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Distribution of DDT residues in fish from the Songkhla Lake, Thailand

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"Capsule": *Low DDT concentrations in fish of a tropical lake may reflect rapid biotic dilution and degradation.*

Abstract

Since the early 1950s DDT has been extensively used in Thailand as a malaria repellent and as an agricultural pesticide, but was finally banned in 1994. In this study concentrations of *p,p′*-DDT, *p,p′*-DDE, and *p,p′*-DDD in 113 fish of four species (*Scatophagus argus, Protosus canius, Channa striata* and *Zonichthys nigrofasciata*) are reported from the large, brackish Songkhla Lake and the Gulf of Thailand. The mean ΣDDT concentrations at different locations in the analysed fish species ranged from 33 to 170 ng/g lipid wt. (0.086–7.7 ng/g fresh wt.). This is well below the recommended maximum residue levels in aquatic animals used for human consumption (5000 ng/g fresh wt.) in Thailand. The comparatively low residue levels could be due to the high temperature and solar radiation in the region, which may result in a high volatilising and degradation rate of DDT. Also, the high productivity of the lake could result in a dilution effect, when DDT is distributed in a large amount of organic matter, followed by a high biological degradation of the substance.

Keywords: Southeast Asia; Organochlorine; Fish; Tropical ecosystem

1. Introduction

Since the early 1950s DDT has been extensively used in Thailand as malaria repellent and as an agricultural pesticide. During the last decade the use has been successively limited and was finally banned for all applications in 1994 (UNEP, 1997). In spite of this, persistent organochlorine pesticides are still circulating in Thai ecosystems that are simultaneously being exposed to increasing pollution loads resulting from rapid population growth, urbanisation and agroindustrial development (Hungspreugs et al., 1989).

This is the case in the region surrounding the Songkhla Lake, a lake that is regarded as one of the most important natural resources in southern Thailand, with fishing and aquaculture as the main economic activities. The lake provides food, water for irrigation and domestic use, means of transportation and recreation to the people of its surroundings (Penpolcharoen, 1994). The biodiversity is remarkably high, with a great variety of fish, molluscs, crustaceans, insects, amphibians, reptiles and birds. Approximately 40 of the fish species in the lake are used for human consumption (Scott, 1989).

Agricultural land covers about half of the Songkhla Lake drainage basin. The lowlands are suitable for rice cultivation whereas rubber and fruit plantations dominate the upland areas. Since the 1970s the extensive agricultural activities of the area have brought about increasing use of agricultural chemicals, mainly fertilisers and pesticides (VKI, 1997). In spite of this, it is estimated that 15–30% of the annual crop production is lost to pest, disease and weed competition (Pipithsangchan et al., 1997).

Although DDT for agriculture use was banned in 1983, residue levels can still be recorded in various crops indicating present usage on agricultural lands around the lake (Pipithsangchan et al., 1997). Some research has been conducted on the organochlorine pesticide

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contamination of the abiotic environment in Songkhla Lake (Kanatharana et al., 1994; VKI, 1997). However, no studies have investigated the distribution of organochlorine pesticide residues in aquatic organisms and a base-line study of pesticide contamination in fish is needed since fish is considered to be an important human food source in the area (VKI, 1997). This study provide more information on the residues of DDT in tropical aquatic environments. Models of the redistribution of contaminants such as DDT from tropical areas into temperate areas have been reported (Wania and Mackay, 1993). However, the base of such models is the understanding of the fate of compounds such as DDT in tropical environments. It is, therefore, important to provide more information on background contaminant concentrations in tropical environments.

This study quantifies 1,1-dichloro-2,2-bis(4-chlorophenyl)ethene (p,p'-DDE), 1,1-dichloro-2,2-bis(4-chlorophenyl)ethane (p,p'-DDD) and 1,1,1-trichloro-2,2-bis(4-chlorophenyl)ethane (p,p'-DDT) residues in four species of fish from three different trophic levels obtained from a number of locations throughout the Songkhla Lake. The concentrations are related to ecosystem structure, and compared to level in fish from temperate brackish water regions and to health limits set up by the Thai Ministry of Public Health. Reference samples were collected in the Gulf of Thailand and South Chinese Sea.

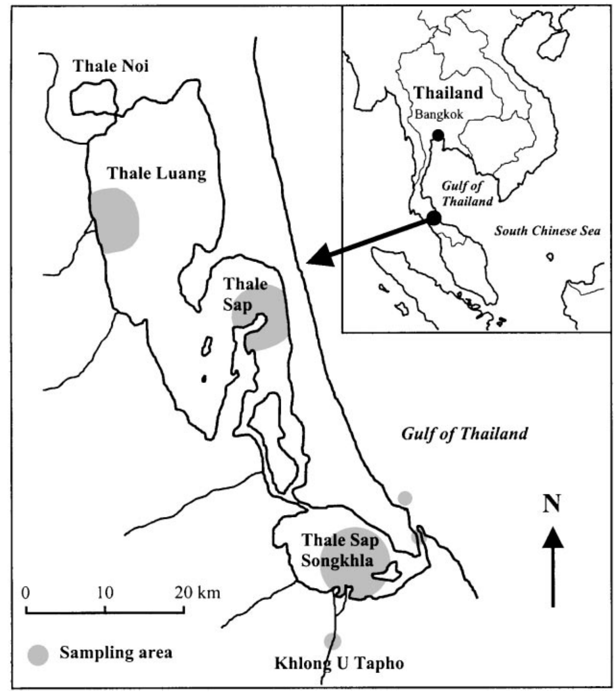

Fig. 1. Sampling areas in the Songkhla Lake basin and the Gulf of Thailand, Thailand.

2. Materials and methods

2.1. Study area

The Songkhla Lake, with an area of 1040 km^2, is the largest lake in Thailand and one of the largest lakes in southeast Asia (Penpolcharoen, 1994). It is located on the southern peninsular of Thailand at latitude 7°43′ to 8°00′ N and longitude 100°05′ to 100°15′ E (Scott, 1989) (Fig. 1).

The Songkhla Lake drainage basin has an area of about 8020 km^2 and consists mainly of farmland, forests and swamps. In the west it is bordered by the Khao Banthad mountain range, in the east by the Gulf of Thailand and in the south it extends across the plains towards the Malaysian border. There are no major rivers in the area, but the lake is fed by many small streams and man-made canals, as well as by run-off from the steep, mainly forested mountains in the west (VKI, 1997).

The lake is 90 km long and 20 km wide and very shallow, with an average depth of 1–1.5 m (Scott, 1989). It can be subdivided into four distinct parts forming three shallow basins connected to each other and to the sea by channels. There is a steep salinity gradient from the northern to the southern part of the lake, changing from fresh to brackish water (Scott, 1989). The northern part of the lake is called Thale Luang (490 km^2, 3‰ salinity) and the middle part Thale Sap (380 km^2, 11‰). The southernmost Thale Sap Songkhla (190 km^2, 20‰) is connected to Thale Sap by a short 8-m-deep channel and opens towards the Gulf of Thailand near the town of Songkhla (VKI, 1997).

2.2. Sampling

The sample points were located along a gradient throughout the lake, stretching from the fresh waters of Thale Luang, through Thale Sap to the almost marine Thale Sap Songkhla (Fig. 1). Fish samples were obtained from fishermen at local fish markets during the dry season of March and April 1997. The fish were not gutted and in most cases still alive. The length of the fish was measured from the front-tip of the mouth to the beginning of the caudal fin with a precision of 0.1 cm and the fish weight was determined with a precision of 1 g.

Four different fish species were collected, one herbivorous (*Scatophagus argus*), one omnivorous benthic dwelling (*Protosus canius*) and two carnivorous species (*Channa striata* and *Zonichthys nigrofasciata*). Two carnivorous fish species were used since the freshwater *Channa striata* could not be found in Thale Sap Songkhla. *Channa striata* was collected from the freshwater Khlong U Tapho, a small canal 4 km south of Thale Sap Songkhla as well as from the sampling areas in Thale Luang and Thale Sap. The additional marine carnivore, *Z. nigrofasciata,* was collected from the opening between Thale Sap Songkhla and the Gulf of

Thailand. It can, however, not be excluded that the *Z. nigrofasciata* sampled at the opening actually dwell most of their time in the Gulf of Thailand. As reference samples, *S. argus* and *Z. nigrofasciata* were also collected from the Gulf of Thailand and the South Chinese Sea, respectively. The sampled fish species were recommended by local fish biologists, and are of commercial interest and commonly consumed by the human population around the lake.

2.3. Chemicals

All solvents were of highest purity commercially available. Acetone and *n*-hexane were purchased from Fisher Scientific Ltd. (Loughborough, UK) and methyl *tert*-butyl ether (MTBE) from Rathburn Chemicals Ltd. (Walkerburn, Scotland, UK). Sulphuric acid (p.a.) from Merck (Darmstadt, Germany) was used. Silica gel 60–200 Mesh (Macherey-Nagel, Düren, Germany) was activated at 280°C for 24 h before being used.

2,2′,5,5,6′-Tetrachlorobiphenyl was used as surrogate standard. *p,p*′-DDE and *p,p*′-DDD were purchased from Dr. Ehrenstorfer GmbH (Augsburg, Germany) and *p,p*′-DDT from Aldrich Chemical Co. (Milwaukee, WI, USA).

2.4. Analysis

The chemical analyses were performed according to Jensen et al. (1983). A known amount of dorsolateral fish muscle was homogenised in a 2:1 mixture of acetone–*n*-hexane with an IKA T25 homogeniser (Labassco AB, Partille, Sweden). The lipid fraction was extracted with a 9:1 mixture of *n*-hexane–MTBE and separated from an added aqueous phase of sodium chloride (0.9%) in orthophosphoric acid (0.1 M). The organic solvents were allowed to evaporate and the lipid content was determined gravimetrically. The surrogate standard was added to the samples and the lipids were dissolved in *n*-hexane and partitioned with concentrated sulphuric acid in order to remove the bulk lipids. The samples were then transferred to a column containing 0.5 g of silica gel–concentrated sulphuric acid (2:1 on a weight basis) prepared in a Pasteur pipette for additional clean up. The samples were washed out with 8 ml of *n*-hexane.

One solvent sample was analysed in parallel with every 10 samples. In cases where blanks contained traces of analytes these amounts were subtracted from the samples. The recovery of the surrogate standard was not determined in this study, but has been reported for a similar method using one additional silica gel–sulphuric acid column to be 83% with a standard deviation of 7% ($n=130$) (Olsson, 1999).

The purified samples were analysed using a Varian 3400 gas chromatograph (GC) equipped with a Varian 8200 autosampler and an electron capture detector. The separation was performed on a J&W Scientific BD-5 capillary column (30 m, 0.25 mm i.d. and 0.25 μm film thickness), with hydrogen as carrier gas (1 ml/min) and nitrogen as make-up gas (30 ml/min). The injection volume was 1.5 μl.

The temperatures of the injector and detector were 250 and 360°C, respectively. The GC oven temperature was initially set at 80°C for 2 min, then raised by 10°C/min to 300°C and maintained for 6 min.

2.5. Statistics

Differences within species among sampling locations were investigated using analysis of variance (ANOVA) one-factor analysis. Differences were investigated using log-transformed concentrations, whereas length and weight data were untransformed. The Tukey's test for unequal sample sizes was used for mean separations (Spjotvoll and Stoline, 1973). Correlations between different parameters were investigated using linear regression on log-transformed data.

3. Results

Weight and length data of the analysed fish are given in Table 1. The sampled material of *Protosus canius* and

Table 1
Biological data (fish weight and fish length) for the fish species sampled from various parts of the Songkhla Lake basin, Thailand, in March and April 1997

Sampling location	No. of fish	Weight (g) mean±S.D.[b]	Length (cm) mean±S.D.[b]
Protosus canius			
Thale Luang	10	307±193^{AB}	35.8±7.3^{AB}
Thale Sap	9	449±215^{A}	41.9±6.5^{A}
Thale Sap Songkhla	10	162±31^{B}	30.5±1.2^{B}
Scatophagus argus			
Thale Luang	9	113±6^{A}	13.8±0.4^{A}
Thale Sap	10	162±23^{B}	15.1±0.7^{B}
Thale Sap Songkhla	8	102±11^{A}	13.2±0.5^{A}
Gulf of Thailand	10	139±17^{C}	15.2±0.5^{B}
Channa striata			
Thale Luang	8	244±38	26.8±1.4^{A}
Thale Sap	9	200±51	24.6±2.2^{A}
Khlong U Tapho	10	217±26	25.4±1.0^{AB}
Zonichtys nigrofasciata			
Thale Sap Songkhla[a]	10	248±27^{A}	24.3±1.2
South Chinese Sea	10	278±33^{B}	24.1±0.9

[a] Samples collected in the outlet connecting Thale Sap Songkhla and the Gulf of Thailand.

[b] Superscript capital letters denote significant difference among locations for means; means that do not share the same letter are significantly different ($P<0.05$).

S. argus shows significant differences ($P<0.05$, ANOVA) in length and weight between sampling locations. There is also a significant ($P<0.05$, ANOVA) difference in weight for *Z. nigrofasciata* and in length for *Channa striata*. However, in most cases the size differences are rather small and the fact that the differences are significant is due to the uniform samples at each location. The lipid content of the analysed fish is shown in Table 2. A significant difference in the lipid content of *S. argus* was found in-between locations ($P<0.001$, ANOVA). No such difference was found for the other species.

Concentrations on a lipid weight basis of *p,p*′-DDE, *p,p*′-DDD and *p,p*′-DDT and the sum of these compounds (ΣDDT) in the various fish species in different parts of the lake are shown in Table 2. *p,p*′-DDE was found in all analysed samples. *p,p*′-DDD and *p,p*′-DDT could not be quantified by GC in a proper way in the samples of *Protosus canius* from Thale Sap and Thale Sap Songkhla due to contamination during the preparation of the samples. However, *p,p*′-DDD and *p,p*′-DDT was detected and quantified in all remaining samples. The relative composition of the different DDT compounds in the fish species sampled at different locations is shown in Fig. 2. The most striking difference is the high relative amount of *p,p*′-DDT found in *Z. nigrofasciata* from both locations, where *p,p*′-DDT is actually the major constituent of ΣDDT. There were no significant spatial differences in concentrations of *p,p*′-DDE for *Protosus canius* and *Channa striata*. For *Channa striata* significant differences were found for *p,p*′-DDD, *p,p*′-DDT and ΣDDT whereas for *S. argus* and *Z. nigrofasciata* there were significant differences for all analysed substances. However, the most obvious

Table 2
Lipid amount (%) and concentrations (ng/g) of *p,p*′-DDE, *p,p*′-DDD, *p,p*′-DDT and ΣDDT in fish from the Songkhla Lake basin, Thailand, sampled in March and April 1997[a]

Sampling location	*n*	Lipid amount (%)[c]	Residue concentrations[c]			
			p,p′-DDE	*p,p*′-DDD	*p,p*′-DDT	ΣDDT
Protosus canius						
Thale Luang	10	**0.89**	**16**	**11**	**7.0**	**35**
		(0.69–1.13)	(11–23)	(6.9–18)	(4.7–10)	(24–51)
Thale Sap	9	**0.89**	**16**	imp.[d]	imp.	imp.
		(0.62–1.29)	(11–26)			
Thale Sap Songkhla	10	**0.96**	**22**	imp.	imp.	imp.
		(0.86–1.06)	(15–31)			
Scatophagus argus						
Thale Luang	9	**1.52**[A]	**96**[A]	**49**[A]	**25**[A]	**170**[A]
		(1.22–1.88)	(86–110)	(39–62)	(20–30)	(150–200)
Thale Sap	10	**2.60**[AB]	**19**[B]	**7.5**[B]	**8.9**[B]	**36**[B]
		(1.86–3.63)	(14–26)	5.5–10)	(6.3–12)	(27–49)
Thale Sap Songkhla	8	**5.18**[B]	**20**[B]	**6.8**[B]	**8.1**[B]	**35**[BC]
		(3.88–6.92)	(15–27)	(5.6–8.2)	(5.3–12)	(26–47)
Gulf of Thailand	10	**1.59**[A]	**36**[B]	**19**[C]	**14**[AB]	**71**[C]
		(0.99–2.57)	(21–62)	(12–30)	(7.7–26)	42–120)
Channa striata						
Thale Luang	8	**0.55**	**29**	**16**[AB]	**14**[A]	**61**[AB]
		(0.32–0.96)	(16–55)	6.7–37)	8.4–23)	33–120)
Thale Sap	9	**0.52**	**22**	**5.0**[A]	**2.9**[B]	**33**[B]
		(0.33–0.82)	(11–41)	1.9–13)	(1.3–6.5	(17–64)
Khlong U Tapho	10	**0.42**	**42**	**23**[B]	**19**[A]	**87**[A]
		(0.38–0.47)	(35–50)	(18–30)	(14–26)	(72–100)
Zonichtys nigrofasciata						
Thale Sap Songkhla[b]	10	**3.91**	**14**[A]	**9.7**[A]	**20**[A]	**45**[A]
		(3.14–4.87)	(8.3–25)	(6.4–15)	(13–32)	(28–72)
South Chinese Sea	10	**5.00**	**6.7**[B]	**5.4**[B]	**7.2**[B]	**19**[B]
		(4.09–6.11)	(5.4–8.4)	(4.5–6.5)	(5.5–9.4)	(16–24)

[a] Concentrations are given in ng/g on a lipid weight basis as geometric means (in bold) and 95% C.I. (in parentheses). The lipid amount given in per cent of fresh weight is also included.

[b] Samples collected in the outlet connecting Thale Sap Songkhla and the Gulf of Thailand.

[c] Superscript capital letters denote significant difference among locations for means; means that do not share the same letter are significantly different ($P<0.05$).

[d] imp., Impossible to quantify on GC due to impurity.

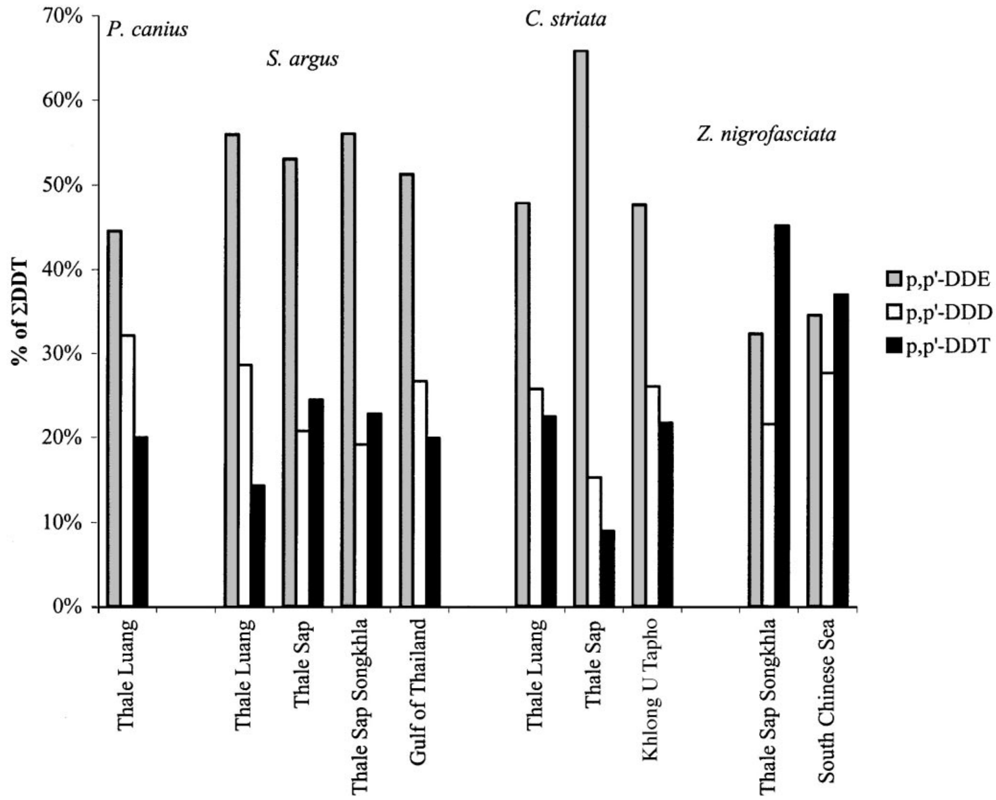

Fig. 2. The relative composition of *p,p′*-DDT and its metabolites (*p,p′*-DDD and *p,p′*-DDE) in different fish species from different locations in the Songkhla Lake basin and the Gulf of Thailand, Thailand.

spatial difference was found for *S. argus* with the highest concentrations at Thale Luang compared to the other sampling locations. The differences between the concentrations at Thale Luang and the minimum concentrations at Thale Sap and Thale Sap Songkhla were about a factor of five. Log-transformed *p,p′*-DDE concentrations were significantly negatively correlated to the lipid percentage of the muscle tissue in *S. argus* ($P<0.0001$, $r^2=0.42$), *Channa striata* ($P<0.0001$, $r^2=0.71$) and *Z. nigrofasciata* ($P<0.05$, $r^2=0.26$). This relation is shown for *S. argus* and *Channa striata* in Fig. 3.

4. Discussion

The magnitude of the ΣDDT in the fish was similar to concentrations reported in earlier studies in other parts of Thailand. Kannan et al. (1995) found ΣDDT residues with a mean of 120 ng/g lipid wt. (lw) (range 10–360 ng/g lw) in fish from the Bangkok region. Organochlorine pesticides have also been studied in green mussels (*Perna viridis*) from the Gulf of Thailand. The ΣDDT concentrations found in a survey by Siriwong et al. (1991) were in the range of 35–280 ng/g lw ($n=521$) and 66–379 ng/g lw ($n=281$) in a similar survey by Ruangwises et al. (1994).

The identified ΣDDT concentrations in the analysed fish from Songkhla Lake were well below the maximum residue levels in aquatic animals for human consumption as recommended by the Ministry of Public Health in Thailand, i.e. 5000 ng/g fresh wt. (fw) (NEB, 1989). The maximum ΣDDT concentrations in the sampled fish were about 8 ng/g fw, with a mean concentration of 0.5 ng/g fw, which cannot be regarded as hazardous, even when stricter restrictions are considered, such as the maximum Swedish ΣDDT level allowed in fish for human consumption, 500 ng/g fw (SLV, 1979).

The interspecies and spatial variation in concentration of *p,p′*-DDE were quite small within the Songkhla Lake basin with the exception of the herbivorous *S. argus* (Table 2). For *S. argus* large variations between and within locations were observed for both ΣDDT concentrations as well as lipid contents. A large variation in sexual development was also observed for this species at the different locations as well as within the locations (data not shown). The spawning period has by several authors been reported to dramatically increase the concentrations of lipophilic substances, such as ΣDDT, also if concentrations are based on lipid weight (Edgren et al., 1981; Bignert et al., 1993). The decrease of lipid storage in connection to the spawning has been suggested to be the major reason. It is, therefore, possible that the observed variation for *S. argus* in residue

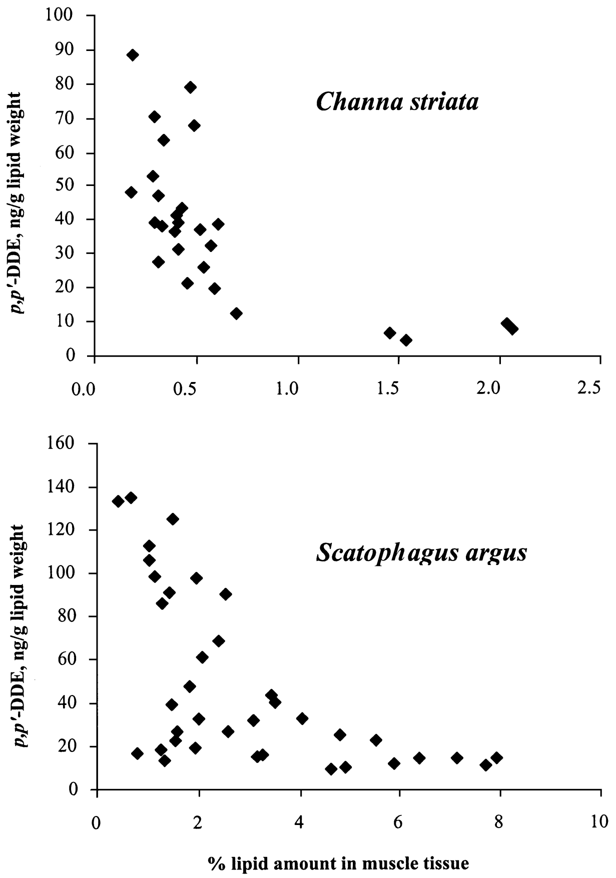

Fig. 3. Concentrations of *p,p'*-DDE in two fish species (*Channa striata* and *Scatophagus argus*) from the Songkhla Lake basin, Thailand, in relation to the lipid amount in muscle tissue.

concentrations between locations is due mainly to different reproductive phases and not spatial differences. In Fig. 3 the concentrations of *p,p'*-DDE on a lipid weight basis are presented versus the lipid amount expressed as percentage of fresh weight. The observed pattern (especially for *S. argus*) is similar to that reported by Bignert et al. (1993) in herring from different seasons, representing different reproductive stages. Thus, this further implies that different reproductive stages might be a more relevant parameter than distinct spatial exposure to explain the observed geographical differences in concentrations for *S. argus*.

Due to the discussed uncertainty regarding reproductive status of the investigated species it is speculative to draw conclusions about small differences in residue concentrations between species and locations. On the contrary, the similarities with small differences between species and locations are quite remarkable. The differences are small despite the fact that fish species from different trophic levels are included in this study. Spatial variations are in fact to be expected since DDT has been used quite extensively until 1983 and thereafter for malaria vector control by the Ministry of Public Health (Ruangwises et al., 1994).

Compared to temperate areas the ΣDDT concentrations in the analysed fish were low. In the brackish Baltic Sea in northern Europe, where the salinity gradient is similar to Songkhla Lake, the ΣDDT concentration in the mid 1990s in the omnivorous perch (*Perca fluviatilis*) was approximately 150 ng/g lw (Bignert, 1999) and approximately 500 ng/g lw in planktivorous herring (*Clupea harengus*) (Bignert et al., 1998). These values are higher than the levels in Songkhla Lake although the countries around the Baltic Sea banned the use of DDT in the early 1970s.

Several theories based on abiotic processes have been presented to explain low residue concentrations of organochlorine substances in tropical environments. Low residue concentration might be due to high temperatures and strong solar radiation resulting in high degradation rates (Samuel and Pillai, 1989). The high temperatures also lead to increased volatilisation that might result in processes often referred to as global chromatography (Sleicher and Hopcrafft, 1984; Wania and Mackay, 1993; Larsson et al., 1995). However, biological events might also influence the concentrations. The high biomass and primary production in the lake (4.24 g O_2 m^{-2} day^{-1}) (National Research Council of Thailand, 1984) likely contribute to a dilution effect of contaminants due to their distribution in the large amount of organic matter (Olsson and Jensen, 1975; Taylor et al., 1991; Larsson et al., 1992). The high productivity also increases the sedimentation of particular matter and might result in a removal and binding of contaminants to the sediments (Larsson et al., 1992). Furthermore, the high productivity and microbial activity probably increase the biological degradation of *p,p'*-DDT (Olsson and Jensen, 1975).

The low concentrations of ΣDDT in the pelagic Gulf dwelling *Z. nigrofasciata* indicate that the air transport of DDT and condensation on open water in the region is small. However, the contribution of DDT to ΣDDT in *Z. nigrofasciata* was high (Fig. 2). A pattern of a higher amount of DDT than DDE is similar to the pattern reported in fish from pelagic areas in the Swedish archipelago during the time of DDT use in northern Europe (Olsson and Jensen, 1975). The pattern of DDT compounds in *Z. nigrofasciata* thus indicates that DDT products are still circulating in the Thai environment.

Comparing the composition of DDT constituents in the marine *Z. nigrofasciata* with the other species that can be regarded as limnic (Fig. 2), the latter have a much higher degree of the more persistent metabolite *p,p'*-DDE. The same pattern has been reported for pike from the Swedish archipelago, with more *p,p'*-DDE in nutrient-rich areas close to the coast (Olsson and Jensen, 1975). In line with the results discussed by Olsson and Jensen (1975) it can be suggested that the limnic environment in the lake ecosystems, compared to the marine environment in the Gulf of Thailand, is extreme in the amount of microbial activity with a high biotic degradation as a consequence. The fast degradation of *p,p′*-DDT into its metabolites thus conceals the exposure of *p,p′*-DDT to the lake ecosystem, even though it is likely that the exposure is higher for fish living in the lake than for fish in the Gulf, due to the higher absolute values of ΣDDT in fish from the lake.

This study provides environmental data for a better understanding of the fate of DDT in tropical aquatic environments. The presented results indicate that the generally low ΣDDT concentrations in the Songkhla Lake mainly can be explained by biotic dilution and biotic degradation processes.

It also demonstrates the need of further studies on the fate of DDT in the tropics in order to evaluate global fate models. Although the reported ΣDDT concentrations in fish of Songkhla Lake do not seem to pose any threat to the human population of the area, environmental impacts should not be disregarded, especially since no studies have been conducted to deny their presence. Further research aiming at an overall assessment of the environmental pollution status in the Songkhla region, including more information on the ecological effects of organochlorine contamination, would thus provide important information for managing the lake's resources.

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