 30a and 30b show the effect of Fastac concentration on blowfly settling times. In both trials, the blowflies settled on the insecticide treated fish, for shorter periods, than on the control fish. This repellent action was most apparent at the 0.006% and 0.05% concentrations. Those flies, that did settle for longer periods on the insecticide treated fish, tended to engage in mouthparts cleaning, instead of feeding, indicating that Fastac has an irritant effect. The repellent effect, is also demonstrated by the blowfly counts (Figs. 31a to d and 32a to d), which show that fewer blowflies were observed on the fish treated with 0.006% and 0.05% Fastac, than on the control and 0.001% treated fish.

Table 16, shows that dipping in Fastac, was also effective in reducing blowfly oviposition and larval infestation, during processing. Fastac was found to be larvicidal, and larvae, present on the fish that were salted in the open tank, were killed by the Fastac dip, after salting.

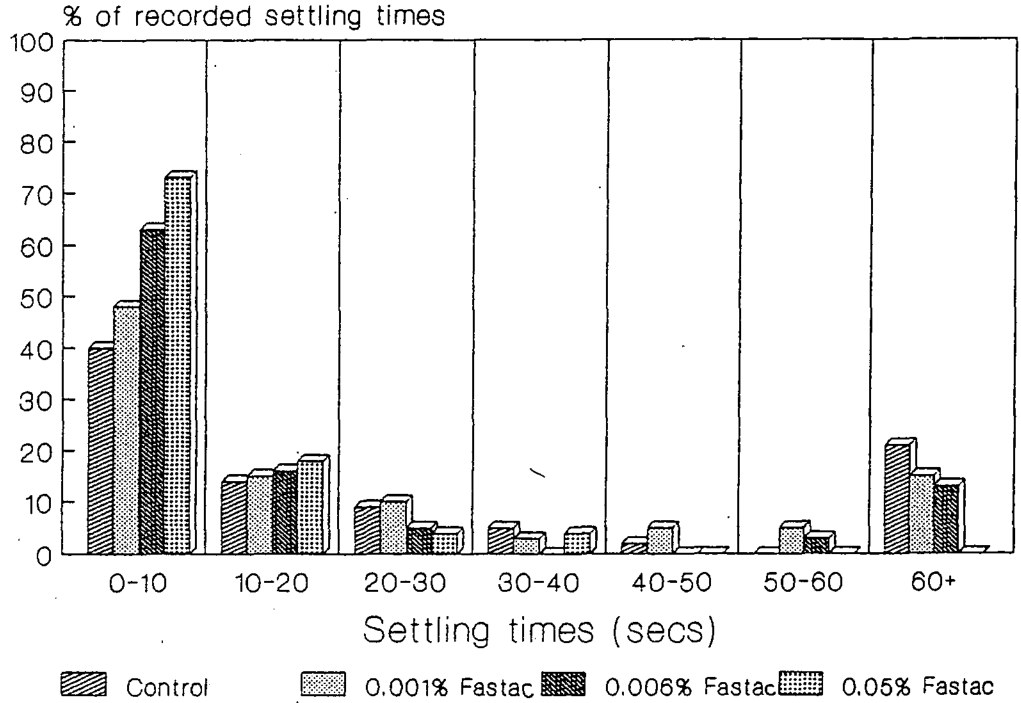
Table 16: The effect of insecticide treatment on blowfly infestation of *A. thalassinus* during processing under dry season conditions - summary of field trial results, Cirebon, July 1984.

Trial	Treatment	No. egg batches after 2 days	χ^2	% fish with grade 2 infestation by 3rd day: am	χ^2	pm	% fish with each level of damage LEVEL: 0	L	M	H
1.	0.05% dip	1	7.7**	0	5.1*	0	100	0	0	0
	0.006% dip	1	7.7**	7	3.1	0	100	0	0	0
	0.001% dip	5	2.7	50	0	0	100	0	0	0
	Control	12		70		0	100	0	0	0
2.	0.05% dip	0	10.1**	0	2.2	0	100	0	0	0
	0.006% dip	2	5.8*	0	2.2	0	100	0	0	0
	0.001% dip	1	7.7**	0	2.2	0	100	0	0	0
	Control	12		17		67	33	33	33	0

*$p<0.05$, **$p<0.01$

Fig. 30: Effect of insecticide treatment on blowfly settling times on drying *A. thalassinus*. Cirebon, July, 1984

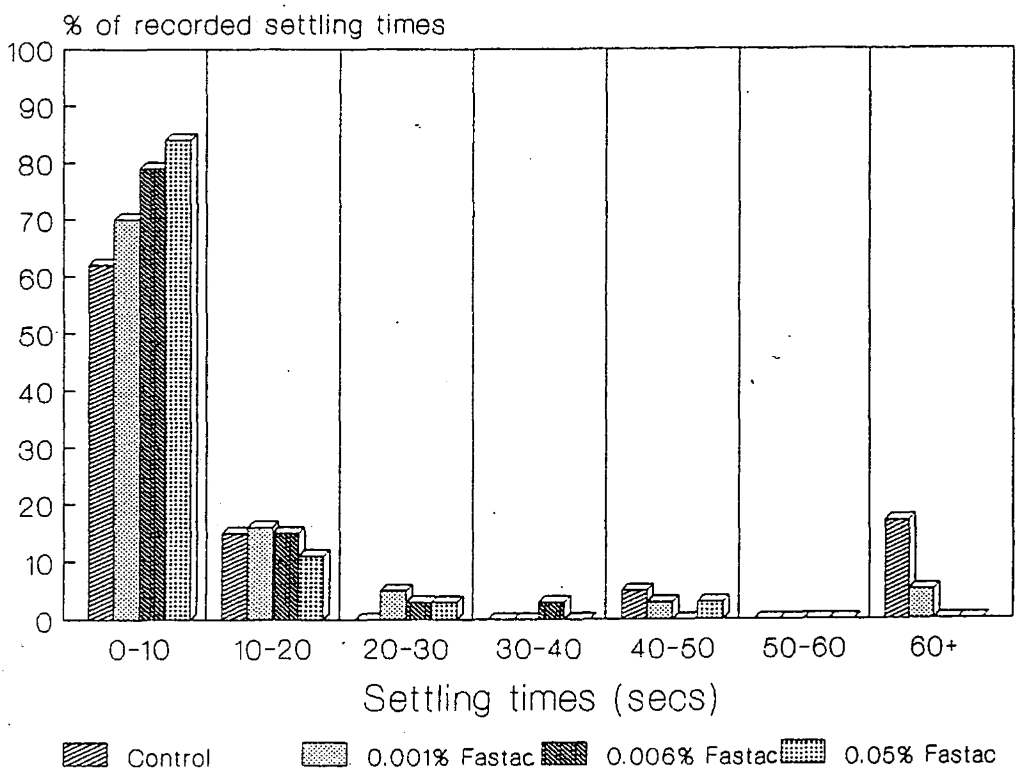
(a) Trial 1



169 counts taken

	Mean settling time (s.)	
Control	21	
0·001% Fastac	23	t = 0·5
0·006% Fastac	16	t = 1·0
0·05% Fastac	9	t: 3·3, $p < 0{\cdot}01$

(b) Trial 2



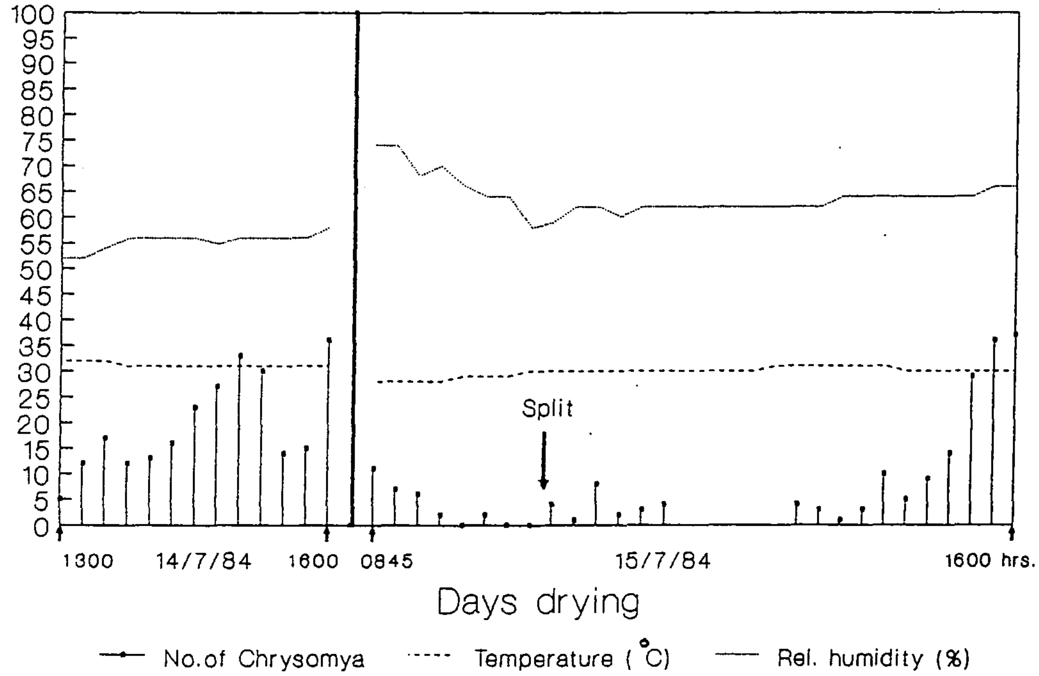
154 counts taken

	Mean settling time (s.)	
Control	18	
0·001% Fastac	11	t = 1·69
0·006% Fastac	7	t = 3·00, $p < 0{\cdot}01$
0·05% Fastac	10	t = 2·06, $p < 0{\cdot}01$

Fig. 31: Effect of insecticide on blowfly settling on *A. thalassinus* during drying. Cirebon, July, 1984

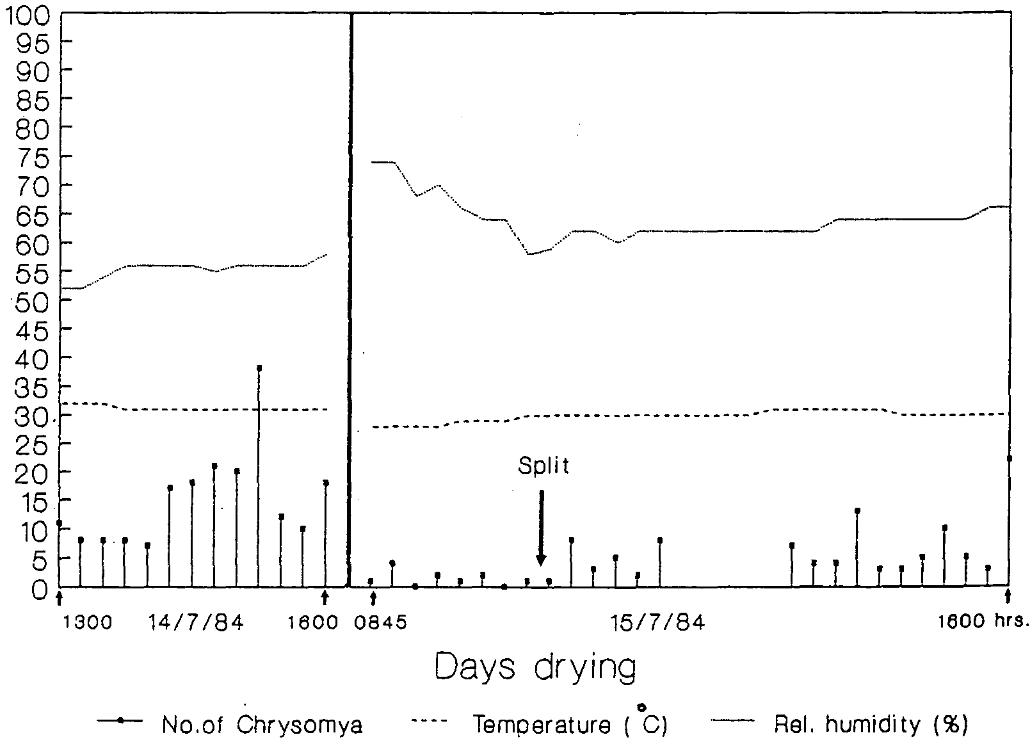
(a) Trial 1. Control fish

Ordinate values are nos. of Chrysomya, air temperature and relative humidity



Instantaneous counts taken at 15 minute intervals

(b) Trial 1. Fish dipped in 0.001% Fastac



<u>Mean count</u>

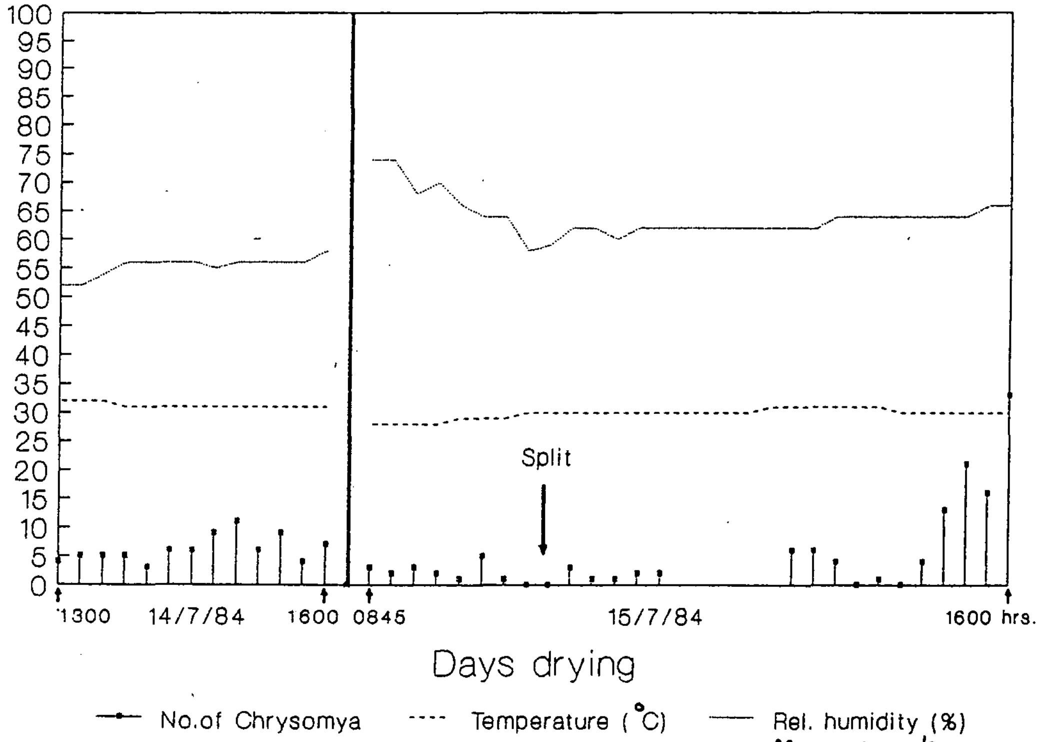
(a) 11·6

(b) 8·0

$t = 2{\cdot}7,\ p < 0{\cdot}05$

Fig. 31 (contd.)

(c) Trial 1. Fish dipped in 0.006% Fastac



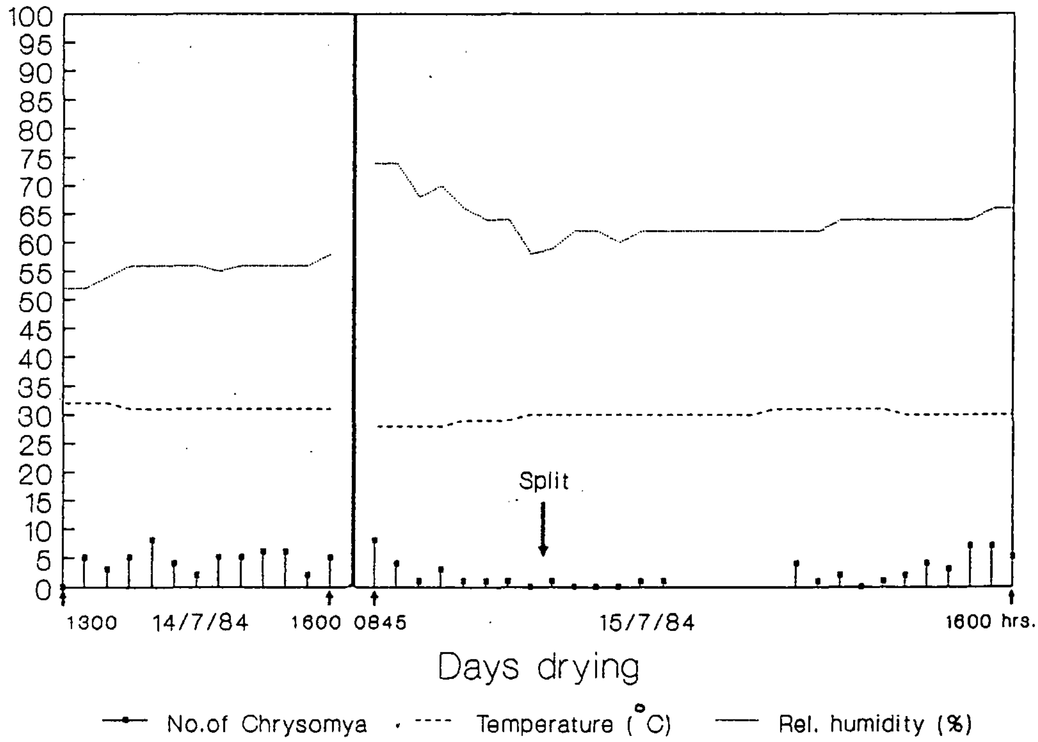
Mean count

(a) 11·6

(c) 5·4

$t = 4{\cdot}8,\ p < 0{\cdot}01$

(d) Trial 1. Fish dipped in 0.05% Fastac



Mean count

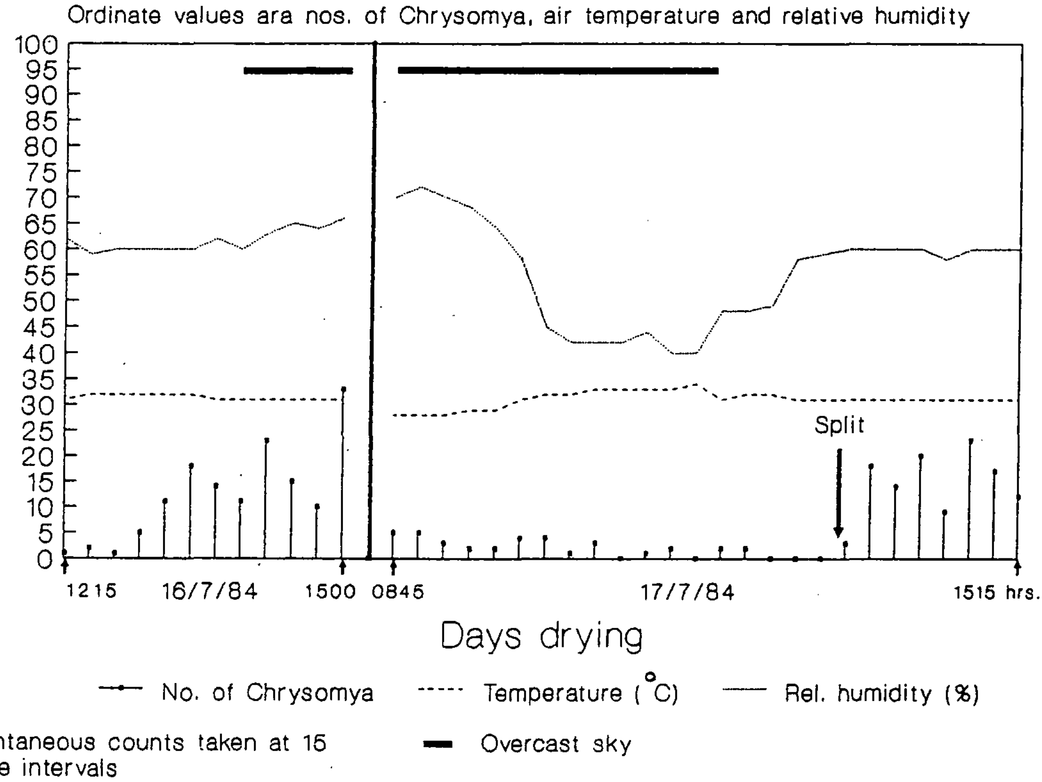
(a) 11·6

(d) 2·9

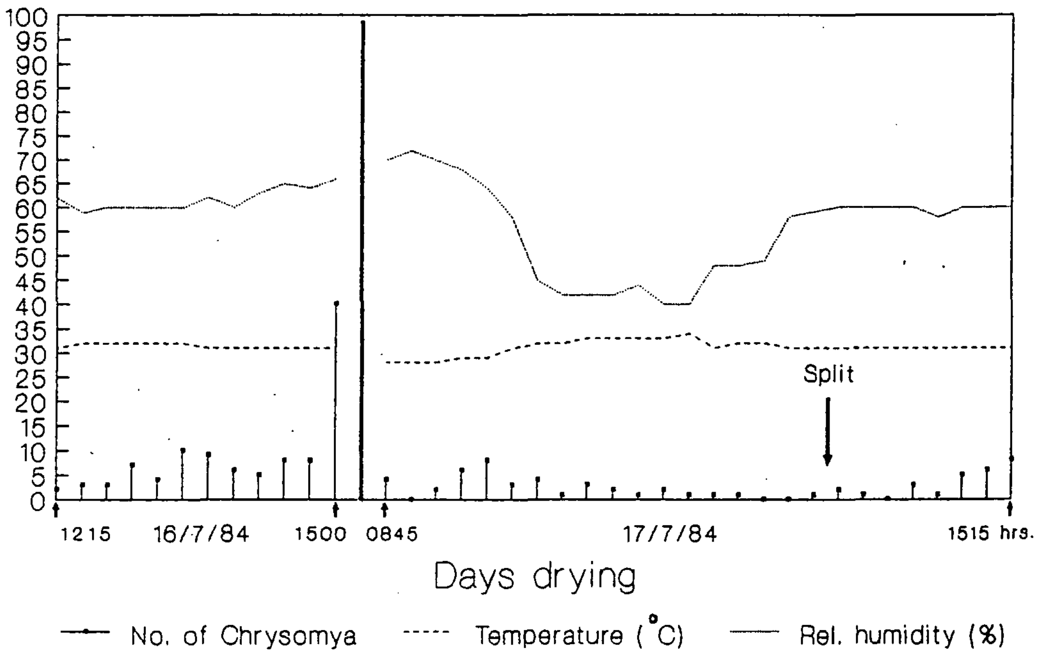
$t = 5{\cdot}4,\ p < 0{\cdot}01$

Fig. 32: Effect of insecticide on blowfly settling on *A. thalassinus* during drying. Cirebon, July, 1984

(a) Trial 2. Control fish



(b) Trial 2. Fish dipped in 0.001% Fastac



<u>Mean count</u>

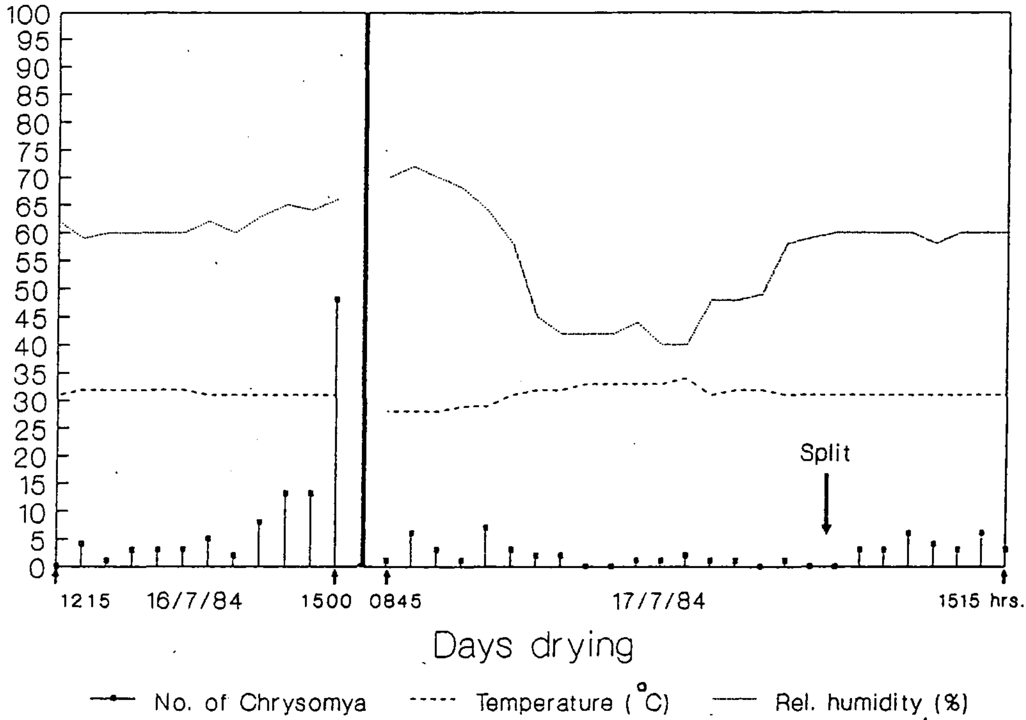
(a) 7·59

(b) 4·38

$t = 3{\cdot}08, p < 0{\cdot}05$

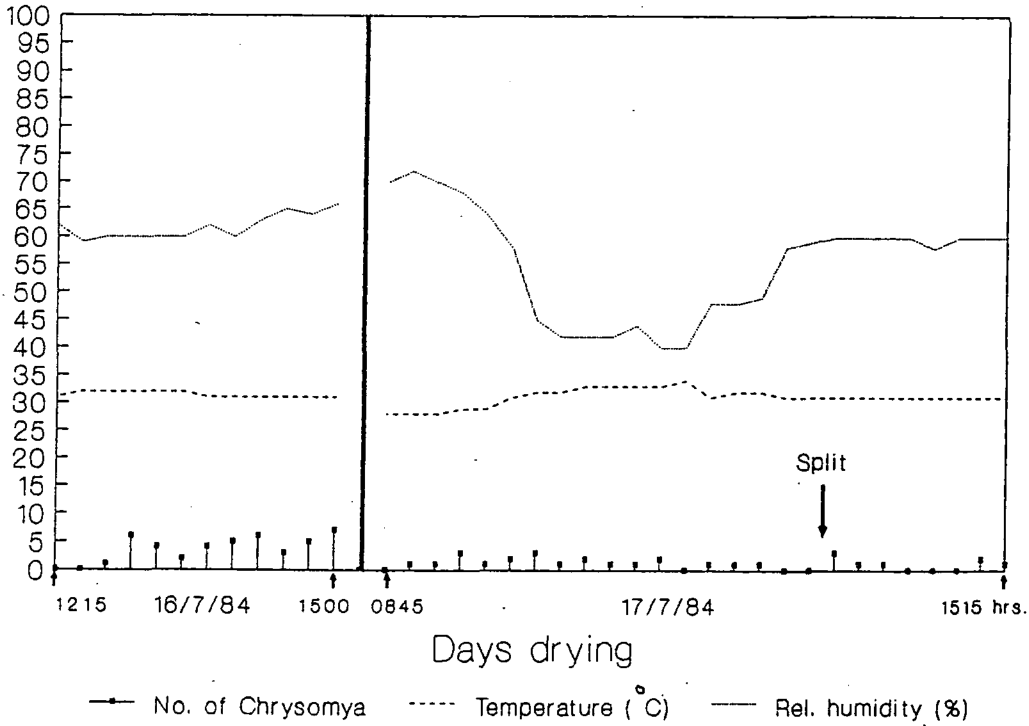
Fig. 32 (contd.)

(c) Trial 2. Fish dipped in 0.006% Fastac



Mean count
(a) 7.59
(c) 4.18
$t = 3.16, p < 0.05$

(d) Trial 2. Fish dipped in 0.05% Fastac



Mean count
(a) 7.59
(d) 1.85
$t = 4.71, p < 0.01$

Damage to the infested fish, was less than that observed at the end of similar trials, carried out during the rainy season. This could be due to either the fish taking longer to dry in the rainy season, or to fewer egg batches being laid on the fish, during the dry season trials.

Observation of insect infestation during storage (Table 17), suggested that treatment with 0.006% and 0.05% Fastac, had a repellent effect against *Dermestes* sp. The presence

Table 17: The effect of insecticide treatment on insect infestation during storage.

Trial	Treatment	*Dermestes* cast skins	χ^2	Dead *N. rufipes*		Insect damage
1.	0.05% Fastac dip	0	2·2	7	5·1*	0
	0.006% Fastac dip	0	2·2	22	20**	0
	0.001% Fastac dip	5	0	0	0	0
	Control	4		0		0
2.	0.05% Fastac dip	0	73**	40	38**	0
	0.006% Fastac dip	0	73**	3	1·3	0
	0.001% Fastac dip	0	73**	1	0	0
	Control	75		0		0

* $p<0\cdot05$ ** $p<0\cdot01$

of numerous, dead *Necrobia rufipes*, on fish treated with 0.006% and 0.05% Fastac, demonstrated the toxicity of Fastac to this organism. The lack of visible, insect damage to any of the stored fish, reflects the relatively low level of insect infestation, which could be due to the Fastac treated fish having an overall, repellent effect, or the fish being protected by the sacks they were stored in. The latter explanation is unlikely, however, as the sacks were unsealed and also contained holes.

Residue analysis results, for the fish used in the wet and dry season trials, are given in Table 18.

Table 18: Fastac residues in salted, dried *A. thalassinus* samples, processed in Cirebon, 1984.

% Fastac in dip solution	Salted weight (g)	Dried weight (g)	Storage time (weeks)	Fastac residue (mg/kg dwb)	Fastac residue (mg/kg wwb)
0	1393	742	1	<0.05	-
	1434	828	2	<0.05	-
	1644	960	2	<0.05	-
	2295	1313	4	<0.05	-
	1059	613	14	<0.05	-
	2006	1165	14	<0.05	-
0.001	1653	878	1	0.46	0.30
	1803	1087	1	0.26	0.14
	1236	712	14	0.26	0.16
	1865	1104	14	0.22	0.13
0.006	452	238	1	2.4	1.4
	1093	573	1	1.5	0.9
	1151	614	1	0.9	0.6
	2236	1361	1	1.3	0.7
	1128	634	14	1.2	0.7
	2031	1184	14	0.8	0.4
0.0125	1073	606	2	2.4	1.4
	1402	757	2	1.6	1.0
0.025	1159	582	2	3.6	2.2
	1366	722	2	2.6	1.2
0.05	394	196	1	30	18
	1840	1099	1	7.5	4
	1578	812	4	2.8	1.4
	2169	1172	4	2.6	1.3
	3048	1667	4	1.6	0.8
	1071	612	14	4.0	2.2
	2020	1234	14	2.6	1.6

Initial uptake of Fastac, was volumetrically estimated for fish with a salted weight of 1500g, to be as follows: 0.4 mg/kg for the 0.001% dip, 2.4 mg/kg for the 0.006% dip, 5 mg/kg for the 0.0125% dip, 10 mg/kg for the 0.025% dip and 20 mg/kg for the 0.05% dip (all estimates on wet weight basis).

Size can be seen to be an important factor in determining residue levels. Smaller fish, in each case, have higher residue levels than larger fish, which have undergone the same treatment. This is to be expected, since insecticide uptake on dipping, depends on surface area, whereas residue levels are quoted in terms of unit weight of the fish. Assuming the initial uptake estimates are reasonably accurate, it can be seen that Fastac levels decreased markedly after drying and 1 week's storage, and then, less rapidly over the subsequent storage period. In general, the residues declined by approximately 50% during the 15 weeks storage period. For the 0.006% Fastac treatment, which gave complete protection against blowfly infestation during drying, Fastac residues were less than 2 mg/kg (wwb) after drying and 1 week's storage. For the 0.05% Fastac treatment, which gave complete protection against insect infestation during processing and storage, Fastac residues were approximately 2 mg/kg after 14 weeks' storage. For comparison, a 0.125% pyrethrin dip synergised with 0.25% piperonyl butoxide gave residues of 12 mg/kg and 70 mg/kg respectively, when used on fish in Malawi (FAO. 1981).

4.4 Conclusions

[illegible]

1. Extended salting of fish, using a pickle cure method, did not significantly increase salt concentration or protect *A. thalassinus* from infestation by *C. megacephala* and *L. cuprina* during processing.

2. Repairing and guarding the tank with a closely fitting lid prevented blowfly infestation during salting and was a technique readily adopted by the processor.

3. Screening during drying, although effective in reducing infestation, proved inconvenient and unacceptable to the processor.

4. The synthetic, pyrethroid insecticide, Fastac, had a marked repellent effect against blowflies, when applied as a dip at concentrations of above 0.001%.

5. Dipping the fish in Fastac, prevented blowfly infestation at concentrations as low as 0.003% and prevented damage at concentrations down to 0.001%.

6. Dipping the fish in Fastac, appeared to have a repellent effect against *Dermestes* sp. at concentrations of 0.05 and 0.006%, over a 15 weeks storage period.

7. The most effective damage prevention method, was a combination of screening during salting and dipping in Fastac before drying.

8. Fastac residues were markedly influenced by the size of the fish, being much higher for small than large fish.

9. For the 0.006% treatment, residues of less than 2 mg/kg (wwb) were found after drying and 1 week's storage. For the 0.05% Fastac treated fish, residues of approximately 2 mg/kg (wwb) were found after 14 weeks storage.

10. Fastac residues declined during drying and subsequent storage.

5. FIELD INFESTATION AND LOSS REDUCTION TRIALS CONDUCTED IN WEST JAVA DURING 1985 AND 1986.

5.1 Introduction

In January 1985, a short visit was made to W. Java, in order to collect further data on the effectiveness of screening and Fastac treatment, in preventing insect infestation of *A. thalassinus* during processing and storage, under wet season conditions. The visit also allowed further data on salt levels in salted, dried *A. thalassinus* to be collected, in support of the series of laboratory investigations, into the effects of salt content on blowfly oviposition and larval infestation, detailed in Section 8.

The earlier field investigations, described in Section 4, had demonstrated that the pyrethroid insecticide, Fastac, was effective at controlling insect infestation of salted, dried fish, when applied at very low concentrations. However, this insecticide needed to be cleared, for use on fish, by the FAO/WHO Joint Meeting on Pesticide Residues (J.M.P.R.), before it could be recommended to fish processors in Indonesia.

Field experiments, carried out in Malawi and the Gambia (Walker and Evans, 1984), have shown that dipping fresh fish in 0.06% and 0.03% aqueous emulsions of pirimiphos-methyl (commercial name Actellic or Silosan), protected them from blowfly infestation. On being advised by the Overseas Development and Natural Resources Institute, that an application for pirimiphos-methyl, to be registered for use on fish, had already been submitted to the J.M.P.R. and that similar clearance for Fastac would take at least 2 years to obtain, it was decided, that subsequent investigations into the effectiveness of insecticide treatment, in controlling insect infestation of cured fish in Indonesia, should concentrate on evaluating pirimiphos-methyl, instead of Fastac.

Pirimiphos-methyl is an organo-phosphorous compound, with the chemical name O-2-diethylamino-6-methylpyrimidin-4-yl OO-dimethyl phosphorothioate. It has a low toxicity (LD_{50} rats 2,050 mg/kg) and is metabolised and excreted, in the urine and faeces of mammals, within 4-5 days. The major metabolites, are the parent hydroxypyrimidine and related N-dealkylated compounds. Neither pirimiphos-methyl, nor its decomposition products, accumulate in the tissues. Pirimiphos-methyl has been in widespread use, principally in public health and the protection of stored food products, since the early 1970s' and no cases of poisoning or injury to health, as a result of its use, have been reported (ICI, 1982).

In order to be in a position, to make positive recommendations on the use of insecticides to reduce losses, before the end of 1986, 2 series of trials, to evaluate the efficacy of pirimiphos-methyl, in controlling insect infestation of salted, dried *A. thalassinus*, under both dry and wet season conditions, were carried out. Subsequent to this decision being taken, the J.M.P.R. cleared pirimiphos-methyl for use on fish and recommended a Maximum Residue Level (M.R.L.) of 10 mg/kg.

In view of the promising results already obtained with Fastac, it was also decided to carry out further, comparative trials using this insecticide, should pirimiphos-methyl fail to control insect infestation, under the processing conditions prevailing in Indonesia.

In addition, a further trial, evaluating the effectiveness of screening *A. thalassinus* throughout both processing and storage, was carried out under rainy season conditions.

5.2 Field trials conducted under rainy season conditions between January and May 1985.

5.2.1 Effects of screening on insect infestation of *A. thalassinus* during processing and storage.

Materials and methods

The fish were initially processed by the standard method described in Section 3 and, after salting in the closed tank, were weighed, stratified and divided into 2 groups of 11 fish. The control fish had a mean, salted weight of 1,743 ± 469g and the experimental, screened fish had a mean, salted weight of 1,810 ± 512g. After salting, the fish were washed and placed on interwoven bamboo mats, before being placed out to dry. Each tray of fish, to be protected by screening, was placed in a fly screen, of the design described in Section 4 and these fish were screened throughout the drying process. The control fish received no protection during drying. On the 2nd drying day, the fish were split and apart from occasional turning, they received no further treatment during drying. On completion of drying, the fish were individually weighed and assessed for blowfly infestation and damage, using the criteria given in Section 4.2.1.

Before being put into store, the screened fish were individually enclosed in stapled, polythene bags and then placed together in an interwoven bamboo basket, the entrance to which was covered with a cement bag. The control fish were not enclosed in polythene bags, before being placed in the basket. The fish were left in a store room, at the processor's premises, for 3 months. After storage, each fish was weighed and examined for insect infestation, damage and microbial growth. A semi-quantitative assessment of insect infestation was made for each batch and the insect debris at the bottom of each basket was collected, for later examination.

Results and discussion

Screening the fish throughout the drying process, reduced drying rates and resulted in the screened fish taking 1 day longer to dry, than the unscreened fish (Table 19). Even after 5 days, the screened fish had only shown a weight loss of about 39%, which was significantly less than the unscreened fish (Student t-test, $p<0.01$). The extended drying period was found to be unacceptable to the processor. During earlier trials, when the fish had only been screened for the first 2 days of drying, it was found that screening did not have a significant effect on drying rates. The fish in this trial, were screened throughout drying, to prevent the possibility of *Dermestes* infestation during drying, from interfering with the storage trials. As the main reason for screening fish during routine processing, would be to protect them from blowfly infestation, it would not be necessary to screen them for longer than the first 2 days of drying, when they are most attractive to blowflies and vulnerable to infestation (see Fig. 6, Section 3).

Table 19: Effect of screening on weight loss of *A. thalassinus* during drying.

Treatment	Mean, salted weight (g)	Mean, dried weight (g)	% weight loss	Drying time (days)
Unscreened	1,743	950	46.0	4
Screened	1,810	1106	39.4	5

The control fish became heavily infested with *C. megacephala* larvae and suffered consequent damage during drying (Table 20). Salt content of about 23% (dwb) failed to provide protection against infestation.

Table 20: Effect of screening on blowfly infestation of *A. thalassinus* and damage during drying.

Treatment	% fish with grade 2 infestation at end of drying	% fish with each level of damage LEVEL 0	L	M	H
Unscreened	45	18	0	27	54
Screened	1	91	9	0	0
	$\chi^2 = 1.5, p > 0.05$	χ^2 on no. with M/H damage $: 7, p < 0.01$			

Although screening was again demonstrated to be an effective method of reducing infestation and damage during drying, further work on improved screen designs, needs to be done, before the method will be acceptable to processors.

The unscreened control fish, were subjected to heavy infestation with *Dermestes maculatus*, which caused severe damage during storage and damage (Table 21).

Table 21: Field observations of effects of screening on insect infestation of *A. thalassinus* and damage during storage.

Treatment	Number of *Dermestes* larvae	χ^2	adults	χ^2	shed larval skins	χ^2	% fish with moderate to high damage	χ^2
Unscreened	>700	700**	21	19**	>700	718**	100	9**
Screened	0		0		18		0	

** $p < 0.01$

Screening with polythene bags, appeared to give good protection against insect infestation, under the prevailing storage conditions, when numerous, unscreened fish were also present and exposed to insect attack. It is possible that, in the absence of unprotected fish, the *Dermestes* larvae would have bored through the polythene bags, to gain access to the fish inside. Although the screened fish had not suffered insect damage during storage, they were relatively damp and of poor quality, due to microbial spoilage. The fish smelled strongly of ammonia and were partially covered with an orange, microbial slime. The unscreened fish had relatively dry surfaces, did not smell of ammonia and had less microbial coverage. The higher microbial activity in the screened fish is not surprising, since the mean weight loss during drying was only 39.4% and during storage 0.7%, compared with a mean weight loss of 46% during drying and 12% during storage, in the unscreened fish. Whereas the unscreened fish, after 3 months storage, contained on average about 46% moisture and almost 13% salt (Table 22), it can be calculated that the screened fish would have contained almost 60% moisture and about 10% salt and, consequently, had a higher water activity than the unscreened fish. This would make the screened fish more susceptible to microbial spoilage.

Table 22: Analytical data for individual fish samples of cured *A. thalassinus* that had been stored for 3 months.*

Weight of each fish (g)	% Moisture mean	sd	% NaCl (wwb) mean	sd	% NaCl (dwb) mean
835	47.7	0.3	13.6	0.8	26.0
1222	49.8	0.6	11.7	0.3	23.3
470	44.8	0.3	12.6	0.3	22.8
499	40.6	1.0	12.0	0.1	20.2
1238	43.7	1.0	12.6	0.4	22.4
865	46.3	0.3	13.4	0.0	24.9
504	46.4	1.2	13.5	0.8	25.2
818	43.3	0.9	10.8	0.3	19.1
1273	47.2	2.6	11.9	0.2	22.5
Overall mean	45.5	2.7	12.5	0.9	22.9

* Values are means of triplicate determinations (3 flesh samples from each fish): SD = standard deviation (n-1), wwb = wet weight basis, dwb = dry weight basis.

The results of the storage trial, indicate that if this type of product is to be successfully stored in polythene bags, it will be necessary to dry the fish to a much lower water content before storage. As this will fundamentally alter the nature of the product, it will probably meet with consumer resistance and not be acceptable to the processor.

5.2.2 Effect of Fastac application on insect infestation and losses of *A. thalassinus* during processing and storage.

Materials and methods

The 44 fish, used in this experiment, were processed by the usual method and, after salting, were divided into 4 groups of similar size distribution. Before drying, the control fish were individually dipped for 2 x 3 second periods in water and the insecticide treated fish were dipped for the same period in an aqueous emulsion of Fastac, at the required concentration. Details of fish weights and insecticide treatments are given in Table 23. The trays of fish were then placed out to dry as normal. Trays containing fish of the same treatment were arranged into blocks and the blocks of different treatments were arranged in random order on the drying racks. Apart from splitting and occasional turning, they received no further treatment. On completion of drying, the fish were individually weighed and assessed for blowfly infestation and damage. Before being put into store, each group of fish was placed into a separate, interwoven bamboo basket and covered with cement bag paper. They were then stored for 3 months at the processor's premises, before being weighed once more and assessed for insect infestation and damage.

After storage, 9 fish, 3 from each insecticide treatment, were sealed in polythene bags, frozen and returned to the U.K. for proximate analysis.

Results and discussion

Weight loss, during the 4 day drying period, ranged from 42.9 to 46.0% (Table 23). The decrease in weight was mainly due to water loss, although consumption of fish by blowfly larvae will have accounted for some of the weight loss, particularly in the control fish. There was, however, no significant difference in weight loss between any of the groups.

Treatment with Fastac, provided protection against blowfly infestation during drying (Table 24).

Table 23: Summary of weight changes to fish during drying and treatments used in Fastac evaluation trials.

Treatment	Mean, gutted weight after salting (g)	Mean weight after 4 days drying (g)	t	% weight loss	Number of fish
Control	1,743 ± 469	950 ± 298		46.0	11
0.001% dip	1,791 ± 435	1,016 ± 276	0.54	43.7	11
0.006% dip	1,775 ± 576	1,003 ± 364	0.37	44.2	11
0.05% dip	1,708 ± 420	967 ± 267	0.14	42.9	11*

* 1 fish taken by cat before end of drying.

Table 24: The effect of Fastac application on blowfly infestation and damage during drying.

Treatment	% fish with grade 2 infestation at end of 4 days drying	χ^2	% fish with each level of damage LEVEL 0	L	M	H	χ^2
Control	45		18	0	27	54	
0.001% dip	19	0.57	82	0	18	0	3.27
0.006% dip	0	3.2	0	0	0	0	7.10**
0.05% dip	0	3.2	0	0	0	0	7.10**

**$p<0.01$

The 0.001% Fastac dipped fish, suffered some infestation and damage during processing, but less than the control fish. The 0.006% and 0.05% Fastac treatments, provided complete protection against infestation during drying. The weight loss figures are consistent with the control fish having suffered higher losses during drying, than the insecticide treated fish.

The control and 0.001% Fastac treated fish suffered *Dermestes* infestation and damage during storage (Tables 25 and 26). There was little evidence of *Dermestes* infestation in the 0.006% Fastac treated fish, although numerous larvae, tentatively identified as Ephydrids, were present. Ephydrid larvae are known, albeit infrequently, to occur in carrion (Erzinclioglu, *pers. comm.*) and have been observed to infest dried fish in India (Soans and Adolph, 1971).

Moderate to high damage was caused to 36% of the 0.006% Fastac treated fish during storage. The absence of Ephydrid larvae from the control fish, might be accounted for by competition with *D. maculatus*. However, large numbers of both *D. maculatus* and Ephydrid larvae were found on the 0.001% Fastac treated fish, which could be explained by a relatively high concentration of Fastac, at the early stages of storage, inhibiting early infestation by *D. maculatus*, thereby allowing a population of Ephydrid larvae to become established. *Dermestes* infestation may have commenced once the Fastac residues had fallen to a sufficiently low level. Unfortunately, it was not possible to test this hypothesis by making sequential observations during storage, due to the author's absence from Indonesia. The 0.006% Fastac treatment was therefore effective in controlling *D. maculatus*, but not Ephydrid infestation, during storage. The 0.05% Fastac treatment provided complete protection against both types of organism (Table 25).

Table 25: Field observations of effects of Fastac application on insect infestation and damage during storage.

Treatment	Number of *Dermestes* larvae	adults	χ^2	shed larval skins	Number of Ephydrid(?) larvae	% fish with moderate to high damage	χ^2
Control	>700	21		>700	0	100	
Fastac 0.001%	>900	18		>1000	>100	100	
Fastac 0.006%	1	0	19**	0	>500	36	2·4
Fastac 0.05%	0	0	19**	0	0	0	9·01**

** $p<0{\cdot}01$

Table 26: Laboratory counts of insects present on fish samples and debris returned to the U.K. (Samples examined within 1 week of return to U.K.)

Treatment	*Dermestes* in debris larvae	χ^2	adults	χ^2	shed larval skins	χ^2	*Dermestes* on fish larvae	adults	Ephydrid larvae on fish
Control	21		15		962		-	-	-
Fastac 0.001%	8	4.96*	3	6.70**	527	126**	126	2	7
Fastac 0.006%	0	19**	0	13**	0	962**	0	0	49
Fastac 0.05%	0	19**	0	13**	0	962**	1	0	0

*$p<0.05$, **$p<0.01$

Relatively high salt levels (Table 22) were seen to offer no protection against *Dermestes* or Ephydrid infestation during the storage trials. This observation has been confirmed in subsequent storage trials conducted by the author (see Section 6). The protective effect of salt is debatable, as field and laboratory trials conducted by other authors have yielded conflicting data. Green (1967), found that heavy salting prevented beetle attack during trials in Saudi Arabia, whereas Aref *et al.* (1965) found that heavily salted fish could suffer appreciable beetle damage during extended storage in Mali. Proctor (1972), found a salt content of 8-10% greatly reduced the damage caused by *D. ater* and *D. maculatus* in Zambia. Mills (unpublished FAO report 1979) reported that light brining, to give salt contents of 2-3%, reduced infestation of fish in Chad, but van der Meeren (1979) was unable to demonstrate any protective effect, during similar trials in the Ivory Coast. Wood *et al.* (1987) conducted a series of laboratory trials on laboratory strains *of D. maculatus* and *D. frischii* that had not previously been exposed to salt and found that a salt content, greater than 9%, largely protected salted, dried whiting from damage, reduced weight losses and greatly inhibited insect development. Variations observed in the field, could be due to insect species differences, preferences for different types of fish, storage conditions or the local availability of unsalted fish. It could also be that local tolerance to high salt concentrations has developed in *Dermestes* strains, found in areas where salting fish before drying is commonly practiced.

Infestation, during this wet season trial, was greater than that observed during the previous dry season. This could be a seasonal effect or be due to the difference in storage methods. In the dry season trial, the fish were stored in sacks, whereas during the wet season trial they were stored in baskets, which may have facilitated easier insect access to the fish.

5.3 Field trials conducted under dry season conditions during October 1985 and rainy season conditions during January 1986.

5.3.1 Effects of Actellic and Fastac application on insect infestation of *A. thalassinus*, during processing and storage.

Materials and methods

Trials 1-4 were conducted on *A. thalassinus* during October 1985, under dry season conditions and Trial 5 was conducted in January, 1986, under rainy season conditions.

Trial 1 investigated the effects of insecticide application on blowfly settling activity and oviposition during processing and insect infestation during storage. Trials 2-4, in addition, investigated the effects of insecticide application on infestation and damage by blowfly larvae. Due to lack of time in Indonesia, it was not possible to record larval damage at the end of Trial 5, although blowfly activity, oviposition and infestation data were obtained. All fish were processed by the standard method and a summary of the numbers of fish, used in the trials, drying periods and the insecticide treatments evaluated, is given in Table 27.

Table 27: Summary of fish numbers and insecticide treatments used in trials.

Trial	Treatment	Number of fish	Drying period (days)
1.	Control, water dip	11	
	0.06% Actellic dip	11	4
	0.006% Fastac dip	11	
2.	Control, water dip	35	
	0.03% Actellic dip	35	4
	0.06% Actellic dip	35	
3.	Control, water dip	15	
	0.015% Actellic dip	21	3
	0.03% Actellic dip	15	
4.	Control, water dip	15	
	0.015% Actellic dip	18	4
	0.03% Actellic dip	18	
	0.003% Fastac dip	15	
5.	Control, water dip	13	
	0.03% Actellic dip	13	4-5*
	0.045% Actellic dip	13	

* Drying period subsequently reported by processor.

After the fish had been salted and washed, those to be treated with Actellic, were individually submerged in 6 litres of emulsion for 15 seconds, before being allowed to drain for 3 seconds and placed on the drying trays. This regime had been developed in laboratory trials at the Overseas Development and Natural Resources Institute. The fish to be treated with Fastac, were dipped into 6 litres of emulsion for 2 periods of 3 seconds, prior to draining for 3 seconds and placing on the drying trays. To be sure that the fish did not deplete the active ingredient in the dip, not more than 15 fish were dipped in any preparation of emulsion. The fish were then placed on trays and the trays, containing different treatments, were arranged in random order on the drying racks. After the fish were further split, on the 2nd or 3rd day of drying, a measured quantity of insecticide used for dipping, of the same concentration, was painted on the newly exposed flesh, using a 2.5 cm brush.

Blowfly activity, mainly *C. megacephala*, during drying was monitored by taking counts at 15 or 30 minute intervals. Blowfly settling times were also recorded. The fish were inspected for blowfly egg batches during the first 2 days of drying and assessed for larval infestation at daily intervals. Infestation and damage were graded using the criteria given in Section 4.2.1. No blowfly damage data is given for Trial 5, because, due to lack of time, the author had to leave Cirebon before drying was completed. In Trial 1, the total numbers of individual eggs, present on the fish, were determined by collecting all of the egg batches and counting the eggs under a stereomicroscope.

At the end of drying, the fish were weighed, assessed for damage and placed in woven bamboo baskets. The baskets were covered with plastic sheeting and stored at the processor´s premises for 10 weeks.

Fish samples, for determination of pirimiphos-methyl residues, were taken at the end of both the drying and storage periods. Each sample was wrapped in aluminium foil and sealed in a polythene bag, before being deep frozen and air freighted to ODNRI, for analysis by gas-liquid chromatography

The fish were examined for insect infestation after 10 weeks storage. Adult insects were collected, for identification, and infestation with *Dermestes* larvae was estimated by recording the number present on each fish. An indication of infestation history, was obtained by counting the number of shed, larval skins. In addition, the fish were weighed and assessed for *Dermestes* damage.

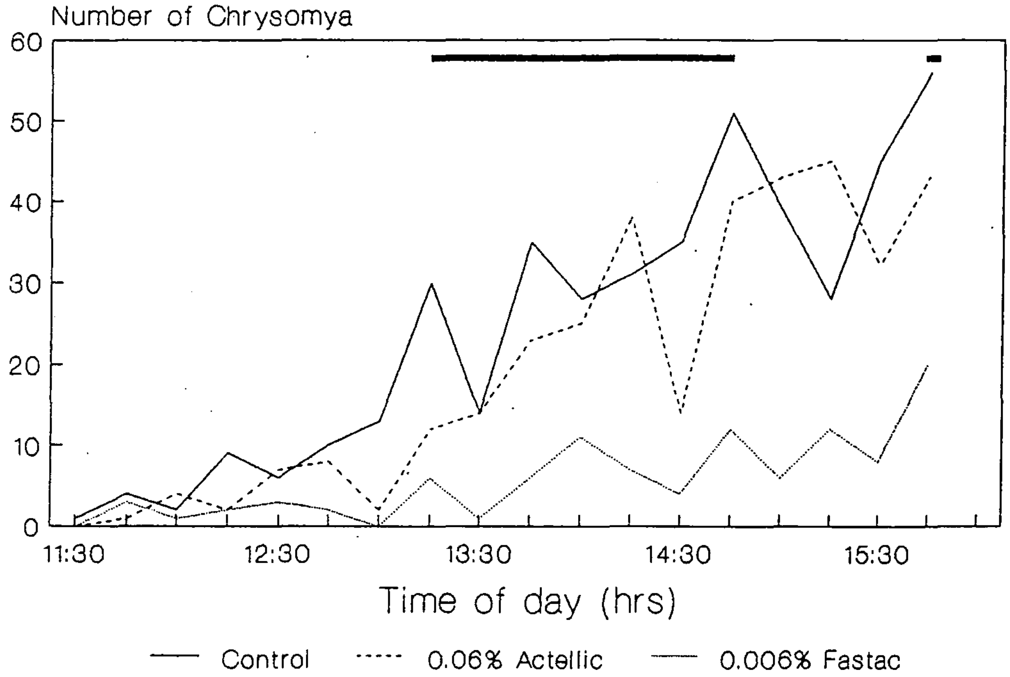
Results and discussion

Figure 33 confirms that Fastac has a repellent effect against blowflies. The application of 0.06% Actellic had no detectable effect on settling blowflies, in any of the trials. The blowfly counts correspond with the observed settling times on the control and insecticide treated fish (Fig. 34). The results indicate that blowflies settled for shorter periods on fish treated with Fastac, than on either the control or Actellic treated fish.

Table 28 suggests that treatment with 0.06% Actellic resulted in an increase in the number of egg batches and total number of eggs laid on the 1st drying day. The egg numbers, per egg batch, appeared to be similar in the control and 0.06% Actellic groups. An increase in egg batches laid on bonga fish, when treated with 0.06% Actellic, had been reported in the Gambia by Walker and Evans (1984). There was a large variation in egg numbers between trays of the same treatment, in both the control and Actellic treated fish. This could be a consequence of group oviposition, which is discussed further in Sections 7 and 8.

As observed in previous trials with *A. thalassinus*, the application of Fastac markedly reduced oviposition.

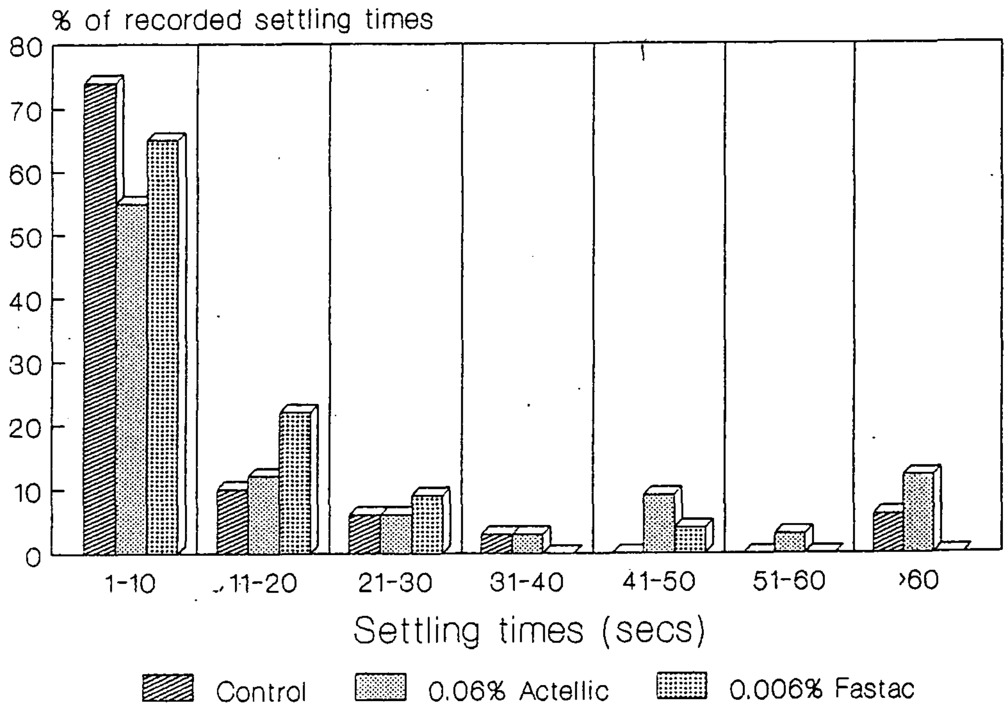
Fig. 33: Effect of insecticides on blowfly settling on *A. thalassinus* during drying. Cirebon, October, 1985



Instantaneous counts taken at 15 minute intervals

Control mean count = 24, Actellic mean count = 20, t = 2.09, $p > 0.05$
Fastac mean count = 6, t = 5.8, $p < 0.01$

Fig. 34: Effect of insecticide treatment on blowfly settling times on drying *A. thalassinus*. Cirebon, October, 1985



87 counts taken

Table 28: Blowfly egg batches and numbers of eggs oviposited on *A. thalassinus* during the first 2 drying days of Trial 1.

Treatment	Tray No.	No. of fish	No. of egg batches Day 1	No. of egg batches Day 2	Total no. of eggs Day 1	Total no. of eggs Day 2
Control	1.	3	10	3	1098	130
	2.	4	6	2	579	2*
	3.	4	8	2	935	132
Mean no. per fish			2.2	0.6	237	124
0.06% Actellic	1.	3	20	0	2741	0
	2.	4	12	4	979	278
	3.	4	12	5	915	168
Mean no. per fish			4	0.8	421 t=4.6+	41 t=1.6
0.006% Fastac	1.	3	2	0	4	0
	2.	4	2	0	15	0
	3.	4	1	0	3	0
Mean no. per fish			0.5	0	2 t=6.1++	0 t=17.9++

+p<0.05, ++p<0.01

* Figure artificially low as 1 egg batch could not be removed from fish.

During subsequent trials (Table 29), the Actellic treatment was demonstrated to have no repellent effect against ovipositing blowflies and, in most cases, more egg batches were laid on the Actellic treated fish, than on the control fish. Treatment with Fastac again reduced the number of egg batches oviposited on the fish in Trial 4 and it was also noted, that the sizes of the egg batches present on the Fastac treated fish were very small, less than 25% of the egg numbers, by eye estimate, relative to the egg batches on the control fish.

Table 29: The effect of insecticide treatment on blowfly oviposition, infestation and damage to *A. thalassinus* during processing- summary of results of 5 trials.

Trial	Treatment	No. egg batches after 2 days	χ^2	% fish with grade 2 infestation after 3 days	χ^2	% fish with each level of damage LEVEL 0	L	M	H	χ^2
1.	Control	31		-		-	-	-	-	
	0.06% Actellic	53		-		-	-	-	-	
	0.006% Fastac	5		-		-	-	-	-	
2.	Control	-		50		23	53	22	2	
	0.03% Actellic	-		0	15**	100	0	0	0	
	0.06% Actellic	-		0	15**	100	0	0	0	13.07**
3.	Control	19		67		27	26	47	0	
	0.015% Actellic	58	18.7**	10		96	4	0	0	
	0.03% Actellic	33	3.2	7		100	0	0	0	
4.	Control	26		67		0	13	67	20	
	0.015% Actellic	40	2.56	5		90	5	5	0	
	0.03% Actellic	26	0	0		95	0	0	5	
	0.003% Fastac	9	7.71**	0		100	0	0	0	
5.	Control	21		85		-	-	-	-	
	0.03% Actellic	16	0.43	0	9.1**	-	-	-	-	
	0.045% Actellic	20	0.02	0	9.1**	-	-	-	-	

** $p<0.01$

There was a marked reduction in infestation of all of the Actellic treated fish (Table 29). Both the 0.03%, 0.045% and the 0.06% Actellic dips proved very effective in controlling infestation. Whilst limited infestation of some Actellic treated fish was recorded, it was noted that the larvae on these fish were small compared to those present on the Control fish. The presence of dead, 1st instar larvae on the treated fish, demonstrated Actellic to be larvicidal in its effect.

Each of the insecticide treatments greatly reduced damage during drying (Table 29). Damage was prevented by Actellic at concentrations of 0.03% and 0.06% and by Fastac at a concentration of 0.003%.

The fish became infested with *D. maculatus*, *D. carnivorus* and *D. ater*, during the 10 week storage period (Table 30). Infestation was similar to that observed between July and October, 1984, but low compared with the storage trial carried out between January and May, 1985. From the limited data available, it appears that stored fish are more susceptible to beetle infestation during the latter part of the rainy season, than at other times of the year. Although infestation pressure was low during this trial, the results obtained, show that insecticide treatment during processing, gave protection against beetle infestation and damage during storage. The 0.03% and 0.06% Actellic dips, both provided complete protection against beetle infestation and damage. Fish treated with 0.015% Actellic were found to be free of larvae and damage, but the presence of shed larval skins, indicated some early infestation during the storage period in Trial 4. The 0.003% Fastac treatment also gave complete protection, although the presence of some shed skins on the 0.006% Fastac treated fish (Trial 1), again indicated some past infestation.

The pirimiphos-methyl residue data, obtained at the end of drying and after 10 weeks storage, are summarised in Table 31.

During the storage period, mean residues for Trials 2-4 dropped by approximately 50%. The remaining, mean residue of 2.1 mg/kg would most likely be sufficient to give further protection against *Dermestes* infestation. It should be noted, however, that there is a wide variation in the final residues, the highest value being almost 5 times as high as the lowest.

Table 30: *Dermestes* infestation and damage during storage. (See details of measures at foot of table)

Trial	Treatment	Larval infestation	Shed, larval skins	Damage	No. of fish
1.	Control	0.08 ± 0.29	1.54 ± 1.69	0.55 ± 0.52	11
	0.06% Actellic	0	0	0	11
		t = 0.9	t = 0.03	t = 3.4**	
	0.006% Fastac	0	0.10 ± 0.32	0	11
		t = 0.9	t = 2.8*	t = 3.4**	
2.	Control	1.52 ± 1.82	6.07 ± 8.34	0.90 ± 0.62	29
	0.03% Actellic	0	0	0	23
		t = 4.5**	t = 3.9**	t = 8.2**	
	0.06% Actellic	0	0	0	23
		t = 4.5**	t = 3.9**	t = 8.2**	
3.	Control	0.09 ± 0.3	0.50 ± 1.73	0.50 ± 1.73	12
	0.015% Actellic	0	0	0	11
		t = 1	t = 1	t = 1	
	0.03% Actellic	0	0	0	12
		t = 1	t = 1	t = 1	
4.	Control	0.87 ± 1.41	3.67 ± 3.18	0.60 ± 0.74	15
	0.015% Actellic	0	1.07 ± 1.79	0	15
		t = 2.4*	t = 2.8**	t = 3.1**	
	0.03% Actellic	0	0	0	15
		t = 2.4*	t = 5.5**	t = 3.1**	
	0.003% Fastac	0	0	0	15
		t = 2.4*	t = 5.5**	t = 3.1**	

Larval infestation: mean number of larvae on each fish ± standard deviation.

Shed, larval skins: mean number of skins on each fish ± standard deviation.

Damage: mean score relating to following criteria:-
Score 0 = no beetle damage, 1 = light beetle damage, 2 = moderate beetle damage ± standard deviation.

*$p < 0.05$

**$p < 0.01$

Table 31: Pirimiphos-methyl residues, at the end of drying and after 10 weeks storage, in fish treated with a 0.03% pirimiphos-methyl dip before drying.+

Trial	Pirimiphos-methyl treatment	Pirimiphos-methyl residues (mg/kg) ± standard deviation at end of drying	Pirimiphos-methyl residues (mg/kg) ± standard deviation after 10 weeks storage	t
2.	0.03%	4.6 ± 2.2	2.4 ± 1.4	2.24*
3.	0.03%	4.6 ± 1.5	1.5 ± 0.5	6.59**
4.	0.03%	4.9 ± 0.9	2.3 ± 0.8	4.12**
Mean of 3 trials		4.7	2.1	

*$p < 0.05$, **$p < 0.01$

Photo-chemical degradation and hydrolysis of the phosphorus ester side chain, to form hydroxypyrimidine compounds, are the principal causes of the decrease in pirimiphos-methyl residues, during processing and storage (ICI, 1982). The hydroxypyrimidines produced, are members of a class of compounds known to be of low toxicity and also occur as products of animal metabolism.

5.4 CONCLUSIONS

Trials carried out between January and May, 1985

1. Treatment of *A. thalassinus* with a Fastac dip at a concentration of either 0.05% or 0.006% provided protection, during drying, against infestation by blowfly larvae (principally *C. megacephala*) and prevented subsequent damage under wet season conditions. Damage by blowfly larvae was reduced at the lowest concentration of Fastac used, 0.001%.

2. There was no significant difference between the percentage weight loss of the control and insecticide treated fish during drying.

+See appendix 3 for details of residues resulting from dips at other concentrations.

3. Salt concentrations of approximately 23% (dwb) failed to provide protection against blowfly larvae during drying.

4. Screening during drying protected the fish against blowfly infestation.

5. The percentage weight loss of the screened fish during drying was markedly lower than for the unscreened fish, even after the screened fish had been dried for an extra day.

6. Treatment of *A. thalassinus* with 0.001% Fastac did not provide protection against infestation and damage by *D. maculatus* during storage.

7. Treatment of *A. thalassinus* with 0.05% and 0.006% Fastac provided protection against infestation and damage by *D. maculatus*.

8. The 0.001% and 0.006% Fastac treated fish became infested with Diptera larvae, tentatively identified as Ephydrids, during storage.

9. The unscreened fish lost approximately 10% in weight during storage. The screened fish lost, on average, less than 1% in weight.

10. A mean salt concentration of approximately 23% (dwb), failed to provide protection against infestation by *D. maculatus* and Ephydrid larvae during storage.

11. Screening during storage provided good protection against insect infestation, when unscreened fish were also present in the store.

12 The screened fish suffered greater microbial spoilage during storage.

<u>Trials carried out between October, 1985 and January, 1986</u>

1.Treatment of *A. thalassinus* with an Actellic (pirimiphos-methyl) dip at concentrations of either 0.06% or 0.03% reduced blowfly infestation and prevented damage in the dry season.

2. Treatment of *A. thalassinus* with an Actellic dip at concentrations of either 0.03% or 0.045%, reduced blowfly infestation in the wet season.

3. Treatment of *A. thalassinus* with an Actellic dip at a concentration of 0.015%, reduced blowfly infestation in the dry season, but some damage was observed.

4. Treatment of *A. thalassinus* with a Fastac (alphacypermethrin) dip, at concentrations of either 0.006% or 0.003% had a repellent effect against blowflies, reduced oviposition, reduced larval infestation and prevented damage to the fish by blowfly larvae.

5. The mean residue after processing for the 0.06% Actellic treatment was 9.8 mg/kg, with 50% of the samples containing more than the FAO/WHO maximum residue limit of 10 mg/kg. The mean residue, after processing, for the 0.03% Actellic treatment, was 4.6 mg/kg, with none of the samples containing more than 10 mg/kg.

6. Over a 10 week storage period, pirimiphos-methyl residues fell by approximately 50%, to a mean level of 2.1 mg/kg, in the fish treated with a 0.03% Actellic dip.

7. Treatment of *A. thalassinus* with an Actellic dip at concentrations of either 0.06%, 0.03% and 0.015%, prevented infestation and damage by *Dermestes* during a storage period of 10 weeks.

8. Treatment of *A. thalassinus* with a Fastac dip at concentrations of either 0.006% or 0.003%, prevented infestation and damage by *Dermestes* during a storage period of 10 weeks.

2. Treatment of *A. thalassinus* with an Actellic dip at concentrations of either 0.03% or 0.045%, reduced blowfly infestation in the wet season.

3. Treatment of *A. thalassinus* with an Actellic dip at a concentration of 0.015%, reduced blowfly infestation in the dry season, but some damage was observed.

4. Treatment of *A. thalassinus* with a Fastac (alphacypermethrin) dip, at concentrations of either 0.006% or 0.003% had a repellent effect against blowflies, reduced oviposition, reduced larval infestation and prevented damage to the fish by blowfly larvae.

5. The mean residue after processing for the 0.06% Actellic treatment was 9.8 mg/kg, with 50% of the samples containing more than the FAO/WHO maximum residue limit of 10 mg/kg. The mean residue, after processing, for the 0.03% Actellic treatment, was 4.6 mg/kg, with none of the samples containing more than 10 mg/kg.

6. Over a 10 week storage period, pirimiphos-methyl residues fell by approximately 50%, to a mean level of 2.1 mg/kg, in the fish treated with a 0.03% Actellic dip.

7. Treatment of *A. thalassinus* with an Actellic dip at concentrations of either 0.06%, 0.03% and 0.015%, prevented infestation and damage by *Dermestes* during a storage period of 10 weeks.

8. Treatment of *A. thalassinus* with a Fastac dip at concentrations of either 0.006% or 0.003%, prevented infestation and damage by *Dermestes* during a storage period of 10 weeks.

6. FIELD INFESTATION AND LOSS REDUCTION TRIALS CONDUCTED IN THAILAND DURING 1986/87 AND 1988.

6.1 Introduction

The findings of the survey of cured fish establishments in Thailand, which are described in Section 2, identified similar problems to those being experienced by cured fish processors in Indonesia. Salted, dried fish was again seen to suffer blowfly attack during processing and both salted, dried and smoked fish suffered continued insect attack, principally by dermestid beetles, during storage. Some Thai fish processors have responded to the problem by applying insecticides, in particular the organophosphorus compound, Dipterex (Dimethyl 2,2,2-trichloro-1-hydroxyethyl-phosphate), to their fish. This product is intended for agricultural and horticultural use and is not approved for use on fish.

As a result of these findings, it was decided to carry out 2 sets of field trials, to evaluate the effectiveness of pirimiphos-methyl and deltamethrin in protecting cured fish, processed in Thailand, against insect infestation during processing and storage.

Deltamethrin is a synthetic pyrethroid compound and has the chemical name [S]-alpha-cyano-m-phenoxybenzyl(1R,3R)-3(2,2 dibromovinyl)-2,2 dimethyl-cyclopropane carboxylate. It has an LD_{50} of 67-139 mg/kg for rats and like alphacypermethrin (Fastac), is effective at very low concentrations. Deltamethrin is currently used for public health purposes and protection of stored food products. In addition, Duguet *et al.*(1985) and Golob *et al.*(1987) have evaluated its use in protecting dried fish during storage trials, conducted in Africa.

The 1st set of trials, carried out during the rainy season of 1986 and extended into early 1987, evaluated the effectiveness of dip treatments with pirimiphos-methyl* and deltamethrin in protecting salted, dried fish against insect infestation during processing and storage. In addition, a pilot experiment, evaluating the effectiveness of spray

*Pirimiphos-methyl is active ingredient of brand products Actellic and Silosan.

treatments of the 2 insecticides, in protecting smoked fish against *Dermestes* infestation, was carried out. The processing method and very dry nature of the smoked product, make spraying a more suitable method of application than dipping, which would result in an unacceptably moist product, that would be susceptible to mould spoilage.

In view of the promising results obtained by the pilot experiment (1986/87), a further set of trials, evaluating the effectiveness of spray treatments of insecticides, in protecting smoked fish against beetle infestation, was carried out during the dry season of 1988.

6.2 Field trials conducted between October 1986 and February 1987.

6.2.1 Effects of dip applications of pirimiphos-methyl and deltamethrin on insect infestation of salted, dried fish during processing and storage.

Materials and methods

The 5 trials* were carried out on the premises of 2 commercial, fish processors, located in Samut Sakhon and are detailed as follows.

Trial 1. (Site 1, Khun Maew, 25 Krokrak, Muang, Samut Sakhon).

100 fresh, iced, marine catfish (*Arius* sp.) were gutted, beheaded and individually weighed and tagged, before being immersed for 18 hours in brine. They were then removed from the brining tank, weighed and soaked in river water for 1 hour. The fish were next divided into 4 batches of equal numbers, each batch containing a similar distribution of fish weights. The control fish were individually dipped in 6 litres of water for 15 seconds, drained, transferred to drying trays and placed out to dry. The 2 batches of pirimiphos-methyl treatment fish, were dipped for 15 seconds in 6 litres of 0.03% and 0.06% a.i. pirimiphos-methyl emulsion respectively, drained and placed out to dry. The deltamethrin treated fish were dipped for 2 periods of 3 seconds in 6 litres of 0.003% a.i. deltamethrin suspension,

*At each site, trials ran simultaneously before being drained and placed out to dry. The trays of fish were arranged in a fully random sequence. The fish were sun dried for 2 days and at the end of drying, were examined for blowfly eggs and larvae Three individual, fish samples from each batch, were randomly taken for salt and moisture analysis and the remaining fish were placed in strong paper bags, put into store, at a dried fish retailer's premises at the Tatien market in Bangkok, for 6 weeks and assessed for insect infestation and damage at the end of the storage period. Damage was graded as follows:

Code	Observation
0	No damage.
L	Occasional holes in flesh.
M	Numerous holes, but flesh not eaten out.
H	Flesh completely eaten out.

Trial 2. (Site 1)

100 fresh, iced hardtail scad (*Megalaspis cordyla*) were individually weighed and and immersed in brine for 2 days, before being removed, re-weighed, washed in fresh water and randomly divided into 4 equal batches. Further treatment was as for Trial 1.

Trial 3. (Site 1)

48 fresh, iced Spanish mackerel (*S. commerson*) were gutted, individually weighed and tagged, before being immersed in brine for 2 days. Further treatment was as for Trial 1.

Trial 4. (Site 2, Khun Wichai Mink, 159 Thashalom, Samut Sakhon).

12 fresh, iced queenfish (*S. commersonianus*) were split, gutted and immersed in saturated brine for 24 hours. Further processing treatment was as for Trial 1. After processing, the fish were enclosed in bamboo baskets and stored at Site 2 for 15 weeks.

Trial 5. (Site 2)

48 fresh, iced marine catfish (*Arius* sp.) were immersed whole in saturated brine for 24 hours. On removal

from the brine, the fish were washed in fresh water, filleted and divided into 4 equal batches of similar weight range. Further treatment was as for Trial 4.

Results and discussion

Weight losses for the fish batches used in Trials 1-5 are summarised in Table 32.

Table 32. Changes in fish weights during processing.

Trial no.	Fish species	Ranges of % mean weight losses between treatments after salting	after drying	Range of t-values+
1.	*Arius* sp.	10.8 - 13.5	41.2 - 44.2	0.27 - 0.58
2.	*M. cordyla*	-	8.2 - 9.5	0 - 1.3
3.	*S. commerson*	12.1 - 12.9	27.4 - 28.0	0.07 - 0.5
4.	*S. commersonianus*	4.0 - 5.5	32.9 - 35.1	0.38 - 0.67
5.	*Arius* sp.	-	47.3 - 48.1	0 - 0.05

Within each trial, mean percentage weight losses due to dehydration were not statistically significant, i.e. as expected, the different treatments had no effect on relative weight losses. The salt and moisture contents of fish processed in Trials 1-3 are given in Table 33.

Table 33. Salt and moisture content of fish at end of drying.

Trial	Fish species	Moisture content (%)	Salt content (% dwb)
1.	*Arius* sp.	55.9	29.3
2.	*M. cordyla*	54.8	20.4
3.	*S. commerson*	61.1	26.7

Results of trials 1 and 2 are means of 3 samples, each sample analysed in duplicate. Results of Trial 3 are means of 4 samples.

+ On fish weights after drying

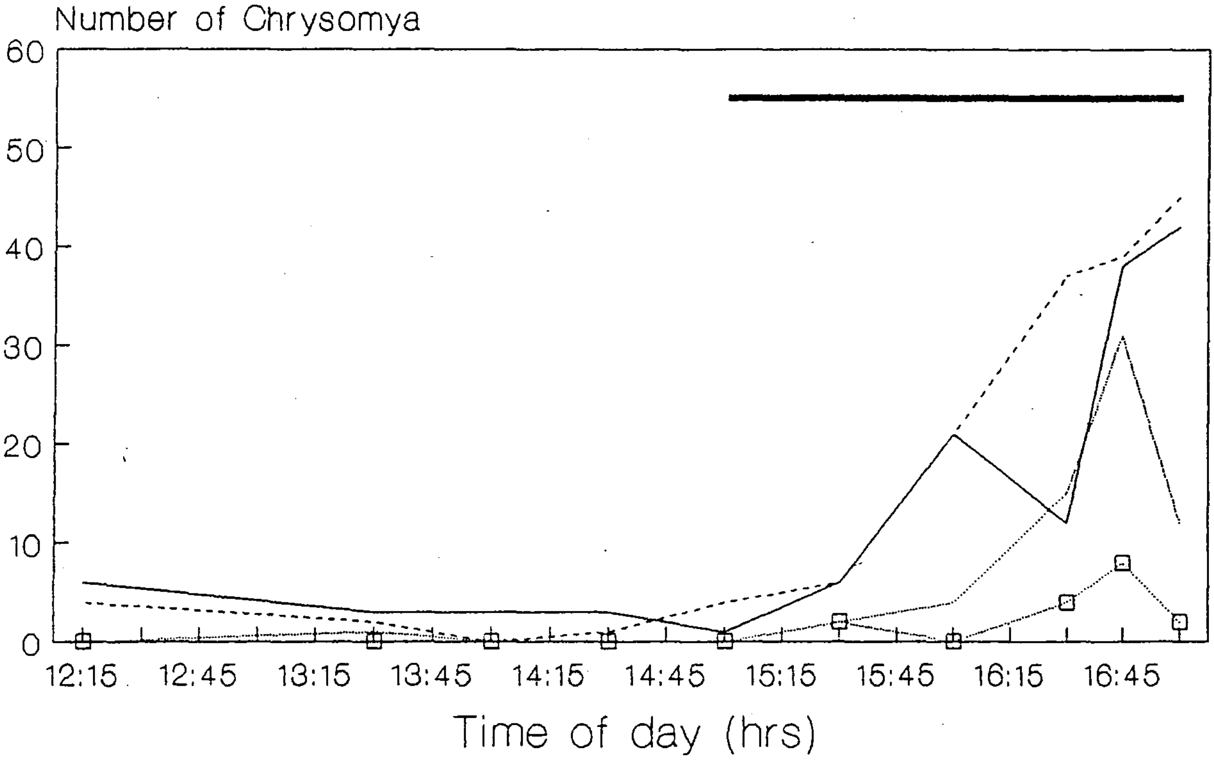
In Trials 1 and 3, percentage moisture content by chemical analysis, corresponded approximately with the percentage weight losses after drying. However, in Trial 2, the percentage moisture of 54.8% did not correlate with the low percentage weight loss of 8.2-9.5%. This might possibly be due to a combination of the relatively high lipid and bone contents in this species.

C. megacephala was the only blowfly species observed during the trials and with the exception of 1 day (October 14th, Figure 35), was present in very low numbers. The pattern of blowfly activity, illustrated in Figure 35, was similar to that observed during earlier trials in Indonesia, where the numbers of blowflies, settling on the fish, increased during the late afternoon. It can also be seen, that of the insecticide treatments, only the 0.003% deltamethrin had an observable, repellent effect against blowflies. These observations support earlier findings in Indonesia, where pirimiphos-methyl and a similar, synthetic pyrethroid insecticide, alphacypermethrin, were evaluated (Esser *et al.* 1986).

Blowfly activity, during the course of the Thailand trials, seemed to be influenced by weather. For most of the time, the weather during the period of the trials, was hot (shade temperatures 33-38°C) and sunny and blowfly activity was low. A build up of cloud cover from 1500 hours on October 14th, was accompanied by a marked increase in blowfly activity. The processor remarked that the blowfly problem was always worse on dull, still, humid days and that, during protracted periods of such weather, his fish suffered heavier infestation, unless protected by insecticide.

Oviposition and infestation with blowfly larvae were restricted to the *S. commerson*, processed in Trial 3 (Table 34). The eggs were either deposited in the abdominal cavity or beneath the opercula and resulted in 92% of the control fish becoming infested.

Fig. 35: Effect of insecticides on blowfly settling on catfish (*Arius* sp.) during drying. Samut Sakhon, Oct., 1986



— Control ---- 0.03% Actellic ······ 0.06% Actellic -□- 0.003% Cislin (deltamethrin)

antaneous counts of *C. megacephala* ▬ Overcast sky
fish.

Control mean count = 13, 0·03% Actellic mean count = 16, t = 1, $p > 0{\cdot}05$

0·06% Actellic mean count = 6, t = 2·2, $p > 0{\cdot}05$

0·003% Cislin mean count = 2, t = 2·7, $p < 0{\cdot}05$

Table 34. Blowfly infestation of *S. commerson*, processed in Trial 3.

Treatment	No. of egg batches	% larval infestation 0	L	M	H
Control	3	8	42	17	33
0.03% pirimiphos-methyl	1	100	0	0	0
0.06% pirimiphos-methyl	0	100	0	0	0
0.003% deltamethrin	0	100	0	0	0

χ^2 on number with moderate-high damage = 4.2, $p < 0.01$

As observed in Indonesia, the relatively high salt content of 26.7% (dwb), failed to protect the fish against infestation. All of the insecticide treatments gave complete protection against larval infestation during this trial.

During storage, the fish were subjected to *Dermestes* beetle attack, particularly at the Samut Sakhon site. Of the 27 beetle samples collected from the stored fish, 22 were identified as *D. maculatus* and 5 as *D. ater*. Occasional *Necrobia rufipes* were also observed on the fish.

Infestation of the salted, dried fish, left in store for 6 weeks at the Tatien market, in Bangkok, was lower than that observed at Site 2 in Samut Sakhon. The marine catfish, processed in Trial 1, showed no evidence of insect infestation and the mean weight loss of 46.5% (Table 35) was solely due to dehydration during storage. Moisture content of the Trial 1 fish dropped from a mean of 55.9% to 17.5% during storage and mean salt content was 29% (dwb).

Dehydration, resulting from the generally dry microclimate of the storage rooms, is certainly the most important cause of weight loss during the storage periods used for these trials. Weight loss, due to insect damage, was relatively insignificant and did not act as a useful indicator of *Dermestes* attack.

There was some evidence of *Dermestes* infestation of the *M. cordyla*, processed in Trial 2, but visible damage was very slight (Table 36). The fish showed a mean weight loss during storage, of 40.25% and mean moisture content

Treatment	Weight before storage (g)			Weight after storage (g)			% weight change	No. Dermestes larvae	No. Dermestes adults		% fish with numerous casts	% damaged fish			
	$\bar{x}$	σ_{n-1}	Σx	$\bar{x}$	σ_{n-1}	Σx			Live	Dead		O	L	M	H
CONTROL	393	159	8,647	201	74	4,426	-49	0	0	0	0	100	0	0	
0.03% PIRIMIPHOS METHYL	367	157	9,185	211	85	5,268 t = 0.4	-42	0	0	0	0	100	0	0	
0.06% PIRIMIPHOS METHYL	80	178	9,500	193	80	4,827 t = 0.4	-49	0	1	0	0	100	0	0	
0.003% DELTAMETHRIN	377	156	9,414	202	81	5,054 t = 0.05	-46	0	0	0	0	100	0	0	

Storage location:- Tatien Market, Bangkok

Table 35 Salted, dried Arius (Trial 1) data collected at the end of storage

Treatment	Weight before storage (g)			Weight after storage (g)			% weight change	No. Dermestes larvae	No. Dermestes adults		% fish with numerous casts	% damaged fish			
	$\bar{x}$	σ_{n-1}	Σx	$\bar{x}$	σ_{n-1}	Σx			Live	Dead		O	L	M	H
CONTROL	142	19	3,549	85	14	2,136	-40	0	0	0	0	68	23	9	0
0.03% PIRIMIPHOS METHYL	149	20	3,721	87	12	2,164 t=0.5	-42	0	0	0	0	96	4	0	0
0.06% PERIMIPHOS METHYL	142	23	3,561	86	16	2,160 t=0.2	-39	0	0	0	0	100	0	0	0
0.003% DELTAMETHRIN	140	18	3,491	84	12	2,112 t=0.5	-40	0	0	0	0	100	0	0	0

Storage location:- Tatien Market, Bangkok

Table 36 Salted, dried *M. cordyla* (Trial 2) data collected at the end of storage.

Piophila pupal cases and occasional Dermestes larval cases present on control fish.

decreased from 54.8% to 34.7%. Mean salt content was 20% (dwb). The 0.03% pirimiphos-methyl treatment reduced *Dermestes* infestation and the 0.06% pirimiphos-methyl and 0.003% deltamethrin treatments, both gave complete protection against infestation during storage.

The *S. commerson*, processed in Trial 3, remained completely free of infestation during storage (Table 37) and the mean weight loss of 52.5% was solely due to dehydration. Mean moisture content decreased from 61.1% to 14.5% and mean salt content was 27% (dwb).

The untreated *S. commersonianus*, processed in Trial 4 and left in store at Site 2, showed evidence of heavy *Dermestes* infestation during the earlier stages of storage, although damage caused was light (Table 38). Mean weight loss during storage was 39.5% and mean moisture content decreased to 15.7%. All insecticide treatments effectively controlled *Dermestes* infestation and damage during storage.

Of the salted, dried fish left in store, the marine catfish, processed in Trial 5, suffered the most significant *Dermestes* damage. The untreated fish suffered a decrease in yield of approximately 5%, compared with the insecticide treated fish, with 75% of the fish showing moderate to high beetle damage (Table 39). The 0.03 and 0.06% pirimiphos-methyl treatments reduced *Dermestes* infestation and damage and the 0.003% deltamethrin treatment gave complete protection. The mean moisture content of the fish after 15 weeks storage was 27.7% and the mean salt content was 14% (dwb).

The results support the findings of Golob *et al.*(1987), during dried fish storage trials conducted in Kenya. Of the 6 insecticides evaluated, only deltamethrin and pirimiphos-methyl were deemed worthy of further investigation as protectants of dried fish against beetle infestation, if protection, for periods exceeding 6 months, was required. The deltamethrin, applied at concentrations of 0.001 and 0.002% was found to provide the most effective control and resulted in residues below the

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Treatment	Weight before storage (g)			Weight after storage (g)			% weight change	No. Dermestes larvae	No. Dermestes adults		% fish with numerous casts	% damaged fish			
	$\bar{x}$	σ_{n-1}	Σx	$\bar{x}$	σ_{n-1}	Σx			Live	Dead		O	L	M	H
CONTROL	391	117	4,698	190	63	2,277	-52	0	0	0	0	100	0	0	0
0.03% PIRIMPHOS METHYL	388	97	4,662	184	47	2,068 $t = 0.3$	-56	0	0	0	0	100	0	0	0
0.06% PIRIMPHOS METHYL	379	70	4,547	197	31	2,360 $t = 0.3$	-48	0	0	0	0	100	0	0	0
0.003% DELTA-METHRIN	371	57	4,453	173	25	2,073 $t = 0.9$	-53	0	0	0	0	100	0	0	0

Storage location : Tatien Market, Bangkok

Table 37 Salted, dried S. commerson (Trial 3) data collected and the end of storage.

Treatment	Weight before storage (g)			Weight after storage (g)			% weight change	No. Dermestes larvae	No. Dermestes adults		% fish with numerous casts	% damaged fish			
	$\bar{x}$	σ_{n-1}	Σx	$\bar{x}$	σ_{n-1}	Σx			Live	Dead		O	L	M	H
CONTROL	2,063	665	6,188	1,170	344	3,510	-43	0	0	0	100	0	100	0	0
0.03% PIRIMIPHOS METHYL	1,787	245	5,362	1,064	154	3,192 $t \approx 0.5$	-40	0	10	0	0	67	33	0	0
0.06% PIRIMIPHOS METHYL	1,847	266	5,540	1,191	119	3,573 $t = 0.1$	-36	0	0	1	0	100	0	0	0
0.003% DELTA-METHRIN	1,584	671	4,752	955	386	2,864 $t = 0.7$	-40	0	0	0	0	100	0	0	0

Storage location :- premises of Mr. Mink, Samut Sakhon

Table 38 Salted, dried S. commersonianus (Trial 4) data collected at end of storage.

Treatment	Weight before storage (g)			Weight after storage (g)			% weight change	No. Dermestes larvae	No. Dermestes adults		% fish with numerous casts	% damaged fish			
	$\bar{x}$	σ_{n-1}	Σx	$\bar{x}$	σ_{n-1}	Σx			Live	Dead		O	L	M	H
CONTROL	186	56	2,235	105	23	1,261	-44	8	0	0	33	17	8	33	42
0.03% PIRIMIPHOS METHYL	185	48	2,225	117	25	1,400 $t=1.2$	-37	0	1	2	8	50	8	42	0
0.06% PIRIMIPHOS METHYL	186	43	2,238	116	23	1,394 $t=1.2$	-38	0	1	10	0	75	25	0	0
0.003% DELTAMETHRIN	185	51	2,224	112	23	1,348 $t=0.7$	-39	0	0	0	0	100	0	0	0

Storage locations:- premises of Mr. Mink, Samut Sakhon

χ^2 on no. fish with M-H damage = 0.64 for 0.03% pirimiphos-methyl ($p > 0.05$), and 7 for 0.06% pirimiphos-methyl and 0.003% deltamethrin ($p < 0.01$).

Table 39 Salted, dried *Arius* (Trial 5) data collected at the end of storage limit of 2 mg/kg set for cereals by the Joint Meeting on Pesticide Residues. Pirimiphos-methyl, when applied at concentrations of 0.01 and 0.02%, gave adequate protection against beetles and residues were equal to or less than the MRL of 10 mg/kg set for fish. Duguet *et al.*(1985) also found deltamethrin, applied as a 0.0025% dip and pirimiphos-methyl, applied at 0.05%, to be effective in controlling *Dermestes* infestation of dried fish in Mali.

6.2.2 Pilot investigation into the effects of spray applications of pirimiphos-methyl and deltamethrin on insect infestation of smoked marine catfish (*Arius* sp.) during storage.

Materials and methods

200 marine catfish were hot smoked for 12 hours, weighed and randomly divided into 4 batches of 50 fish, 3 for insecticide treatment and 1 control batch. The insecticide treatments were 0.03 and 0.06% pirimiphos-methyl and 0.003% deltamethrin. 500 ml of each insecticide solution was prepared and transferred to a hand operated spray gun. Each batch of fish was treated with 80 applications of spray (40 applications to each side of the fish). The control fish were sprayed with water. The weight of solution, used per batch, was measured by weighing the spray gun before and after use. Actual amounts of insecticide formulation, applied to the fish, ranged from 21-27g. After spraying, the smoked fish were sun dried for 3 hours, before being wrapped in paper and stored in bamboo baskets and placed into store at the processor's premises. Other than the insecticide application and sun drying, the treatment was that normally given by the processor, when processing this product.

After 15 weeks, the fish were individually weighed and examined for the presence of *Dermestes* adults, larvae and shed larval skins. Samples of adult insects were collected for identification. Damage was graded as follows:

Code	Observation
0	No damage.
L	Occasional holes in flesh.
M	Numerous holes, but flesh not eaten out.
H	Flesh completely eaten out.

Results and discussion

Of the fish left in store, the smoked marine catfish suffered the heaviest *Dermestes* infestation and damage. All of the untreated, control fish suffered moderate to high insect damage, which resulted in a decrease in yield of approximately 25% (Table 40). The low moisture content of the fish was probably a factor in making them so vulnerable to beetle infestation. The results of the 0.03 and 0.06% pirimiphos-methyl treatments were ambiguous, in that the 0.03% treatment appeared to give better protection than the 0.06% treatment, which resulted in infestation levels that were similar to the control group. Of the 2 insecticides tested, the deltamethrin treatment gave the best protection and appeared to have a repellent effect against *Dermestes*. This effect has also been observed by Taylor and Evans (1982), Duguet *et al.*(1985) and Golob *et al.*(1987). The unusual results of the pirimiphos-methyl treatments could be a consequence of the baskets of fish being moved by the processor, to facilitate building work during the storage period. It is possible that the basket containing the 0.03% pirimiphos-methyl treated fish was placed in close proximity to the basket containing the deltamethrin treatment and that the repellent effect of the deltamethrin might have protected the pirimiphos-methyl treated fish against infestation.

Treatment	Weight before storage (g) $\bar{x}$	σ_{n-1}	Σx	Weight after storage (g) $\bar{x}$	σ_{n-1}	Σx	% weight change	No. Dermestes larvae	No. Dermestes adults Live	Dead	% fish with numerous casts	% Damaged fish O	L	M	H
CONTROL	74.6	34.3	3,728	57.3	25.8	2,807	-25	49	92	2	86	0	0	6	94
0.03% PIRIMIPHOS METHYL	69.3	30.0	3,464	69.4	29.4	3,470 $t = 1.5$	0	4 $\chi^2 = 36^{**}$	34 $\chi^2 = 26.7^{**}$	0	26 $\chi^2 = 16^{**}$	52	18	24	6
0.06% PIRIMPHOS METHYL	76.6	29.0	3,831	57.2	23.5	2,862 $t = 0$	-25	30 $\chi^2 = 4.6^{*}$	57 $\chi^2 = 7.8^{**}$	0	64 $\chi^2 = 1.6$	0	2	38	60
0.003% DELTAMETHRIN	74.3	29.0	3,713	75.1	29.8	3,756 $t = 2.25^{**}$	+1	3 $\chi^2 = 40^{**}$	13 $\chi^2 = 5.8^{**}$	0	6 $\chi^2 = 35^{**}$	82	10	8	0

Storage location:- premises of Mr. Mink, Samut Sakhon.

$^{*}p \leq 0.05$, $^{**}p < 0.01$

χ^2 on fish with M-H damage = 18, ($p < 0.01$) for 0.03% pirimiphos-methyl, 0 ($p > 0.05$) for 0.06% pirimiphos-methyl and 38 ($p < 0.01$) for the 0.003% deltamethrin treatment.

Table 40 Smoked Arius data collected at end of storage

6.3 Field trials conducted between March and May 1988.

6.3.1 Effects of spray applications of pirimiphos-methyl and deltamethrin on insect infestation of smoked fish during storage.

Materials and methods

Three trials were carried out, at the premises of 2 smoked fish processors in Samut Sakhon, between March and May, 1988. The species of smoked fish, used for the trials were as follows:-

Trial 1 - Snakehead (*Ophiocephalus* sp.).

Trial 2 - Lizardfish (*Saurida* sp.).

Trial 3 - Marine catfish (*Arius* sp.).

The insecticides evaluated, were pirimiphos-methyl (Actellic), supplied as a 50% emulsion concentrate by ICI Asiatic (Agriculture) Co., Ltd. and deltamethrin (Cislin), supplied as a 1% suspension concentrate by Wellcome Thailand Ltd. The insecticides were applied by a hand spraygun at the concentrations and approximate dosages given in Table 41.

Table 41. Details of insecticide applications to fish.

Insecticide	Concentration (% w/v)	Dosage (mg/kg)	Volume applied (ml/kg)
Pirimiphos-methyl	0.05	5	10
	0.1	10	10
	0.2	20	10
Deltamethrin	0.01	1	10
	0.02	2	10
	0.03	3	10

Trial 1

Approximately 12 kg of smoked snakehead was purchased from a processor in Nakhon Sawan and transported to the premises of processor 1 (Khun Teuw), in Samut Sakhon. On arrival at the processor′s premises, the fish were divided into 12 batches. The insecticide treated fish were then sprayed with pirimiphos-methyl at the above concentrations. The control fish were sprayed with water only. Each treatment consisted of 3 replicates. The fish were allowed to sun dry for 2 hours, before being weighed and placed in woven bamboo baskets. The baskets were stored in a random pattern at the processor's premises. At the end of the storage period of 9.5 weeks, the fish were inspected by staff of the Thailand Fish Technology Development Division. The fish were examined for *Dermestes* infestation and all adult *Dermestes*, present on the fish, were collected for identification. *Dermestes* larvae were recorded on each fish as being present or absent. Both the weights of the whole fish and fish fragments in the bottom of each basket were recorded using a Precisa top pan balance, accurate to 1g. The shed skins of *Dermestes* larvae were collected from the bottom of each basket and counted, to obtain an index of infestation during the early stages of storage. An attempt was made to estimate insect damage, using the criteria described in Section 6.2.1, but was unsuccessful, due to the counterparts' lack of experience in assessing insect damage.

<u>Trial 2</u>

Approximately 84 kg of smoked *Saurida* sp. was divided into batches of 4 kg before being treated. The insecticides evaluated, were pirimiphos-methyl and deltamethrin. Method details were as for Trial 1.

<u>Trial 3</u>

Approximately 84 kg of smoked *Arius* sp., processed at the premises of Khun Wichai Mink, were divided into 4 kg batches and received the treatments used in Trials 1 and 2. In this case, however, the fish were stored at the premises of Khun Wichai Mink.

Results and discussion

It can be seen from Table 42, that of the 238 *Dermestes* samples collected, approximately 55% were identified as *D. ater*, 42% as *D. maculatus* and the remaining 3% as *D. carnivorus* and *D. frischii*.

Table 42. Dermestid adults collected from stored fish

Insect species	Trial 1. *Ophiocephalus*	Trial 2. *Saurida*	Trial 3. *Arius*
D. ater	35	63	33
D. maculatus	26	65	8
D. carnivorus	1	6	0
D. frischii	0	1	0
Total	62	135	41

$\chi^2 = 27$

Of the 2 species of fish left in store at Site 1, *Saurida* sp. appeared to be more attractive to dermestid beetles ($p < 0.01$).

In Trial 1, all of the fish lost weight during storage (Table 43). The small differences in weight loss between the control fish, which suffered relatively high *Dermestes* infestation and those treated with insecticide, which suffered little infestation, support the view that weight losses were primarily due to dehydration, rather than to insect damage.

Table 43. Data obtained during storage of smoked *Ophiocephalus* sp. (Trial 1).

	Insecticide dose (mg/kg) of pirimiphos-methyl 0	5	10	20
% change in fish weight	-4	-5	-3	-2
% infestation by *Dermestes*	59	0	10	0
No. shed, larval skins	1127	249	248	42

Both indicators of *Dermestes* infestation, demonstrated that spray applications of pirimiphos-methyl, at dosages of 5, 10 and 20 mg/kg, reduced *Dermestes* infestation. The 20 mg/kg dosage gave the best protection against *Dermestes* infestation.

In Trial 2, there was again no apparent relationship between weight change during storage and insecticide treatment (Table 44). The extremely friable nature of the product meant that the most important cause of weight loss was fragmentation during handling.

Table 44. Data obtained during storage of smoked *Saurida* sp. (Trial 2).

	Insecticide dose (mg/kg)						
	Pirimiphos-methyl				Deltamethrin		
	0	5	10	20	1	2	3
% change in fish weight	-2	0	-2	+2	0	-2	+1
% infestation by *Dermestes*	15	1	11	0	1	10	0
No. shed, larval skins	1285	471	418	85	60	120	34

All of the insecticide treatments resulted in a reduction in *Dermestes* infestation and the shed, larval skin counts suggested that the deltamethrin was the more effective of the 2 insecticides, over the concentration ranges tested.

In Trial 3, only the control group of fish showed a decrease in weight over the storage period (Table 45). The other treatments either showed a weight increase or no change, suggesting that the weight loss by the control fish, may, in part, be due to insect infestation.

Table 45: Data obtained during storage of smoked *Arius* sp. (Trial 3).

	Insecticide dose (mg/kg)						
	Pirimiphos-methyl				Deltamethrin		
	0	5	10	20	1	2	3
% change in fish weight	-4	+5	0	+6	0	+2	0
% infestation by *Dermestes*	33	0	6	3	0	0	0
χ^2		53*	45*	59*	53*	78*	78*
No. shed, larval skins	1803	328	228	279	70	20	0
χ^2		1021*	1221*	1116*	1603*	1744*	1803*

*$p < 0.01$

This hypothesis is supported by the insect infestation results, which again demonstrated the effectiveness of the insecticides used, in controlling *Dermestes* infestation.

The weight increases, shown by the 5 and 20 mg/kg pirimiphos-methyl and 2 mg/kg deltamethrin treated fish could be due to water uptake from the atmosphere. However, the absence of weight increases in the other groups of insecticide treated fish, suggest that weighing errors cannot be ruled out.

6.4 Conclusions

1. Blowfly activity was low during the processing trials and resulted in only the S. *commerson* becoming infested with blowfly larvae.

2. Pirimiphos-methyl, applied as a 15 second, 0.03% a.i. dip and deltamethrin, applied as a 6 second, 0.003% a.i. dip, protected *S. commerson* against blowfly infestation during processing.

3. A 15 second, 0.03% a.i. dip reduced *Dermestes* infestation of salted, dried fish over a 15 weeks storage period.

4. During a pilot experiment, untreated, smoked, marine catfish became heavily infested by *D. maculatus* and *D. ater* and suffered physical losses of approximately 25% over a 15 weeks storage period.

5. A pilot experiment indicated that a spray application of 0.003% deltamethrin was effective in preventing *Dermestes* infestation of smoked, marine catfish over a 15 weeks storage period.

6. Subsequent field trials, demonstrated that spray treatments with pirimiphos-methyl, at dosages of 5 - 20 mg/kg and deltamethrin, at dosages of 1-3 mg/kg were effective in reducing *Dermestes* infestation of 3 species of smoked fish over a 9.5 weeks storage period.

7. Both the pilot experiment and subsequent field trials, showed that the spray application of deltamethrin, at 1-3 mg/kg, was more effective in protecting against *Dermestes* spp. during storage, than the pirimiphos - methyl at higher concentrations.

7. GENERAL BIOLOGICAL RESULTS ON *C. MEGACEPHALA*, OBTAINED DURING FIELD TRIALS AND LABORATORY INVESTIGATIONS

7.1 Introduction

C. megacephala is widely distributed throughout the Oriental and Australasian regions and also occurs in neighbouring parts of the Palaearctic region, e.g., China and Japan. It is not, as yet, widespread in Africa, but a few specimens, probably introduced by shipping, have been identified in Ghana, Senegal, Mali and South Africa (Prins, 1979), and the species has also been recently introduced to the Canary Islands (Baez *et al.*,1981). *C. megacephala* is commonly found near human dwellings and creates a great nuisance in fish markets and other areas where fish is handled (Zumpt, 1956). During the course of the fieldwork conducted in South East Asia, *C. megacephala* has been observed at all salted, dried fish processors' premises, in 7 provinces of Indonesia and 3 provinces of Thailand and is the species of blowfly most commonly associated with cured fish sites in South East Asia. Although *L. cuprina* was frequently observed at fish processing sites, the results given in Table 5 and field observations suggested it was a pest of minor importance, compared with *C. megacephala*.

The life history of *C. megacephala*, based on laboratory observations, has been described by Wijesundara (1957a), who has also studied the effect of humidity on longevity of the adults (Wijesundara, 1957b). Studies of the seasonal occurrence of *C. megacephala* in West Bengal have been carried out by Roy and Dasgupta (1975). A further investigation, of seasonal occurrence of *C. megacephala* in Calcutta, was also carried out (Das and Dasgupta, 1982a), and in addition, Das and Dasgupta (1982b), studied sex ratios of specimens caught in traps, set in a zoological garden in Calcutta. Several taxonomic and distribution studies have been conducted on *C. megacephala* in the Oriental and Australasian regions, Kurahashi (1970, 1971, 1972, 1977, 1979a, 1979b, 1982), Singh *et al.* (1979).

C. megacephala is primarily regarded as a pest of medical and veterinary importance (Tumrasvin *et al.*, 1979) and although adults have been described as causing a nuisance in areas where fish is handled (Zumpt, 1956), there have been no references to *C. megacephala* larvae infesting and causing losses in traditionally processed fish. In view of the finding that *C. megacephala* is a major cause of losses in salted-dried fish in South East Asia, further field and laboratory data on the biology of the fly, particularly in relation to its status as a pest of cured fish, were collected during the course of the infestation reduction experiments and are described below.

7.2 Comparison of sex ratios of laboratory reared flies, with flies recorded visiting fish during processing in Indonesia

Materials and methods

Laboratory populations of *C. megacephala* were established from pupae, imported from Indonesia. Details of maintenance are given in Section 8. Dead flies were routinely collected from the cages, and at the end of the useful life of each cage population, the flies were killed and sexed.

Field observations were made at the premises of Hadji Sonja, in Cirebon, West Java, during the processing of *A. thalassinus*. The numbers of male and female *C. megacephala*, present on the fish during drying, were routinely recorded at 15 or 30 minute intervals. In addition, counts were made of male and female flies present on the fish during salting. Accurate sexing, by eye morphology, could be done without catching or disturbing the flies.

Results and discussion

It can be seen from Table 46, that male and female flies were produced in approximately equal numbers in the laboratory cultures. A paired Student t-test, demonstrated that the small difference, between the number of male and female flies, was not significant ($p > 0.05$).

Table 46. Sex ratios of laboratory reared *C. megacephala*

Cage population	Number Males	Number Females	Ratio of male to female flies
1	34	22	1.5
2	182	179	1.0
3	131	196	0.7
4	121	133	0.9
5	253	210	1.2
6	46	77	0.6
7	128	112	1.1
8	277	227	1.2
9	766	725	1.0
10	191	183	1.0
11	724	654	1.1
12	167	217	0.8
13	144	151	0.9
14	98	79	1.2
15	244	232	1.0
16	278	272	1.0
17	299	195	1.5
18	318	334	1.0
19	231	276	0.8
Total =	4,632	4,474	

Wijesundara (1957a), recorded sex ratios of cage populations of *C. megacephala*, derived from gravid female flies, trapped at a local market, and found a slight preponderance of males over females in every case. The mean proportion of males to females was 1.13 to 1, and a chi^2 test, applied to his results, demonstrated a significant difference between the numbers of males and females (X^2=4.26, $p<0.05$).

Table 47 shows that the incidence of female *C. megacephala* was far higher than the incidence of males, during trials conducted at the processing site in Indonesia, indicating a behavioural difference between the sexes. This can be explained by the female flies having both a higher protein requirement than the males, in order to produce eggs, and by the presence of gravid flies, using the fish as oviposition medium.

Table 47. Proportion of female *C. megacephala*, expressed as percentages of the total daily blowfly counts recorded on *A. thalassinus*, during the course of 11 processing trials in Indonesia

Proportion of female flies (% total count) on fish during salting		Proportion of female flies (% total count) on fish during drying	
99	(121)	96	(452)
100	(182)	92	(222)
100	(15)	94	(476)
100	(114)	96	(111)
100	(159)	91	(476)
100	(53)	91	(1285)
100	(95)	92	(906)
100	(40)	97	(190)
		89	(1089)
		92	(706)
		96	(2936)

Actual total counts given in parentheses.

Das and Dasgupta (1982b), investigated sex ratios of blowflies in Calcutta and found the sex ratio of captured specimens to be dependent on the nature of the bait used in the fly traps. Almost equal numbers of male and female *C. megacephala* adults were trapped on baits of ripe jackfruit, ripe mango and molasses, whereas 85.5% of the flies captured by the traps containing fish were female. It would seem that the sugar rich baits are used as an energy source by both sexes and that traps, containing such baits, give a more accurate representation of the real proportions of female and male flies in the population. Roy and Dasgupta (1975) also found that fish baited traps yielded a far higher proportion of female flies than males, when they monitored seasonal occurrence of flies in West Bengal. In view of the finding that male and female *C. megacephala* are produced in equal numbers, the only other possible explanation for a significant difference between the real proportions of the two sexes, as opposed to the proportions at a particular site or attracted to a particular bait, would be differential mortality rates. Details of investigations into the mortalities of male and female flies are given in

Section 7.6 and indicate that there is no difference between the mortalities of laboratory reared, male and female *C. megacephala* adults, over periods of up to 98 days.

7.3 Ovary stages of female flies visiting *A. thalassinus* during processing

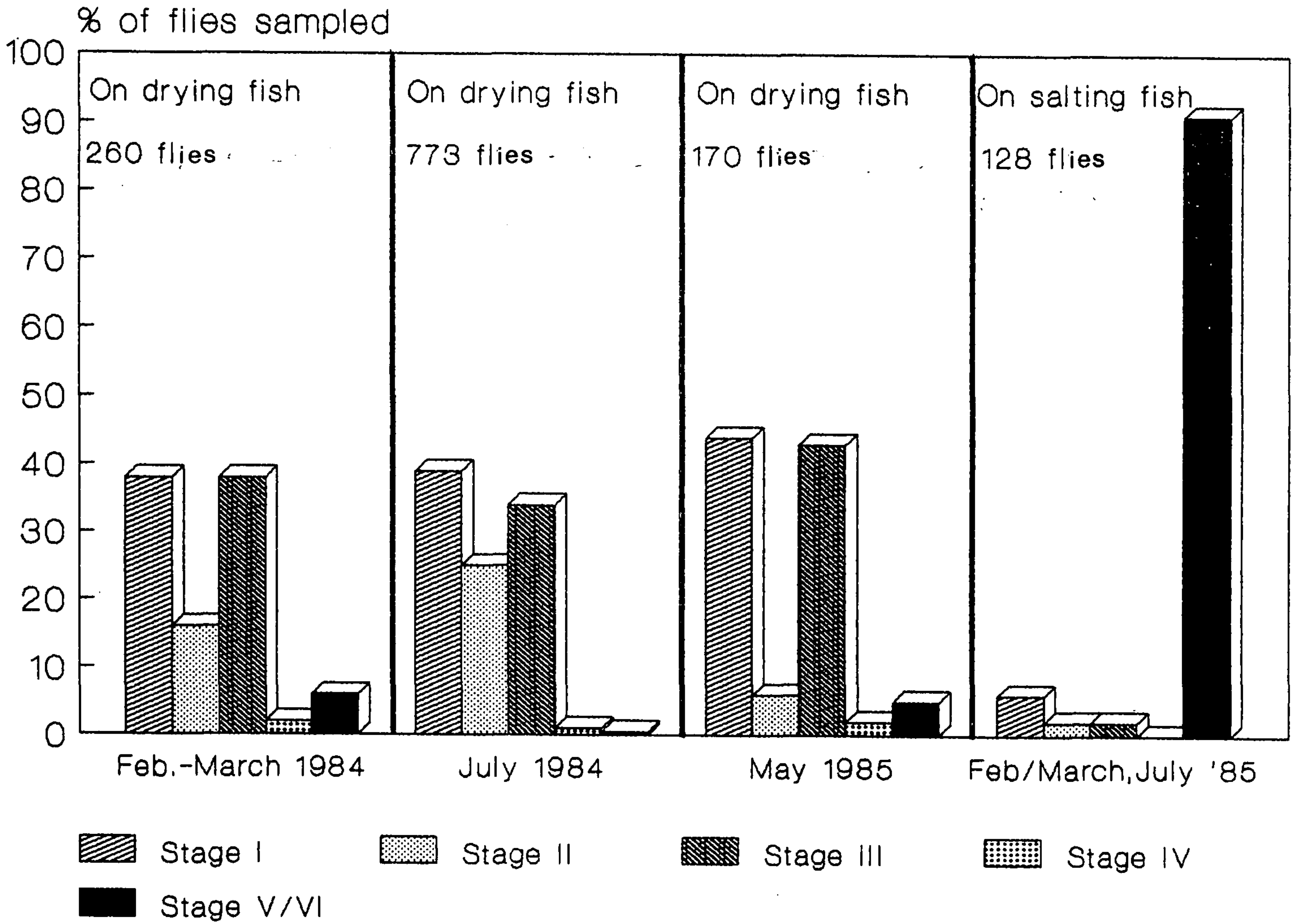
Materials and methods

Female flies were collected from fish in the salting tank and fish on the drying racks during the first 2 days of drying. The flies were captured manually, with the aid of a small, polythene bag. This method was found to be more efficient, under normal processing conditions, than either water or bottle traps or hand netting. After killing by ethyl acetate, the flies were preserved in 70% alcohol and returned to Grimsby. In the laboratory, the ovaries were removed and staged, according to oocyte characteristics, based on morphological criteria described in Trepte and Trepte-Feuerborn (1980). Because of the difficulty in discriminating between stages V and VI, these were treated as one stage and coded stage V/VI.

Results and discussion

Figure 36 shows the distribution of ovary stages of flies collected from fish on the drying racks, during trials conducted in Indonesia in February-March 1984, July 1984 and May 1985, and also gives the distribution of stages, obtained from flies collected from fish in the salting tank, during 1985. It can be seen that the majority of female flies, present on the drying fish at each of these different seasons, were sexually immature, whereas the majority of flies, on the fish being salted, were gravid. It can be concluded that most of the flies on the drying fish were there to feed and that the flies on the salting fish were primarily using the fish for oviposition. It was at the salting stage, that the fish appeared to be most attractive to ovipositing blowflies and as the flies were attracted indoors to the fish in the salting tanks, it can be surmised that at the salting stage of processing, *A. thalassinus*

Fig.36: Ovary stages of *C. megacephala* collected from *A. thalassinus*.



releases a volatile substance that has a powerful, differential attractive effect on gravid *C. megacephala* females.

The nature of attractant volatiles, released by fish during processing, warrants further investigation, with a view to developing a lure, which could be mixed with an insecticide and evaluated as a method of blowfly control.

7.4 Development and egg capacities of ovaries of laboratory reared *C. megacephala*

Materials and methods

A population of 1000 flies, was maintained in a cage, at a mean temperature of 28°C and reared on fresh, bloody liver, which was presented to the flies on alternate week days. Sucrose and water were made constantly available to the flies. At weekly intervals, after adult emergence, 20 females were removed, killed and dissected to remove the ovaries. As the main aim of the investigation was to determine the time necessary for the ovaries to reach maturity, the immature eggs were staged using a simplified version of the method given above, and divided into stages I, II/III, III/IV and V/VI. In addition, the numbers of eggs present in the ovaries of 5 female flies, were recorded each week.

Results and discussion

It can be seen from Table 48, that the oocytes of *C. megacephala*, can reach maturity within 1 week of emergence. In the main, however, there was synchronous oocyte development, and between 2 and 3 weeks were necessary, for the majority of flies in the culture, to reach maturity. The period required for oviposition to first occur, varied from 7 to 10 days, which compares with a period of 8 to 9 days observed by Wijesundara (1957a).

Table 48. Oocyte development in ovaries of laboratory reared *C. megacephala*. 20 flies examined each week.

Culture age (weeks)	% of flies at each stage of ovary maturity I	II/III	III/IV	V/VI
1.	45	30	15	10
2.	15	20	25	40
3.	0	0	10	90
4.	0	0	10	90
5.	0	0	0	100
6.	0	0	5	95

The numbers of mature oocytes (stages V/VI) in the ovaries of *C. megacephala* samples, removed from the cage population at weekly intervals, between weeks 2 and 6, are given in Table 49. A two-way analysis of variance of this data demonstrated no significant difference between the egg capacities of replicate samples of the same age, or between replicates of different ages over the period of 6 weeks ($p > 0.05$ for both variables).

Table 49: Total number of eggs present in mature ovaries of laboratory reared *C. megacephala*.

Sample number	Age of samples (weeks) 2	3	4	5	6
1.	224	162	151	259	222
2.	215	228	209	200	254
3.	169	264	258	208	197
4.	238	260	189	266	191
5.	222	288	180	248	237
Mean no.	214	240	197	236	220

The overall mean of the number of eggs present in the mature ovaries was 221 with a range of 151 to 288. This compares with a mean of 254 and a range of 224 to 325, reported by Wijesundara (1957a). Roy (1938) obtained a mean egg count 182 and a range 136 to 211 from gravid females caught in local markets in Calcutta. Differences in mean egg numbers, between the 3 sets of data, almost certainly reflect size differences in the adult female flies. Williams and Richardson (1983), found that the number of ovarioles in *L. cuprina*, *Calliphora stygia*, *C. vicina* and *C. hilli*, was dependent on the pupal size of the samples dissected. Large flies, which had emerged from large pupae, contained higher numbers of ovarioles, than small flies, which emerged from small puparia, produced by larvae which had been subjected to food shortage . Oocyte size, however, was found to be independent of pupal size and was not influenced by food availability during the larval stage.

7.5 Effect of presence of *C. megacephala* eggs on oviposition by *C. megacephala* on fish

Materials and methods

A colony of approximately 550 laboratory reared *C. megacephala* was presented on 18 occasions, over a 3 week period, with a choice of a piece of unsalted cod*, spiked with 50-60 mg of recently laid *C. megacephala* eggs and unsalted cod containing no eggs. The fish, which had previously been cut into blocks of 16cm^2, was, on each occasion, exposed to the culture for 20 minutes. At 5 minute intervals, counts were made of the number of flies ovipositing on each piece of fish. At the end of each trial, the pieces of fish were removed from the cage and the weight of eggs present on each piece was recorded. During the period of the trials, the culture was maintained at a mean temperature of 28°C and a mean relative humidity of 37%.

A series of trials to investigate whether the attractive stimulus, exerted by *C. megacephala* eggs, was visual or olfactory, was subsequently carried, out using a laboratory reared *C. megacephala* population, containing 1000 flies. In each trial, the flies were presented with 3 identical dishes, lined with 3 layers of moist tissue paper. 1 of the dishes contained a known weight of fresh *C. megacephala* eggs, the 2nd dish contained a similar, known weight of eggs that had been boiled in water for 30 minutes and the 3rd dish, the control, contained nothing. The dishes were introduced to the cages in random order and left for 1

* Gadus morhua

hour. At 10 minute intervals, the numbers of male and female flies in each dish were counted and, at the end of each trial, the dishes were removed and the number of newly laid eggs in each dish was recorded.

Results and discussion

The data in Table 50, show the effects of spiking fish with freshly laid *C. megacephala* eggs, on oviposition by *C. megacephala* and indicate that more ovipositing flies were attracted to the spiked fish, resulting in more eggs being oviposited on the fish previously spiked with eggs than on the unspiked fish. Group oviposition was apparent in all of the trials and the sequence of events, was that once one fly had commenced oviposition, it was quickly joined by others, which would crowd together and oviposit on the same site. This resulted in a single, large mass of eggs, originating from several parents.

A one tailed, paired Student t test, demonstrated that the mean ovipositing blowfly count was significantly higher ($p<5\%$) on the fish spiked with eggs and that the weight of eggs, oviposited on the spiked fish, was also significantly higher ($p<5\%$).

The results of a further investigation into whether the attractive stimulus of the eggs was olfactory or visual are given in Table 51 and indicate that oviposition was significantly higher (Student t-test, $p<1\%$) in the dishes containing the fresh eggs, than in the dishes containing the boiled eggs. However, the significantly higher number of eggs oviposited in the dish containing the boiled eggs ($p<1\%$), compared with eggs oviposited in the empty dish, suggests a visual stimulus is also involved

Table 50. Effect of spiking fish with freshly laid *C.megacephala* eggs, on oviposition by *C. megacephala*

Trial	x number of ovipositing flies: spiked fish	x number of ovipositing flies: unspiked fish	Weight of eggs (mg)* oviposited on: spiked fish	Weight of eggs (mg)* oviposited on: unspiked fish
1.	10.5	0	289.8	0
2.	4.0	4.0	117.4	93.5
3.	4.2	3.7	72.0	53.2
4.	5.0	0.7	148.3	27.7
5.	4.0	0	52.9	0
6.	6.0	0.2	94.2	2.0
7.	8.0	1.5	157.7	36.2
8.	5.0	8.2	86.7	199.9
9.	2.2	8.0	46.7	159.6
10.	17.0	0	328.6	0
11.	2.7	0.2	78.9	3.7
12.	0.5	5.5	0.3	93.7
13.	0	2.5	0	68.6
14.	3.7	0.5	42.5	4.9
15.	6.0	0.2	111.2	0.8
16.	3.5	0	38.6	0
17.	2.7	0	102.6	0
18.	3.7	0	42.7	0
Mean	4.9	2.0	100.6	41.3

* 1mg = 9 eggs

Table 51: Comparison of oviposition by *C. megacephala* on fish spiked with fresh eggs with oviposition on fish spiked with eggs that had been boiled for 30 minutes

Trial	No. eggs laid in dish with fresh eggs	No. eggs laid in dish with boiled eggs	No. eggs laid in dish with no eggs
1.	19	1	0
2.	4	0	0
3.	0	4	0
4.	7	1	0
5.	0	0	1
6.	0	0	0
7.	1	2	0
8.	0	0	0
9.	0	0	0
10.	3	3	1
11.	0	0	0
12.	18	0	0
13.	20	10	0
14.	19	6	2
15.	15	4	4
16.	20	9	0
17.	9	2	0
18.	19	15	4
19.	24	18	6
20.	20	22	2
21.	17	14	5
22.	20	9	0
23.	14	4	0
24.	10	10	0
Mean no	10.8	5.6	1.1

dendum

an counts of male and female flies, recorded on each eatment during the course of 24 trials, were as follows:

Sex	Dish containing fresh eggs	Dish containing boiled eggs	Empty dish
male	18.9	14.5	8.3
le	14.8	9.7	7.5

paired Student t-test, demonstrated that the numbers of th sexes, were significantly higher (P< 5%) in the dish ntaining the fresh eggs, than in the empty dish and the sh containing the boiled eggs.

cavities and that the presence of ovipositing flies attracted and stimulated further oviposition in other flies.

It was observed during the *C. megacephala* oviposition trials, described in Section 8, that group oviposition most frequently occurred either in narrow gaps between the fish muscle blocks or between the undersurface of the fish and the floor of the dish. Group oviposition and the location of oviposition in cavities, undoubtedly confers a survival advantage, in that the eggs at the centre of a large mass will be less liable to desiccation than those laid as an individual batch and the choice of small cavities for oviposition will also reduce the possibility of desiccation.

In a further investigation into the nature of group oviposition by *L. cuprina*, Barton-Browne *et al.*(1969) concluded that the stimulus produced by ovipositing females, which caused other females to commence oviposition, was almost certainly chemical in nature and that group oviposition also had a strong physical contact component. The experiments on *C. megacephala*, described in this section, support the view that oviposition is initiated by a chemical stimulus, released by recently laid eggs and that group oviposition is stimulated by the presence of females which are already ovipositing. It would seem reasonable to assume that the chemical stimulus is a pheromone which is released during oviposition. A pheromone has been found to stimulate group oviposition in locusts (Norris, 1964) and there have been several reports of pheromones influencing oviposition behaviour in mosquitoes. Further work on isolating and synthesising chemical analogues of pheromones that stimulate oviposition by blowflies, could be usefully carried out in tandem with investigations into the nature of attractant volatiles, given off by fish, with a view to developing a lure that could be used to attract blowflies away from fish during processing.

7.6 Mortality of laboratory reared *C. megacephala* adults and pupae

Materials and methods

Adult mortality was monitored during the lifespans of 3 cage populations, by recording, at regular intervals, the numbers of dead, male and female flies present in each cage. Temperature and relative humidity were monitored and the flies were given fresh, bloody liver on alternate weekdays, as well as a constant supply of sucrose and water.

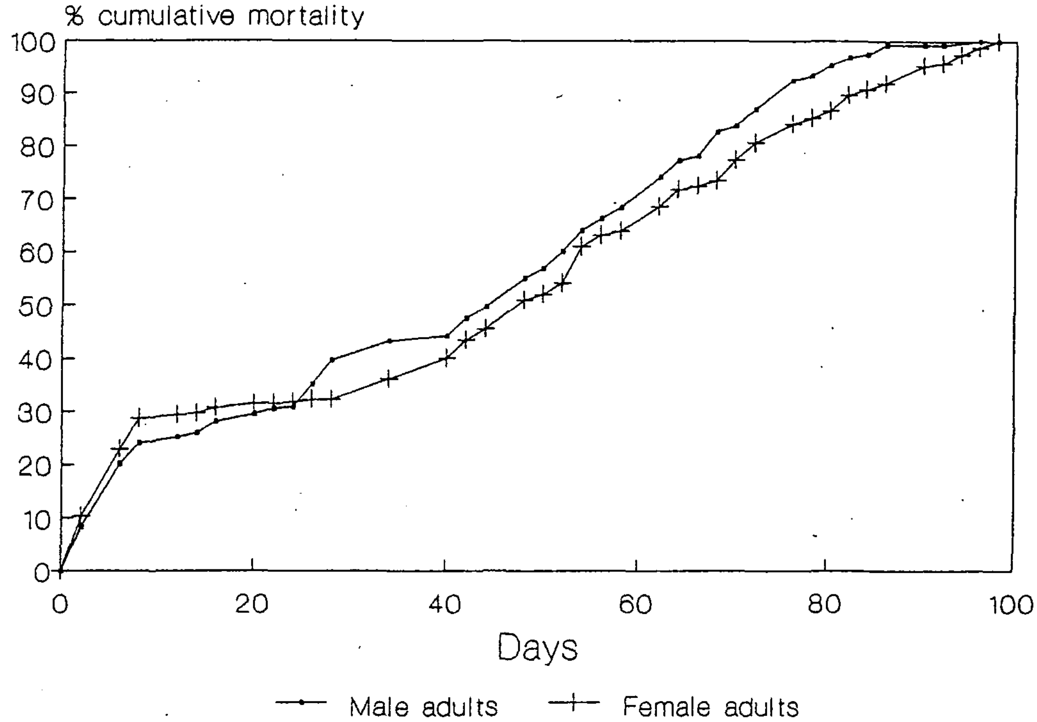
Results and discussion

Figures 37 to 42 give the mortality curves and environmental conditions* of 3 laboratory cultures of *C. megacephala*. The mean lifespans of the flies in the 3 cultures ranged from 47 to 54 days and the longest survival times were 98, 80 and 82 days respectively. These values compare with an average lifespan of 57 days and a maximum duration of 96 days, reported for laboratory cultures of *C. megacephala*, raised under similar environmental conditions by Wijesundara (1957a). There was no apparent difference between the mortality rates of the male and female flies. Two of the populations (see Figs.37 and 41) showed relatively high mortalities during the first 7 days, but in all 3 cultures the mortality rates between days 7 and 40 were relatively low. After 40 to 45 days, the mortality rates tended to increase. Wijesundara (1957b) also found no significant variation between the lifespans of males and females and reported that the mortality was slight and gradual up to 30 to 40 days. In the same series of experiments, Wijesundara (1957b) indicated that the lifespan of *C. megacephala* was longer when cultured at relative humidities of 40 and 60%, than at higher humidities.

Larval and pupal mortalities were monitored during the investigations into the effects of salt content and are discussed in Section 8.

* Spot measurements of temperature and relative humidity taken at daily intervals.

Fig. 37: Mortality of laboratory reared, adult *C. megacephala*



Population 1

Fig.38: Environmental data for Population 1

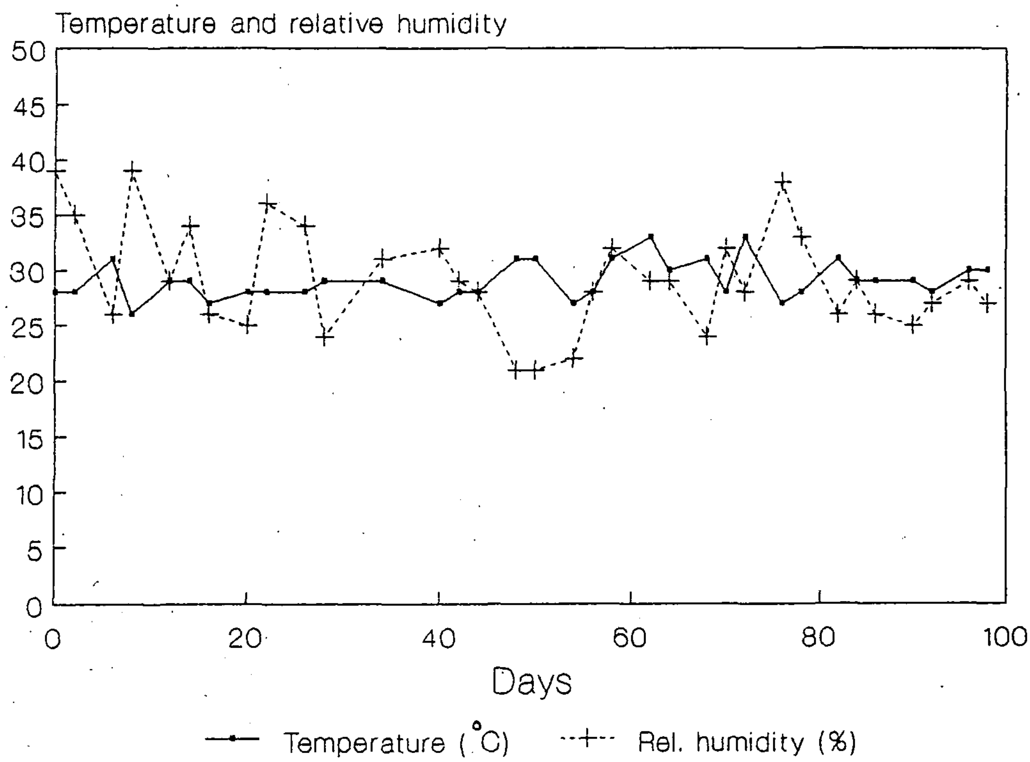
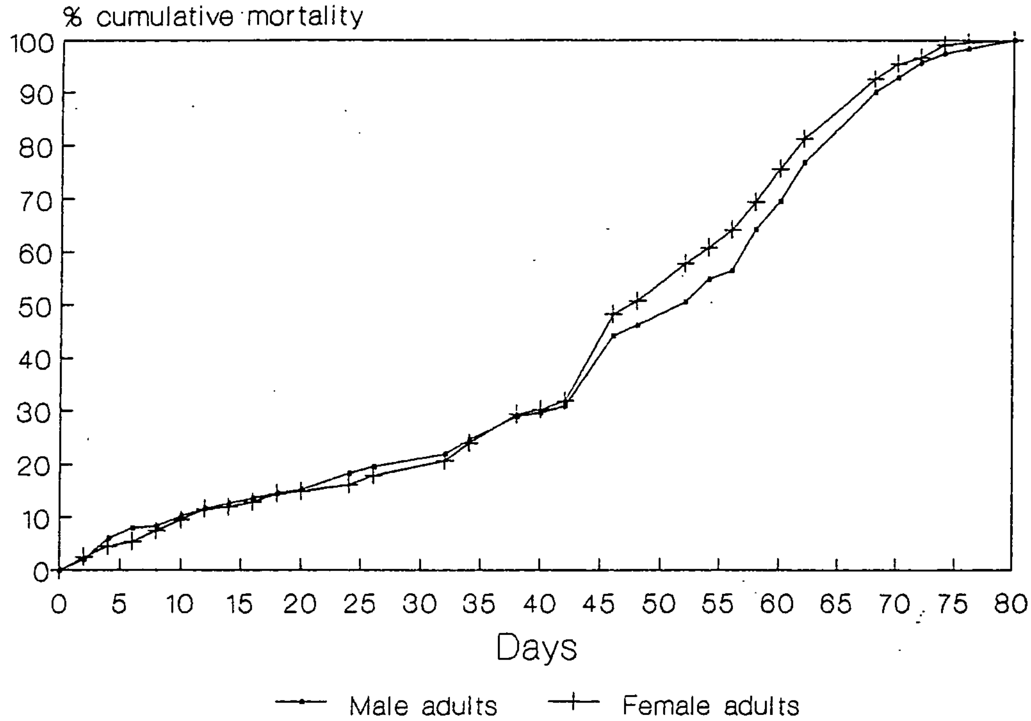


Fig.39: Mortality of laboratory reared, adult *C. megacephala*



Population 2

Fig.40: Environmental data for Population 2

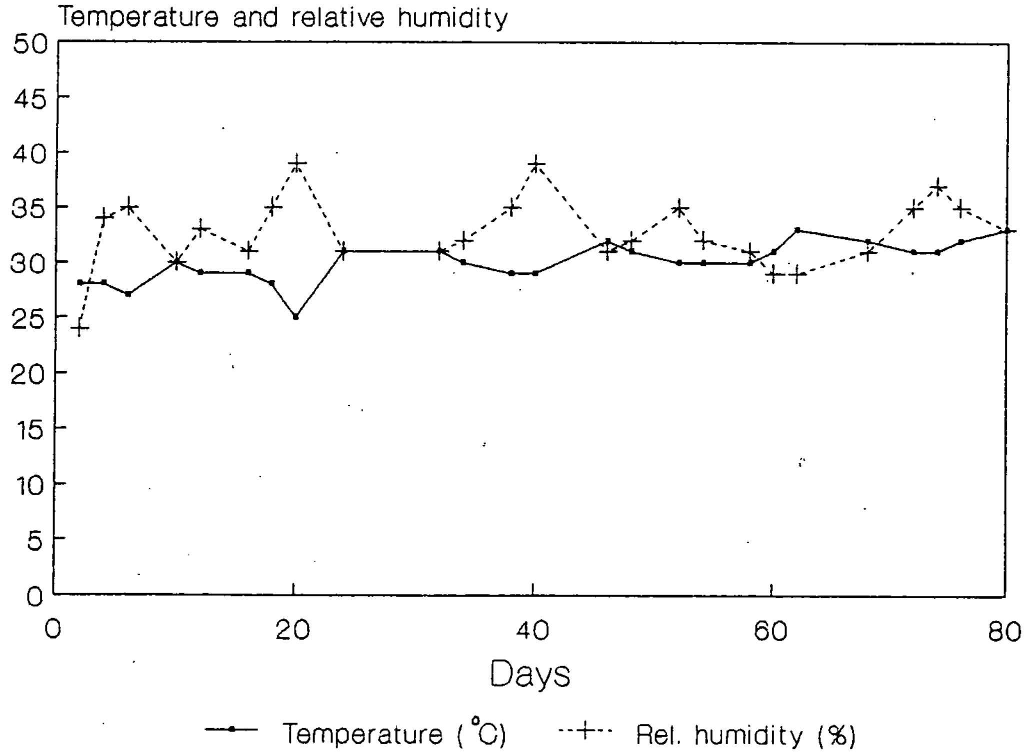
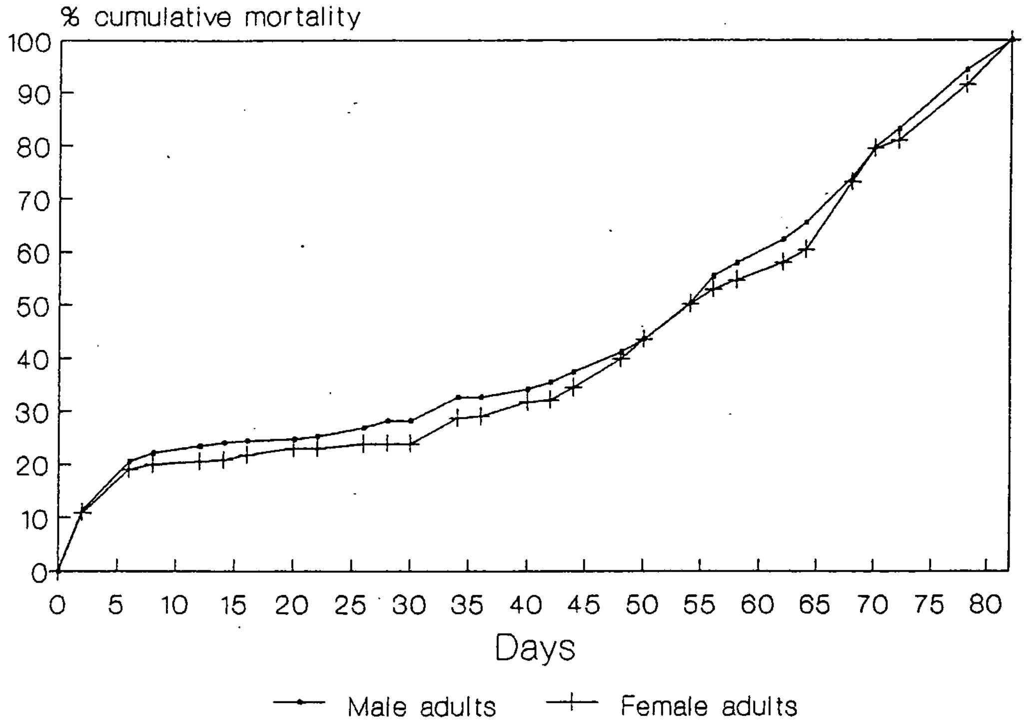
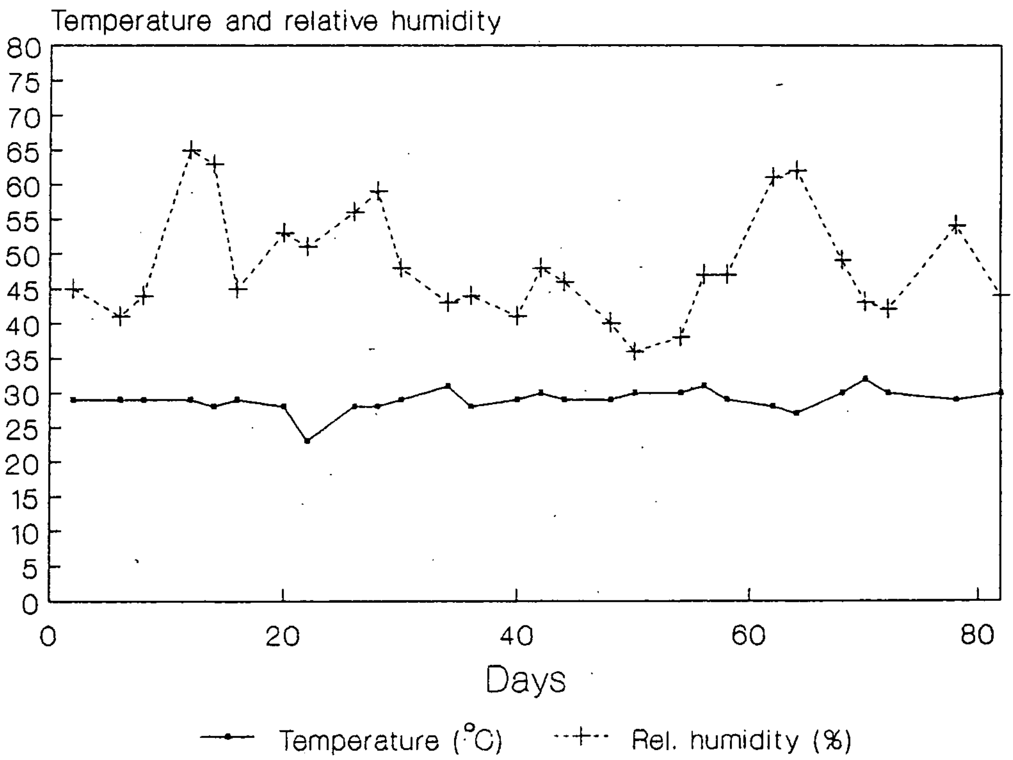


Fig.41: Mortality of laboratory reared, adult C. megacephala



Population 3

Fig.42: Environmental data for Population 3



7.7 Seasonal variation in *C. megacephala* numbers at fish processing site in Cirebon

Materials and methods

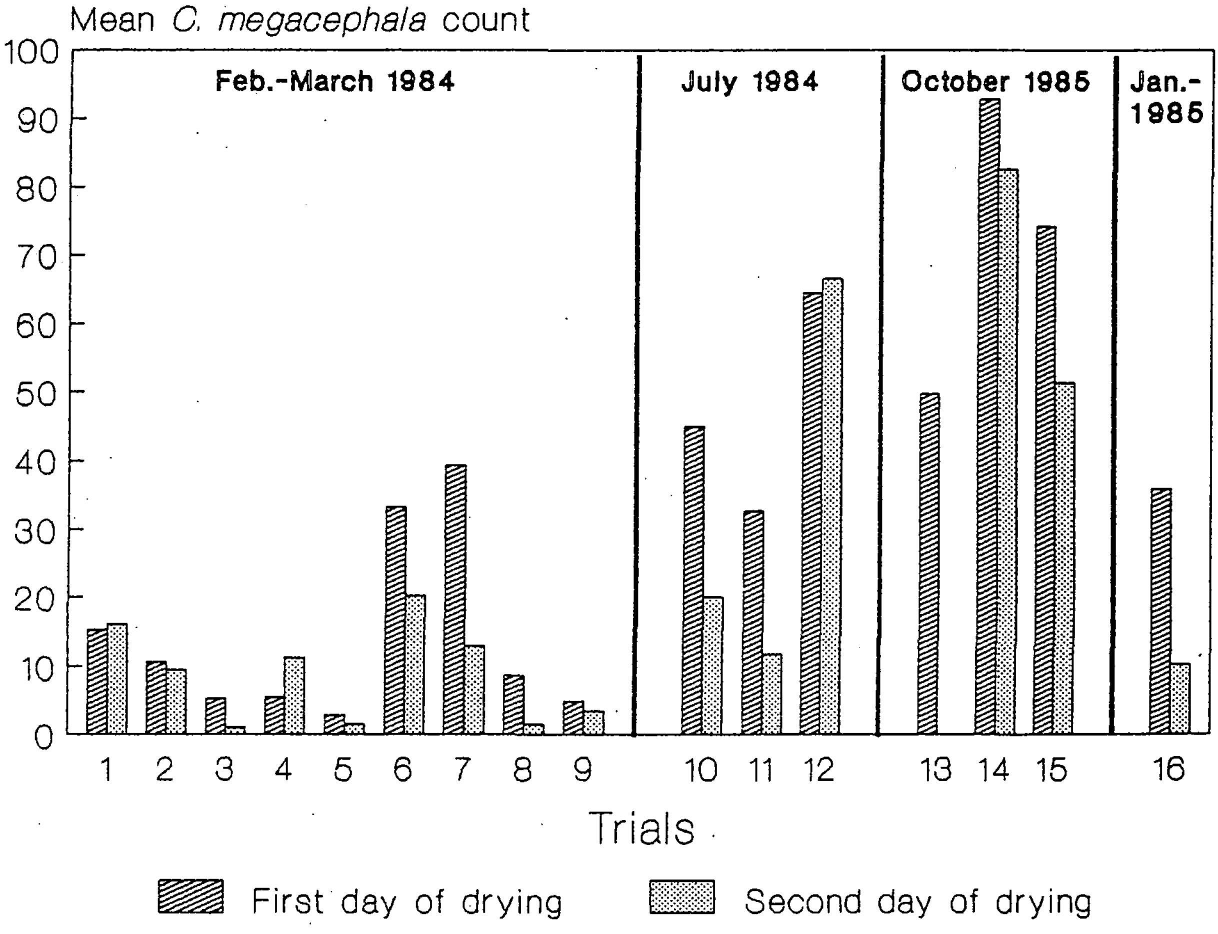
An estimate of the seasonal variation in blowfly numbers was obtained from counts of *C. megacephala* present on *A. thalassinus* during the first 2 days of drying. Counts were made of flies in contact with the fish at 15 or 30 minute intervals and the mean blowfly count for each day was used as an index of blowfly activity. Blowfly activity was monitored during the course of 9 fish processing trials in February and March (wet season) 1984, 3 trials in July (dry season) 1984, 3 trials in October (dry season) 1985 and 1 trial in January (wet season) 1985.

Results and discussion

Observations made at the processing site, during the course of the loss reduction trials conducted between 1984 and 1986, revealed that blowfly activity could vary from day to day, for no apparent reason. No correlations were demonstrated between blowfly activity and temperature, relative humidity, or the number of fish on the drying racks. The blowfly counts, shown in Figures 9, 10, 22, 23, 24, 27, 32 (Section 4) and 33 (Section 5,) show a tendency for blowfly activity to increase during the afternoon and also suggest that activity is higher during overcast conditions. In most cases, activity was at a minimum around the middle of the day. Although blowflies, such as *C. megacephala*, are undoubtedly well protected against the adverse effects of insolation by their reflecting skin surface and thick cuticle, the observed behaviour can be explained as a result of avoidance, by the flies, of the most intense heat and dryness of the day. Hygrophilic blowflies, such as *Calliphora* sp., may develop a clear bimodality of activity during hot periods (Nuorteva, 1965). Bimodality of flight curves may develop in any area, where the temperature around noon, exceeds the thermal preference of a given species. Large, daily fluctuations in blowfly activity are perhaps not surprising, as the processing site was located in a generally insanitary, urban area, close to a refuse dump. In addition, a number of other fish processors were located in the same area and any variations in their processing activity would have affected the blowfly counts on the fish under observation.

Figure 43 shows the mean *C. megacephala* counts (total *C. megacephala* count divided by the number of counts taken) recorded for the first and second drying days of 4 series' of trials, conducted between 1984 and 1985. Although, for the reasons discussed above, such counts cannot be expected to give an accurate estimate of blowfly activity at any one time, they do suggest that blowfly activity was higher in July and October than in January, February and March. This is surprising, as the consensus in the literature, is that blowfly activity is usually higher in the rainy season (November to March in Indonesia), than in the dry season (April to October). The evidence supporting this hypothesis is open to question, as few actual measurements of seasonal variation of blowfly activity have been taken in the tropics. Anggawati *et al.* (1986), monitored fly activity at 2 fish landing sites in Jakarta. They reported that fly abundance was related to availability of breeding sites and seasonal factors, and was highest during the rainy season. While their data on *M. domestica* activity, at one of the sites, supported this conclusion, the numbers of *C. megacephala*, caught on sticky traps, failed to demonstrate any seasonal variation. Roy and Dasgupta (1975), studied seasonal occurrence of flies in an urban in West Bengal, India and found *C. megacephala* to be the most common species present. It was present throughout the year, but was most active from May to October (summer and autumn) when both temperature, relative humidity and rainfall were high. Data collected by Power and Melnick (1945) and Dicke and Eastwood (1952) suggest that fly populations in urban areas may be less stable than in rural areas. Even in rural areas, human activity can generate large fluctuations in local blowfly populations. Nuorteva (1971) recorded a mass occurrence of *Phormia terraenovae* R.-D (Diptera, Calliphoridae), caused by a rendering plant

Fig.43: Mean counts of *C. megacephala* on *A. thalassinus* over first 2 drying days.



storing slaughterhouse waste in the open. The mass occurrence was found to extend over a radius of 4 km around the plant. The breeding media available in domestic and food processing refuse will fluctuate unpredictably, and Norris (1965), suggests that fluctuations in populations of urban flies may be due to changes in practices of refuse disposal. Observations in Cirebon, suggest that variations in blowfly occurrence, probably result from a combination of man´s activities and seasonal factors.

Blowfly larvae require moist, decomposing, organic animal matter as food and their survival will be a function of both the quantity of food available and the length of time it is moist enough to support the larvae. In the rainy season, when fish landings are down, the total amount of available fish will be decreased, but as drying rates may also be reduced, the fish will remain sufficiently moist to both attract ovipositing blowflies and support blowfly larvae for a longer period, which will result in an increase in the local blowfly population. During the dry season, more fish is processed, but drying rates are increased, decreasing both the period during which the fish attract ovipositing blowflies and the length of time the fish will support the larvae.

Variations in the standard of hygiene of a processor (and of his neighbours) can also influence the occurrence of blowflies at the processing premises. Discarded fish offal, domestic waste and blocked drains all too frequently attract and offer breeding sites for blowflies in processing areas and an improvement in hygiene could go some way to reducing the problem of insect infestation of fish during processing. However, fish processing sites are usually located in the poorest, least hygienic parts of town and no matter how carefully an individual processor keeps his premises clean, there will still be an inexhaustible supply of blowflies from the neighbourhood.

Processors frequently report that the blowfly problem is at its worst during the rainy season and that they do not find it necessary to apply insecticides to their fish in the dry season. The results of the field trials described in Sections 3 and 4 suggest that this is more likely to be due to different fish drying rates in the rainy and dry seasons than to seasonal variations in the numbers of blowflies.

7.8 Conclusions

1. There was no significant difference between the numbers of male and female offspring produced by laboratory reared *C. megacephala*.

2. The incidence of female *C. megacephala* at the fish processing site was always significantly higher than the incidence of males.

3. The ovaries of the majority of female *C. megacephala*, collected from fish on the drying racks, did not contain mature eggs.

4. The ovaries of the majority of female *C. megacephala*, collected from fish in the salting tanks, contained mature eggs.

5. The eggs of the majority of laboratory reared flies matured within 3 weeks and the mean egg content of the ovaries was 221.

6. Oviposition on fish spiked with recently laid *C. megacephala* eggs was significantly higher than on fish which had not been spiked with eggs.

7. Recently laid *C. megacephala* eggs had a significantly higher stimulatory effect on oviposition than eggs which had been previously boiled for 30 minutes.

8. The mean lifespans of laboratory reared *C. megacephala* cultures ranged from 47 to 54 days and the longest survival times ranged from 80 to 98 days.

9. There was no significant difference between the mortality rates of the male an female flies.

10. Blowfly activity at the processing site fluctuated from day to day and appeared to be higher in the dry season than in the rainy season.

8. LABORATORY INVESTIGATIONS INTO THE EFFECTS OF SALT CONCENTRATION ON OVIPOSITION AND LARVAL GROWTH AND MORTALITY AND PUPAL MORTALITY IN *C. MEGACEPHALA*.

8.1 Introduction

The field experiments, described in Sections 3-5, showed that salting fish during processing, gave no protection against insect infestation during processing and storage. *C. megacephala* was frequently observed to readily oviposit on fish, both during and after salting, and *C. megacephala* larvae appeared to be tolerant of the high salt concentrations found in both the pickles used for curing, and in the salted fish itself.

Subsequent observations, in 7 provinces of Indonesia and 3 provinces of Thailand, suggest that salt tolerant *C. megacephala* is widely distributed in areas of South East Asia, where fish are traditionally processed by salting and drying (Esser *et al.*, 1988b). In view of the consensus in the literature, that salting has an inhibitory effect on insect infestation and that heavy salting provides complete protection against blowfly larvae (FAO, 1981), it was decided to further investigate the relationship between fish salt concentration and blowfly infestation of salted fish, by carrying out a series of laboratory experiments using *C. megacephala* cultures, established from pupae imported from W. Java, Indonesia. The experiments concentrated on the effects of fish salt content on *C. megacephala* oviposition and larval growth and mortality and pupal mortality.

8.2 Salting treatment of fish used as oviposition and feeding medium.

Cod (*Gadus morhua*), was used as the oviposition/feeding medium, because it has a similar flesh appearance and texture to the flesh of marine catfish (*Arius thalassinus*), which is a commonly salted and dried species in South East Asia, and is particularly vulnerable to blowfly infestation during processing.

The cod fillets were initially submerged overnight in freshwater and then submerged for a further night in brine solutions which ranged in salt concentration from 0% to 30% (w/v). This procedure was similar to the initial processing treatment carried out by traditional fish processors in West Java. After salting, the fish used for the oviposition experiments, was cut into squares of approximately 16 cm^2, sealed in polythene bags and stored at -20°C. The fish used in the larval growth and mortality experiments, was minced and thoroughly mixed before being sealed in polythene bags and stored at -20°C.

Before freezing, samples were taken randomly and analysed for salt and moisture content by using the methods described by FAO (1981). For salt analysis, approximately 2g of representative sample was weighed and macerated in distilled water for about 2 minutes. The extract was transferred quantitatively into a 250 ml volumetric flask and made up to volume. 25 ml aliquots were then titrated against 0.1 N $AgNO_3$, using potassium chromate indicator. Moisture content was determined by taking 2-5 g of a representative sample of flesh and placing it into a previously weighed dish. After re-weighing, the sample was then placed in an oven, set at 105°C and left overnight. On removal from the oven, the sample was allowed to cool for 30 minutes in a desiccator, before being finally re-weighed. The salt and moisture contents of the 4 batches of fish, used for the experiments, are given in Table 52.

Table 52: Salt concentration of the brine used to salt the fish and the mean salt concentration (% dwb) and moisture content (%) of the fish after being salted. Each mean was determined from the results of 3 samples. Standard deviations given in parentheses.

Fish batch		Brine concentration (%)			
	0	5	10	20	30
1. Moisture	83.8(0)	84.9(0.1)	82.7(0.9)	77.4(0.2)	73.4(0.5)
Salt	0.7(0)	13.4(0.4)	28.9(1.7)	39.5(2.1)	47.2(1.2)
2. Moisture	86.2(4.8)	85.6(0.3)	82.2(0.3)	77.6(0.9)	71.6(0.4)
Salt	1.1(0.4)	22.6(0.2)	33.8(0.7)	50.0(0)	53.5(0)
3. Moisture	81.7(0.4)	84.3(0.3)	82.0(0.3)	79.2(0.3)	74.6(0.5)
Salt	1.3(0.3)	14.4(0.4)	21.0(0.5)	33.2(0.6)	40.0(1.4)
4. Moisture	-	83.1(0.2)	81.7(0.5)	78.4(0.6)	77.4(0.8)
Salt	-	10.1(1.2)	14.6(5.3)	25.7(4.6)	30.0(0.6)

8.3 Maintenance of *C. megacephala* cultures.

The cultures were established from *C. megacephala* pupae, imported from Indonesia and were maintained at temperatures of 28 - 30°C, in cages constructed of an aluminium frame, supporting a fine nylon mesh cover. The dimensions of the cages were 74 cm x 51 cm x 35 cm.

Temperature was controlled by an overhead heat lamp, activated by a thermostat, situated inside the cage. The heat lamp provided the only source of illumination during the trials.

The adult blowflies were reared from larvae that were fed on unsalted, fresh cod. After pupation, the newly emerged adults were fed daily on fresh, bloody, lamb's liver. Sucrose and water were made constantly available to the flies.

8.4 Effect of salt concentration of oviposition medium on oviposition by *C. megacephala.*

Materials and methods

This investigation comprised 5 series´ of trials, designed to test the hypothesis that, irrespective of salt concentration, *C. megacephala*, when presented with fish of a range of salt concentrations, would preferentially oviposit on the fish with the lowest salt concentration, but that in the absence of choice, oviposition would still occur on heavily salted fish.

Each cage population was maintained for 3 - 4 weeks before the trials commenced, to allow adequate time for the ovaries of the female, adult flies to mature. The flies were deprived of liver for 1 week before each series of trials commenced,to prevent premature oviposition and reduce the incidence of "spent" female flies.

Each series consisted of 4 trials, carried out on consecutive days. For the first trial of each series, the flies were presented with fish of 4 salt concentrations. In each succeeding trial, the choice of salt concentrations was progressively reduced, by omitting the fish which had the lowest salt concentration in the previous trial. Hence, for the last trial of each series, the flies were presented only with fish of the highest salt concentration. The fish, presented to the flies, had previously been cut into 16 cm^2 blocks and each fish sample was placed in an open, plastic dish. Fish of each salt concentration was presented in triplicate and the dishes were placed in a fully random order on the floor of the cage. After 1 hour, the fish was removed from the cage and the number of eggs present on each fish sample was counted, with the aid of a stereomicroscope.

The approximate numbers of flies present in the cage populations used for the trials were as follows: Series A and B: - 2,000, Series C, D and E: - 1,500.

Results and discussion

Table 53 shows that, with only one exception (Trial E.1), *C. megacephala* preferentially oviposited on the fish present with the lowest salt concentration. Oviposition was not, however, completely restricted to the fish presented with the lowest salt concentration, suggesting that other factors may also influence choice of oviposition site. When choice of fish of different salt concentrations was restricted or absent, the flies readily oviposited on fish with relatively high salt concentrations of 30 to 40% (dwb). These salt levels exceed those usually found in traditionally processed fish in South East Asia (Hanson and McGuire, 1982). Salt concentrations, particularly in salted, dried fish prepared from large species, which are most susceptible to blowfly infestation, are usually less than 30% (dwb). It can also be seen from the very large standard deviations of the mean numbers of eggs present on the fish, that oviposition was not spread evenly between the 3 replicates of any given salt concentration. In general, once one fly had started ovipositing on a particular piece of fish, then other female flies would quickly join it and commence oviposition. This group oviposition would result in a large mass of eggs being deposited on a single piece of fish, whereas the other replicates of the same salt concentration would remain largely free of eggs.

The results obtained during Trial E.1, were certainly anomolous, as no mistakes were made when transferring the fish to the dishes, or in identifying the dishes after the trial. Such an anomolous result could be produced by the group oviposition effect noted above. That is, if a fly started ovipositing on fish of a relatively high salt concentration, the resulting attraction to other flies could outweigh any repellent effect of the high salt content.

Group oviposition occurred during all of the trials and has also been frequently observed on fish being salted during field trials conducted in Indonesia.

Table 53. The effect of salt concentration on oviposition by *C. megacephala*.

(a) Mean number of eggs oviposited on 3 replicates of each salt concentration during Series´ A and B.

Trial	14.4 Mean	14.4 s.d.	21.0 Mean	21.0 s.d.	33.2 Mean	33.2 s.d.	40.0 Mean	40.0 s.d.	% eggs on lowest salt
A. 1.	1700	1864	62	84	0	0	0	0	97
A. 2.			98	96	0	0	0	0	100
A. 3.					234	210	0	0	100
A. 4.							771	1335	-
B. 1.	6512	2463	0	0	0	0	0	0	100
B. 2.			1236	2141	0	0	0	0	100
B. 3.					1025	1579	0	0	100
B. 4.							0	0	-

(b) Mean number of eggs oviposited on 3 replicates of each salt concentration during Series´ C, D, and E.

Trial	10.1 Mean	10.1 s.d.	14.6 Mean	14.6 s.d.	25.7 Mean	25.7 s.d.	30.0 Mean	30.0 s.d.	% eggs on lowest salt
C. 1.	734	1272	0	0	0	0	0	0	100
C. 2.			923	1180	0	0	0	0	100
C. 3.					3479	6026	0	0	100
C. 4.							1199	1267	-
D. 1.	5737	5695	365	633	0	0	0	0	94
D. 2.			768	940	0	0	0	0	100
D. 3.					233	404	0	0	100
D. 4.							1219	2111	-
E. 1.	580	606	52	90	679	1176	0	0	44
E. 2.			81	72	0	0	0	0	100
E. 3.					356	552	0	0	100
E. 4.							0	0	-

Salt concentration (% dwb)

The large standard deviation values reflect the variation in distribution resulting from the group oviposition effect.

The result that *C. megacephala* will readily oviposit on fish with relatively high salt concentrations differs from previously published opinion. Green (1967), Mills (1979), Proctor (1972), Kordyl (1976) and McLellen (1963), all reported that salted fish were unattractive to and Wood

blowflies. More recently, Walker (1985), found that salt appeared to inhibit egg laying on salted *Haplochromis* spp. and *Lethrinops* spp., during field trials carried out in Malawi. However, during Walker's trials, the blowflies found at the processing site, were presented with a choice of unsalted fish and fish of salt concentrations of up to approximately 9%. The laboratory trials, described here and observations by the author at several cured fish processing sites in South East Asia, indicate that, in the absence of such a choice between salted and unsalted fish (which is generally the case at commercial processing sites), salting the fish to the salt levels usually found in South East Asian cured fish, does not impart a repellent effect against ovipositing blowflies such as *C. megacephala* and *L. cuprina*.

8.5 Effect of salt concentration of feeding medium on growth and mortality of *C. megacephala* larvae and mortality of pupae.

Materials and methods

A series of 5 trials was carried out over a period of 6 weeks, using laboratory bred *C. megacephala* larvae. The larvae used in the trials were reared on minced cod, of a range of salt concentrations, which had been processed and analysed using the methods described above.

For each trial, the larvae were reared on fish of 5 salt concentrations. The salt concentrations of the fish used for Trials 1 and 2 were 0.7, 13.4, 28.9, 39.5 and 47.2% (dwb). Trials 3, 4 and 5 used fish at salt concentrations of 1.1, 22.6, 33.8, 50.0 and 53.5% (dwb). At the start of each trial, 1g samples of fish of each salt concentration were placed on small dishes in separate, transparent, plastic jars. Each salt concentration was replicated four times. Twenty five first instar larvae were then weighed and carefully transferred to each sample of fish. The jars were closed, using lids which contained 4 ventilation holes of 1 cm diameter. The holes were covered with fine, nylon mesh to prevent larvae from escaping. The fish was replaced daily, and from day 2 onwards, the quantity of fish used for each replicate, was increased to 12g. The larvae were incubated at 30°C throughout the trials. At daily intervals, counts were taken of all surviving larvae and the larvae from two replicates of each salt concentration were weighed on a top-pan balance, accurate to 0.0001g, until the onset of pupation. Before being weighed, the larvae were allowed to crawl on dry filter paper to remove excess moisture

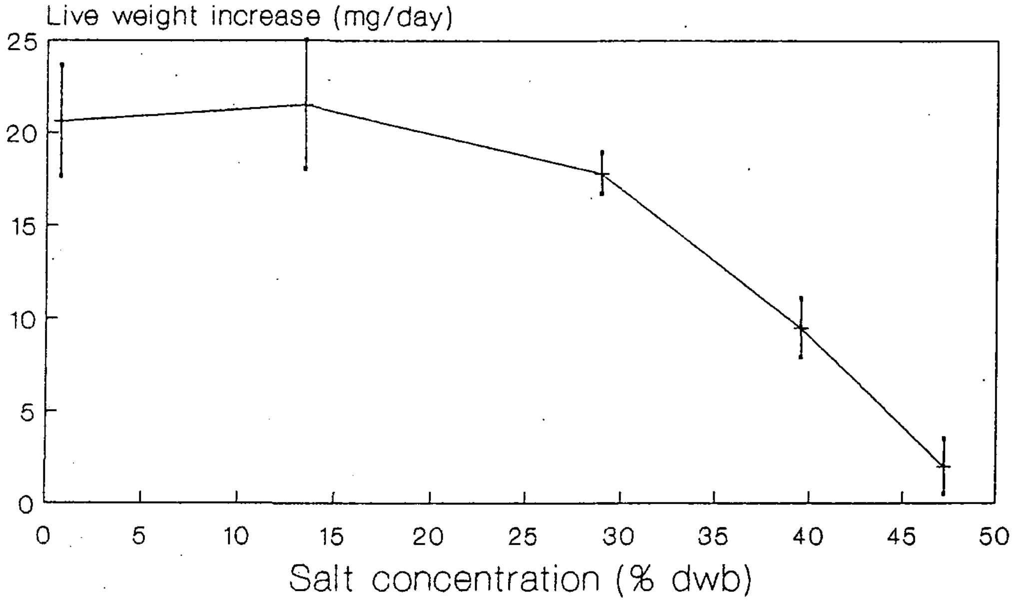
The pupae were tranferred to pupation medium consisting of peat and returned to the incubator. They were monitored at daily intervals for a period of 10 days and adult emergences were recorded. At the end of this period, unemerged pupae were dissected and the state of development of the contents was noted.

Results and discussion

Figures 44 and 45 show the effect of salt content on larval growth rate over the first 3 days of larval growth. Observations at fish processing sites in South East Asia have demonstrated that it is during this period that blowfly larvae cause most damage to the fish. Figure 44 indicates that during the first two trials, a salt content of 28.9% (dwb) results in a reduction in growth rate of 14% and that a salt content of 39.5% (dwb) results in a reduction in growth rate of 54%. Although Figure 44 shows a more rapid decrease in growth rate between salt contents of 28.9% and 39.5% than was obtained by lower salt levels, the difference was not statistically significant due to the small number of samples tested. The results of Trials 3-5 (Fig. 45) support the findings of the first 2 trials and also show that a salt content of 33.8% (dwb) gave a significant reduction in growth rate of 44% (Student t-test, $p<0.01$) and that a salt content of 50% gave a significant reduction in growth rate of 93% (Student t-test, $p<0.001$).

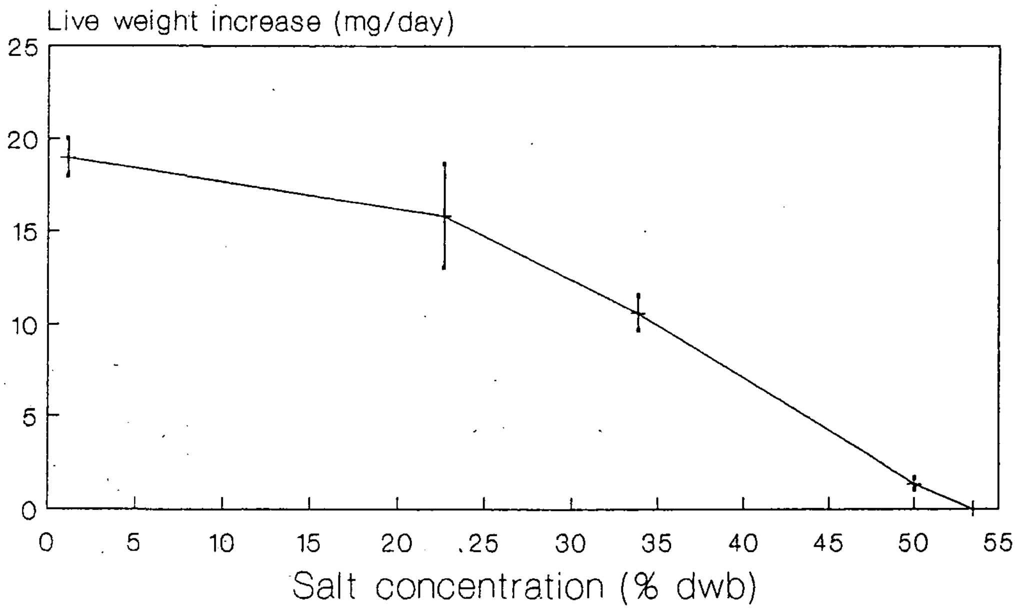
The larval growth curves given in Figures 46 and 47, show that the larvae reared on the unsalted fish grew most rapidly between the 1st and 2nd days of the larval duration and underwent an approximately 5-fold size increase within 24 hours. Salting the fish had the effect of decreasing the

Fig.44: Effect of salt content on larval growth rate of *C. megacephala* over first 3 days of growth at 30°C.



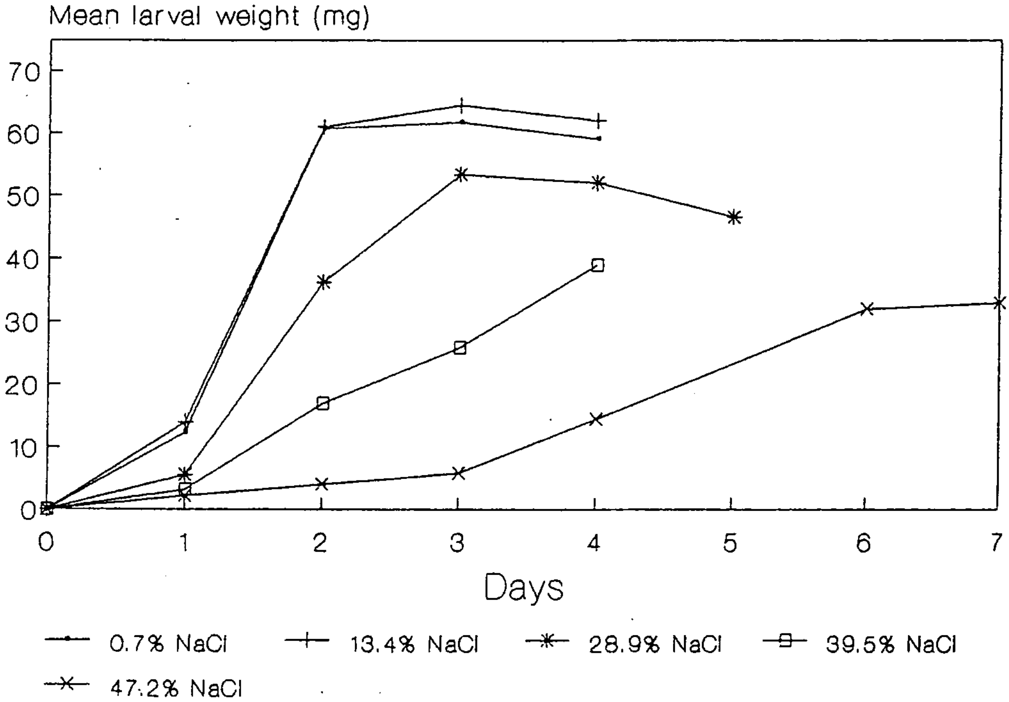
Trials 1 and 2 consolidated results.
Vertical intercepts=1 standard deviation (n=4)

Fig.45: Effect of salt content on larval growth rate of C. megacephala over first 3 days of growth at 30°C.



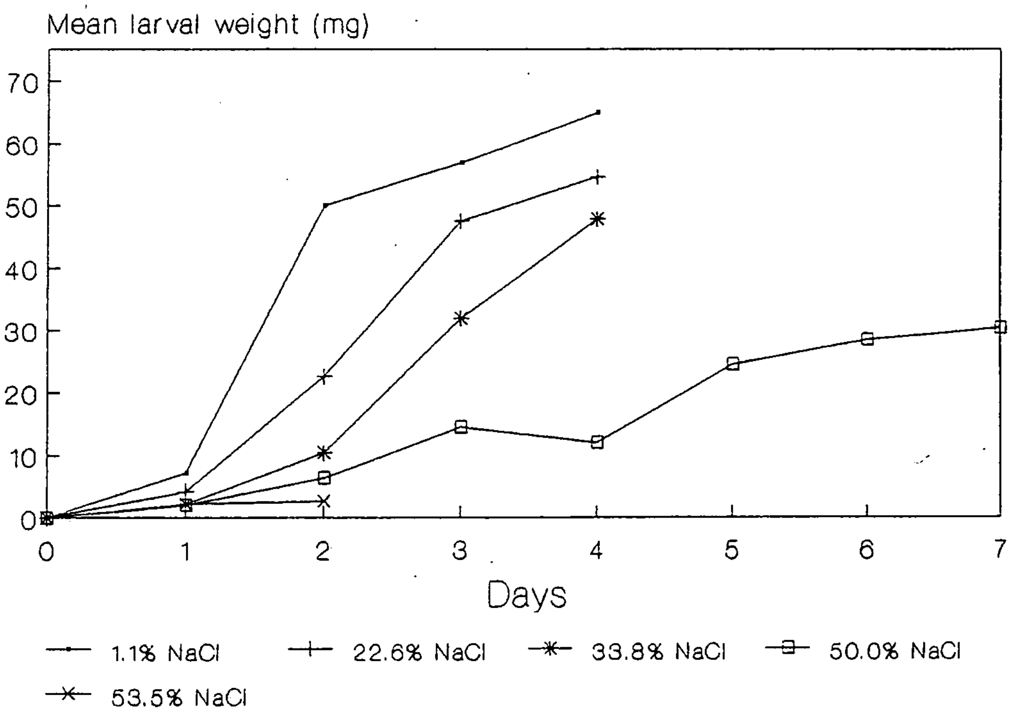
Trials 3-5 consolidated results
Vertical intercepts=1 standard deviation (n=6)

Fig.46: Effect of salt content on amount of larval growth over larval duration at 30°C.



Trials 1 and 2 consolidated results. (n=4)

Fig.47: Effect of salt content on amount of larval growth over larval duration at 30°C.



Trials 3-5 consolidated results (n=6)

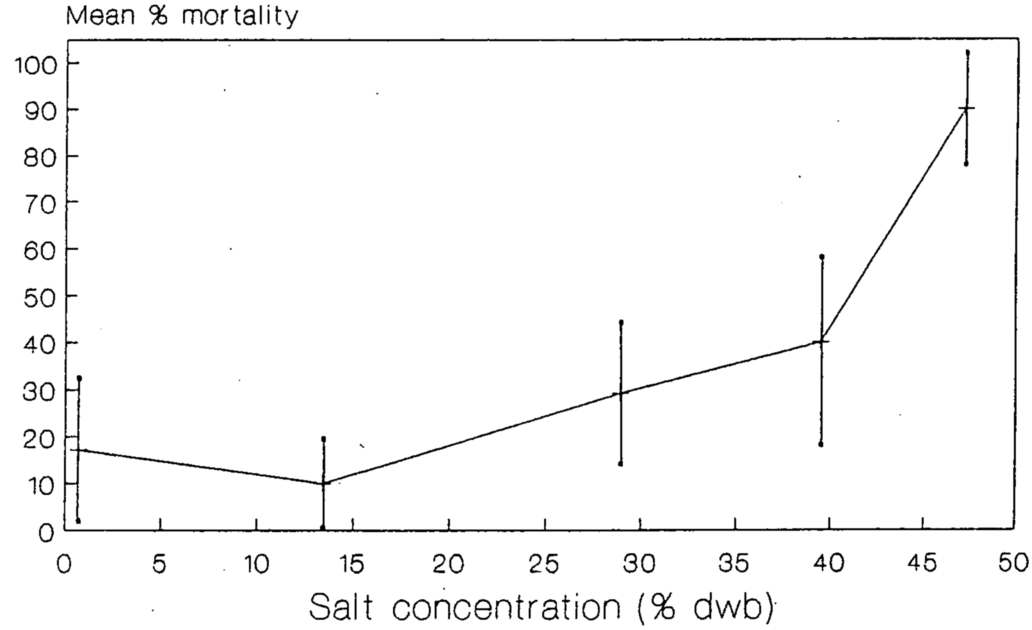
initial growth rate, but over the complete larval duration, salt contents of less than 33.8% (dwb) had little effect on the total amount of larval growth.

The larvae, reared on the unsalted fish, pupated on the 4th day after emergence. Figures 46 and 47 show that salt concentrations of 47.2% and 50%(dwb) caused a delay in the onset of pupation. The few larvae that survived these high salt concentrations, continued to grow at a slow rate for a further 3 days, before pupation commenced.

Figures 48 and 49 demonstrate the effect of salt content on larval mortality (% of larvae that died before pupation). The results show that a salt content of 47.2% (dwb) gave a significant increase in the mortality of *C. megacephala* larvae (Student t-test, $p<0.05$) and indicate that salt contents in excess of 39.5% are necessary to obtain high larval mortalities.

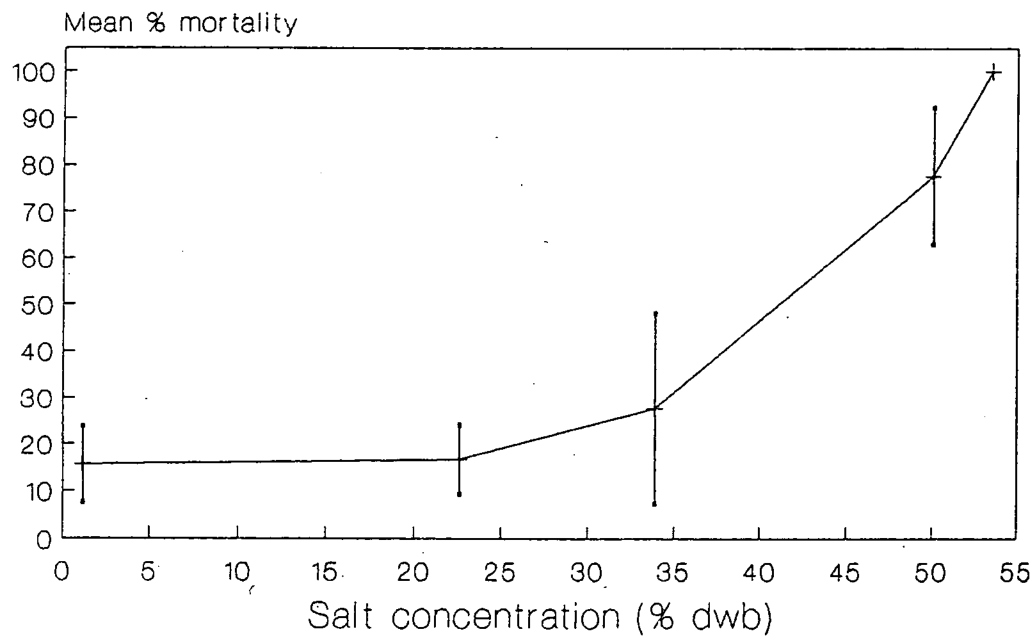
Figures 50 and 51 show the effects of salt content on the mortality of the pupae, which had developed from the larvae that survived the salting treatments. The mean pupal mortalities for Trials 1 and 2 fluctuated between 3.2 an 13.4% within the salt content range of 0.7 to 39.5% and an increase in salt content of the larval diet, over this range, had no effect on mortality. The mean mortality at a salt content of 47.2% increased to 54% but there was no significant difference between this value and those obtained at the lower salt contents. This was due to the large standard deviation that resulted from the small numbers of surviving larvae, that pupated at the high salt concentration. Similar results were obtained in Trials 3 to 5. Over the salt content range of 1.1 to 33.8%, the mean pupal mortalities fluctuated between 6.1 and 7.6% and a salt content of 50% gave a mean mortality of 46%. The large standard deviation, which resulted from the small sample sizes used at this salt content, again precluded demonstration of a significant difference. However, the important point to appreciate, is that some of the larvae reared on food of relatively high salt concentrations were capable of going on to pupate and successfully emerge as adults.

Fig.48: Effect of salt content on larval mortality of *C. megacephala* at 30°C.



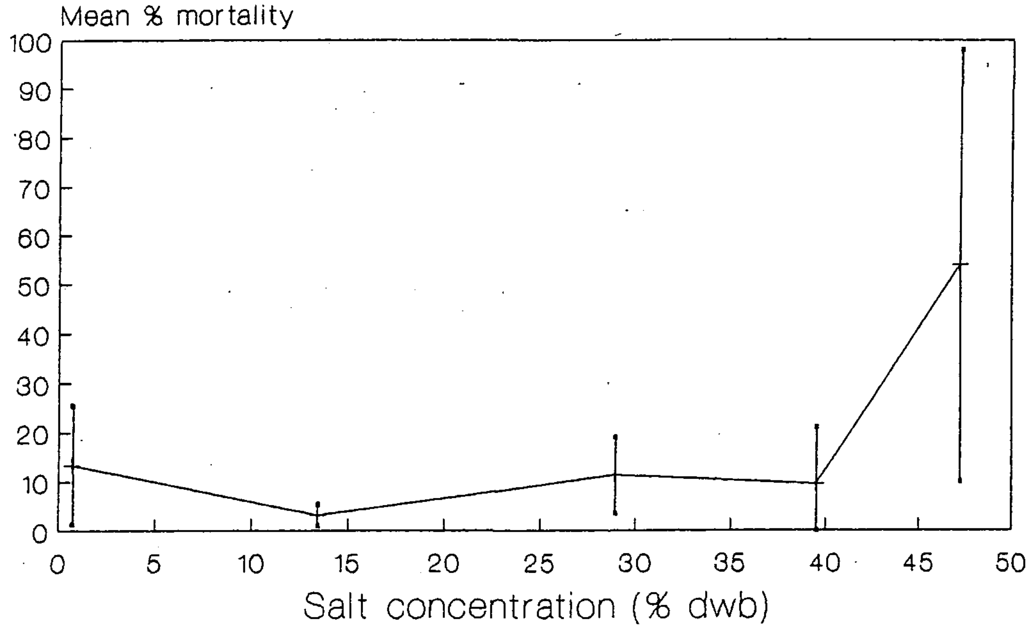
Trials 1 and 2 consolidated results.
Vertical intercepts=1 standard deviation (n= 8)

Fig.49: Effect of salt content on larval mortality of *C. megacephala* at 30°C.



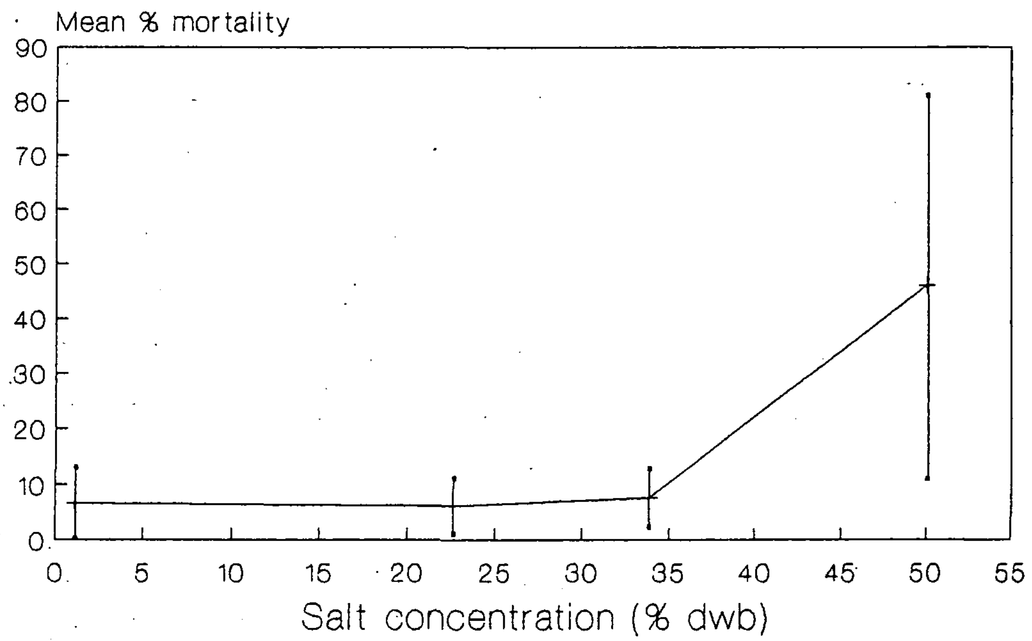
Trials 3 - 5 consolidated results.
Vertical intercepts=1 standard deviation (n=12)

Fig.50: Effect of salt content on pupal mortality of C. megacephala at 30°C.



Trials 1 and 2 consolidated results.
Vertical intercepts=1 standard deviation (n=8)

Fig.51: Effect of salt content on pupal mortality of *C. megacephala* at 30°C.



Trials 3-5 consolidated results.
Vertical intercepts=1 standard deviation (n=12)

Examination of 71 unemerged pupae, revealed that 69% of the pupae had died before developing recognisable structures and 31% had developed into recognisable insects, before death occurred.

The laboratory trials demonstrated that larvae and pupae of *C. megacephala* can tolerate higher salt concentrations than are normally present in traditionally processed, salted, dried fish. The results of these trials suggest that traditional fish processors would need to salt their fish to obtain salt contents of about 40% (dwb) in order to be reasonably certain of controlling infestation by *C. megacephala*. Although it would be possible to modify traditional salting techniques to obtain such high salt concentrations, observations in Indonesia suggest that the resulting, high salt content product would probably be unacceptable to both processors and consumers of salted, dried fish.

The finding that blowfly larvae can tolerate high salt concentrations again appears to contradict previously published reports. Kordyl (1976) observed that brining Lake Tanganyika sardines was sufficient to prevent the development of blowfly larvae and the Tropical Development and Research Institute (1984) reported that a salt concentration in excess of 8% gave larval control of at least 90% during trials conducted in Malawi.

Most of the published observations of blowfly attack on traditionally processed fish have been made in Africa, where fish are seldom salted before drying, under generally drier climates. Of the 2 species of blowflies, observed to infest salted, dried fish in South East Asia, only *L. cuprina* has been reported to infest cured fish in Africa (Walker and Donegan. 1984). Although *C. megacephala* has been introduced to the African continent (Prins, 1979), it has not, as yet, been reported to cause problems to African fish processors. The apparent discrepancy between observations of blowfly oviposition behaviour and larval salt tolerance in Africa and South East Asia, could be due to both *C. megacephala* and *L. cuprina* having evolved a tolerance to the relatively high salt concentrations found in South East Asian fish, which is usually salted before drying. In Africa, where fish is not usually salted before drying, tolerance to high salt concentrations may not yet have been selected for in the blowfly populations associated with the indigenous, traditional, fish processing sites. Should salting before drying be adopted as a method of controlling blowfly infestation in Africa, then the recent introduction of *C. megacephala* to that continent must present cause for serious concern. Further laboratory trials on blowfly species known to infest fish in Africa eg. *Chrysomya albiceps* (Wied.), *Chrysomya chloropyga putoria* (Wied.) and *Chrysomya regalis* (Desvoidy), should be carried out, to determine if salt tolerance is present in local blowfly species', before African processors are advised to adopt the relatively expensive and, probably locally unacceptable, technique of salting their fish before drying.

9. APPLICATION OF RESEARCH FINDINGS TO REDUCING POST-HARVEST LOSSES OF FISH IN SOUTH EAST ASIA AND POSSIBLE AREAS OF FUTURE RESEARCH.

9.1 Summary of research results and their application to reducing post-harvest losses of cured fish.

The field trials and experiments, detailed in the previous sections, have identified and investigated the nature of the major pest problems encountered during the production and distribution of traditionally processed fish in South East Asia. Methods of controlling these problems have been evaluated. The most important of the research findings can be listed as follows:

(i) Traditionally processed, in particular salted, dried fish, is an economically and nutritionally important commodity in South East Asia. Traditional processing is the most widespread method of preserving fish in the region and will continue to be important for the forseeable future. In the more remote areas, it is the only method available to the community. Cured fish assumes particular nutritional importance in inland, rural areas, where it forms a valuable animal protein supplement to the staple diet of rice. For example, in Thailand, large quantities of cured fish, processed in Samut Sakhon, are transported to the relatively impoverished north-east of the country, for consumption by both Thai people and large numbers of Cambodian refugees, housed in camps, along the Thai border.

As well as being an essential source of animal protein to the poorest section of the community, cured fish is also a popular commodity in its own right, and large quantities are processed and sold as high value products. The cured fish industry supports whole fishing communities and will continue to be significant in the economy until refrigeration and freezing facilities become more widespread. At present, such facilities are devoted to fish being processed for export, which has a far higher value than fish destined for domestic consumption.

(ii) Traditionally processed fish suffers serious damage and loss in quality during processing and storage, principally as a result of insect infestation. Microbial spoilage and rancidity are also significant in reducing quality and nutritional value. These losses are particularly significant in small scale processing operations, which lack basic facilities such as piped water and waste and sewage disposal. The relatively low value produce of such operations, is usually destined for the less affluent, who consequently consume fish that is of poor quality, relatively low nutritional value and may contain potentially harmful levels of histamine and lipid oxidation products, which accumulate in fish, during extended storage periods at ambient, tropical temperatures.

(iii) *C. megacephala* and *L. cuprina* are the most important causes of infestation and losses in salted, dried fish during processing. Of the 2 species, *C. megacephala* is of vastly greater importance than *L. cuprina* in South East Asia, and was frequently observed to be present in large numbers, at numerous processing sites visited in Indonesia and Thailand.

(iv) *Dermestes* spp. and *P. casei* are the most important causes of infestation and losses during storage and distribution. *P. casei* was most frequently observed to infest relatively moist, salted, dried fish in store, whereas the dermestid beetles tended to be more serious pests of very dry salted fish and smoked fish.

(v) Cured fish processors and retailers have responded to these problems by illegally applying toxic, household and agricultural insecticides to their fish. This widespread, uncontrolled practice, presents serious, potential short-term and long-term health hazards to both processors and consumers of salted-dried fish. The application of kerosene containing insecticides to the fish is particularly worrying.

(vi) The insects, responsible for infesting salted, dried fish in South East Asia, are tolerant of the relatively high salt concentrations found in the product. Salting the fish, fails to prevent infestation by *C. megacephala, L. cuprina, P. casei* and dermestid beetles.

(vii) During processing, the fish are particularly susceptible to blowfly infestation during the salting and early drying stages of the process. Observations at the processing site in Cirebon, revealed that the blowfly larvae, principally *C. megacephala*, in the salting tank, were tolerant of the high salt concentration of the pickle, produced during salting.

(viii) Laboratory investigations, demonstrated that the larvae of *C. megacephala* can tolerate far higher salt concentrations than are usually found in cured fish in South East Asia. High salt levels also failed to inhibit oviposition by *C. megacephala*. Although salting has been recommended as a method of preventing blowfly infestation of cured fish in Africa, it is clearly not applicable to the situation in South East Asia and should not be pursued as a loss reduction method. Salt tolerance of the other insect species, associated with dried fish, warrants further investigation.

(ix) Repairing and fitting the salting tanks with tightly fitting lids, effectively prevented blowfly infestation during the salting stage of processing. This simple and inexpensive technique impressed and was subsequently adopted by the processor in Cirebon. It´s introduction to other processors, could result in a significant reduction of infestation during processsing, particularly during the dry season, when infestation during drying, assumes less importance. The drying screen design, evaluated during trials in Cirebon, was effective in reducing infestation during drying, but there were practical difficulties associated with it´s use, that made it unacceptable to the processor.

(x) The pyrethroid insecticide Fastac (alphacypermethrin) was very effective in controlling insect infestation during processing and storage, when applied at concentrations as low as 0.003%. Not only was it larvicidal, it also had a marked repellent effect against blowflies, when applied at a concentration of 0.001%. This repellent action is beneficial, in that it will probably reduce microbial contamination of the fish during processing, which in turn should help extend shelf life and reduce the chances of pathogen transmission.

Similar results were obtained with the pyrethroid insecticide Cislin (deltamethrin) and support the view that pyrethroid insecticides should be further evaluated as protectants of cured fish.

(xi) Treatment with the FAO/WHO approved insecticide, pirimiphos-methyl, at a dip concentration of 0.03% a.i., effectively protected cured fish against insect infestation during processing and storage, and represents the only currently available, safe, short-term remedy, that is likely to be adopted by fish processors.

(xii) Observations at the processing site and examination of fly samples, collected from the fish, demonstrated that *A. thalassinus* becomes highly attractive to ovipositing *C. megacephala* during salting, probably as a result of chemical changes to the fish. The nature of this attraction warrants further investigation.

(xiii) Laboratory investigations demonstrated that the presence of *C. megacephala* eggs on fish, stimulated further oviposition by *C. megacephala*. Experiments indicated that this stimulus was chemical, as opposed to visual. Isolation and identification of the chemicals, responsible for this effect, could lead to the development of effective blowfly lures.

(xiv) Blowfly activity, at the processing site in Cirebon, was observed to fluctuate wildly from day to day, probably as a consequence of human activity in the area, rather than climatic effects. This suggests, that taking steps to change, for example, current refuse disposal practices, could contribute towards a reduction in the blowfly population.

The primary aim of research, in the area of overseas development, is that its findings should ultimately assist in the development of the target countries. In the case of the research detailed in this thesis, the findings should assist development, by reducing losses of a nutritionally and economically important commodity, consequently increasing the supply of animal protein available to the population and result in a safer food product, which would benefit the health of the consumer.

Before this can happen, however, it is essential that the relevant sectors in the target country, are made aware of the research findings and are given practical assistance with applying those findings, to solving their problems. Unless this follow-up work is undertaken, the research findings may remain of academic interest only and fail to assist in development.

In order to assist in extending the research findings to the fish processing and distribution sector in Indonesia, a training project, funded by the Overseas Development Administration, was started in 1986.

One of the major aims of this project, is to provide staff of the provincial fish quality control laboratories in Indonesia, with training in loss assessment and safe, loss reduction techniques. To date, training courses have been conducted in East Java, Central Java, West Kalimantan, North Sumatra, South Sumatra and Lampung provinces. Staff from Riau province attended the course in Lampung. Similar training programmes are to be conducted for fisheries department staff of South Kalimantan, South Sulawesi and West Java (including metro Jakarta) provinces. By the time the project finishes in April/May 1989, all of the major salted-dried fish producing areas will have been covered.

Each training course consists of lectures and seminars on loss assessment and reduction, together with practical training in survey and field trial techniques. The course commences with a general lecture on the nature of post-harvest losses and methods of reducing losses. This session, which usually occupies 2-3 hours, is attended by most of the staff of the provincial fisheries departments and interested outside parties eg. staff from local universities, department of health, police etc. Seminars, detailing loss assessment and reduction methods are then held for those staff who will be directly involved with future training of fish processors. Training in survey techniques, is given during visits to fish processors, wholesalers and retailers within the province. This part of the programme can last from 1 to 4 days, depending on the location of the fish processing communities. During this survey, a suitable fish processor who will cooperate with field trials, is identified.

The field trials, evaluating the efficacy of pirimiphos-methyl in reducing blowfly infestation and losses of cured fish, usually occupy about 2 weeks. After an initial trial, demonstrating the methodology of insecticide application and evaluation, the fisheries department staff, conduct under supervision, a similar trial themselves. A final trial, investigating the effect of a variable eg. dipping time, on infestation is also conducted by the participants in the course. This assists consolidation of the techniques already covered and gives experience of how to apply their training to answering questions. During the course of the trials, fish samples are collected for salt and moisture analyses by the fisheries department staff. The remaining fish, processed during the trials, is put into store, usually for a period of about 3 months.

After receiving advice on how to analyse and present the field trial data, the staff who participated in the trials, present their findings at a seminar for the rest of the provincial fisheries department staff.

The fish left in store, is assessed for insect infestation and damage during a short follow-up visit and a final seminar is held for all interested parties.

A major emphasis of each course is to provide practical training in the safe use and evaluation of pirimiphos-methyl. Pirimiphos-methyl has recently received provisional registration, for use on fish, by the Indonesian authorities and is shortly to be marketed, under the brand name Minawet 250 EC, by P.T. I.C.I. Pesticida. This company has considerable experience of supplying pesticides to rice farmers etc., and will, in collaboration with the Department of Fisheries, design can labels that will carry clear, simple instructions on how to safely apply Minawet to fish.

The evaluation trials, carried out during each training course, have consistently demonstrated that pirimiphos-methyl, applied as a 0.03% dip, protects salted-dried fish against blowfly and beetle infestation (Esser *et al.*, 1988b).

It was originally intended that fish processors should take part in the training courses. However, the protracted nature of the Minawet registration process, meant that Minawet could not be legally recommended to fish processors, until it was finally registered at the end of February, 1988. Consequently, the training courses have been restricted to the Fisheries Department staff, with a view to them extending training to the processors, after Minawet had been registered.

If Minawet is to be adopted by the majority of fish processors, it is vital that it is demonstrated to them, that Minawet is effective in controlling insect infestation and that its use makes economic sense. It is also essential that the processors receive training in the safe use of Minawet and that its continued use be monitored, to ensure that residues remain within acceptable limits.

It now seems, that as a result of severe cuts in departmental budgets in Indonesia, this extension will not take place in the absence of further, outside assistance. In order to ensure that the benefits of the research, described here, are realised by fish processors and consumers, further project proposals, aimed at assisting with such an extension programme, are being submitted to the ODA. It is anticipated that once this extension has been carried out, the project work on Minawet will have reached its logical conclusion and that future training and monitoring of processors will conducted by the insecticide manufacturer and the Department of Fisheries.

9.2 Areas of future research

Insect infestation of traditionally processed fish will continue to be a serious problem in the Tropics until safe, effective and economic countermeasures are successfully introduced.

Although pirimiphos-methyl appears to fulfill all of the requirements of a successful remedy, the fact remains that it is still a poison and therefore does not represent an ideal answer to the problem of insect infestation of cured fish. While its current use can be justified on the grounds that it is far safer than some of the insecticides currently being applied to cured fish, it would be unreasonable and debatably amoral, to regard it as a long-term remedy, while failing to continue research into alternative countermeasures, that do not require the application of insecticides.

Screening is a possible remedy that warrants further investigation. Guarding the salting tank with a closely fitting lid, prevented infestation during salting and was a technique readily adopted by the processor in Cirebon (a recent visit to Cirebon revealed that the processor was still using the lid, 4 years later). While the design of the screen, used during the drying trials, presented practical problems and was not adopted by the processor in Cirebon, alternative designs might overcome the problems and should be evaluated. Permanent, walk-in designs may be suitable for premises with large drying areas and although initial, capital outlay would be high, they should be inexpensive to maintain and, in the long term, would probably pay for themselves. For continued protection during storage, suitable, cost effective packaging techniques require further investigation.

The role of volatile chemicals, released by fish during processing, in attracting gravid blowflies is another area that requires further research. Field observations, described in this thesis, suggest that, at certain stages during processing, fish exert a powerful, olfactory stimulus, which results in blowflies being attracted to and subsequently ovipositing on the fish. The isolation and identification of these volatiles could lead to analogues being developed and incorporated into fly traps. In addition, the pheromones that elicit group oviposition, could be further investigated with the same end in mind. Pheromone baited traps have proved successful in controlling other insect pests (Hodges, 1984) and could be integrated with other methods to control infestation.

Further information on naturally occurring insect repellents/insecticides is also required. Asastyasih and Madden (1986), investigated the role of plant products and extracts in preventing blowfly infestation of salted-dried fish and observed that white pepper, garlic, star fruit extract and acetic acid had a repellent effect on *M. domestica* and *C. megacephala*. In the Gambia, the use of lime juice and ground chillies, to control blowfly infestation of sundrying fish, has been observed by Walker and Evans (1984). Pepper is used by some fish processors in Burma, Malaysia and Indonesia, to control blowfly infestation and its use has also been reported in India by Pillai (1957) and Nigeria by Rollings and Hayward (1963). Don-Pedro (1985) found that powdered sun-dried citrus peel could reduce infestation of dried *Clarius* sp. by *D. maculatus*. It must be borne in mind, however, that if any of the natural repellents/insecticides, sometimes used by processors, are effective and economic, then one would expect their use to be more widespread than is apparent. However, it would be worthwhile to identify the active ingredients of these plant products, with a view to developing more efficient repellents.

Further investigation of the biological basis of the high salt tolerance shown by *C. megacephala* larvae should provide useful information on insect salt regulation mechanisms. In addition, the nature of the apparent sensitivity to salt of African blowfly species, needs to be ascertained.

Large blowfly populations result from poor standards of hygiene and sanitation, which themselves are products of poverty and ignorance. Trial, anti-blowfly campaigns should be carried out to see if it is practicable to reduce *C.*

megacephala populations, particularly in the densely populated, urban areas, where fish processing premises are often located.

To be effective, the campaign would require active collaboration between the various departments responsible for public health, sanitation and education, if it is to bring about a sustained improvement in standards of hygiene.

Apart from benefiting fish processors themselves, such a campaign would be worthwhile from a general public health point of view, in contributing to an increase in the standard of health of the community and consequently the standard of living and quality of life of the less affluent in Indonesia.

Blowflies are notorious carriers of disease, particularly the pathogens that cause common diseases in developing countries eg. diarrhoea, dysentery and cholera. Food poisoning microorganisms such as *Staphylococcus aureus* and faecal indicators, belonging to the Enterobacteriaceae and Vibrionaceae, have been isolated from *C. megacephala*, collected at fish processing sites in Jakarta (Anggawati *et al.*, 1986). Blowflies are also thought to have a role in the transmission of tape worm eggs, which they pick up when feeding on human and animal faeces (Lawson and Gemmell, 1985).

Identification of the chemicals that attract blowflies could lead to synthetic analogues being used in a blowfly trapping programme, as part of the wider campaign to reduce the blowfly population.

Such a campaign would be expensive and complicated to administer, but the potential benefits, particularly in the general area of improved public health, should more than justify the effort and expense.

It seems to be the case, that research produces more questions than answers, and the work described in this thesis is no exception. Post-harvest losses in cured fish is unfortunately, for various reasons, a relatively neglected area of research in the Tropics. Further awareness of the problem and of the many questions that still need to be answered, will hopefully, help to correct this state of affairs and stimulate more relevant research, that contributes towards the development of countries in the tropics.

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Appendix 1.

Distribution of principal insect pests at cured fish processing and retail locations visited in Thailand and Indonesia.

	Location	Insect species *Chrysomya megacephala*	 *Lucilia cuprina*	 *Piophila casei*	 *Dermestes* spp.*
T					
H	Samut Sakhon	$+^{52}$	-	-	$+^{265}$
A	Samut Prakhan	$+^{9}$	+	-	-
I	Songkhla	+	+	-	-
L	Bangkok	-	-	-	+
A					
N					
D					
I	Jakarta	$+^{41}$	$+^{2}$	-	+
N	Eretan	+	-	-	-
D	Cirebon	$+^{1870}$	$+^{988}$	+	$+^{15}$
O	Bandung	-	-	-	+
N	Bogor	-	-	-	+
E	Cilacap	+	+	+	+
S	Brondong	+	+	+	+
I	Surabaya	-	-	-	+
A	Muncar	$+^{6}$	-	-	+
	Medan	+	-	-	$+^{26}$
	Tanjung Balai	+	-	+	+
	Palembang	+	+	+	+
	Sungsang	+	+	-	-
	Padang	-	-	-	+
	Pontianak	$+^{35}$	+	-	+
	Banjarmasin	-	-	-	+
	Bangka Island	$+^{5}$	-	-	+
	Mataram	$+^{10}$	-	-	+
	Lombok	+	-	-	-

+ = Observed at field location
- = Not observed at field location
Superscript no. = number of insect samples returned to U.K.
* *Dermestes* complex consisted of *D. maculatus, D. carnivorus, D. ater* and *D. frischii*.

Appendix 2

Counts of *C. megacephala* and *L. cuprina* recorded on drying *Arius thalassinus* during 1984.

Counts		
C. megacephala	*L. cuprina*	Ratios
452	26	17:1
222	2	20:1
476	7	68:1
111	10	11:1
476	11	43:1
1285	30	43:1
906	44	21:1
190	18	11:1
1089	29	38:1
706	15	47:1
2936	18	163:1

Appendix 3

Insecticide residues (mg/kg) at the end of drying of *A. thalassinus*.

Treatment	Size category	Number of fish analysed	Mean residue ± standard deviation
control	Small Medium Large	4 4 4	<0.05 <0.05 <0.05
0.03% pirimiphos-methyl	Small Medium Large Mean	4 4 4 -	3.6 ± 2.0 5.2 ± 2.7 4.9 ± 1.9 4.6 ± 2.2
0.06% pirimiphos-methyl	Small Medium Large Mean	4 4 4 -	10.9 ± 1.1 9.3 ± 1.4 9.2 ± 2.3 9.8 ± 1.7
control	-	3	<0.1
0.015% pirimiphos-methyl	-	10	2.8 ± 0.8
0.03% pirimiphos-methyl	-	3	4.6 ± 1.5
0.015% pirimiphos-methyl	-	3	3.3 ± 1.4
0.03% pirimiphos-methyl	-	3	4.9 ± 0.9

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