Bull. Environ. Contam. Toxicol. (1987) 38:461–466

Bulletin of Environmental Contamination and Toxicology

Fish Head Acetylcholinesterase Activity After Aerial Application of Temephos in Two Rivers in Burkina Faso, West Africa

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The operative technique presently being used in the on-going Onchocerciasis Control Programme (OCP) in the Volta River Basin in West Africa, is a weekly aerial application of the Organophosphorus larvicide, temephos 0,0,0'0'-tetramethyl 0,0'-thiodi-p-phenylene phosphorothioate), to all breeding sites of Simulium damnosum s.l. in the rivers in the programme area. The control programme is scheduled to last for twenty years, and began in 1974.

One of the main aspects of the OCP is the monitoring of the environment to assess the effects of the larvicide application on the non-target organisms. The importance of this aspect of the programme cannot be over-emphasized since many of the treated rivers flow into man-made lakes of major economic importance to the countries in the region and moreover all the treated rivers are associated with the economically important fish industry (Davies et al. 1978). Since 1975, hydrobiological teams have been monitoring the long term effect of the larvicide on the fish populations in the rivers in the treated areas. The short term effects of temephos on fish under laboratory conditions have been reported by several workers including Butler (1965), Von Windegulth and Patterson (1966) and Galleta (1968). Abban and Samman (1980) have given an account of the effect of temephos treatment on fish catches in the rivers Oti and White Volta in Ghana.

This paper reports on the effect of six years of aerial application of temephos on the acetylcholinesterase (AChE) activity per whole head of four commercial fish species from two treated rivers in the Ouagadougou area in Burkina Faso. It is hoped that this study will assist in providing a basis for the overall evaluation of the OCP monitoring programme.

MATERIALS AND METHODS

Four commercial fish species, namely Tilapia galilaea, Tilapia nilotica, Alestes nurse and Schilbe mystus, were randomly caught with cast nets from two treated rivers, the Red Volta and the

White Volta, and from an untreated barrage at Loumbila near Ouagadougou. At the time of the fish collection, both rivers had received six years of weekly temephos treatments at a dosage rate of 0.05ppm per 10 minutes of river discharge. The sampling points were at Pont de Po on the Red Volta and at the point where the Ouagadougou-Kou Pela road crosses the White Volta. The fish collection was done during the peak of the river discharge. The fish from the untreated barrage were used as controls.

The colorimetric method of Ellman et al. (1961) was used to measure the acetylcholinesterase (AChE) activity. The fish was decapitated and the operculum removed. The head was weighed and homogenised using a Potter Elvehjin homogeniser in a 0.1m phosphate buffer solution (pH 7.0) so as to make a 10% homogenate solution. The fish homogenate was next diluted with the phosphate buffer to make a 0.1%solution. 4.0ml of this homogenate solution was measured into the photometric cuvette and 0.01ml dithiobisnitrobenzoic acid (DTNB) solution (0.01M) was added and mixed. The spectrophotometric zero was set with this solution after which 0.01ml acetylthiocholine (ASCh) iodide solution (0.2M) was added and mixed. Immediately after mixing, the rate of the yellow colour production was followed by measuring the absorbance (A) at 412nm every 30 seconds for 120 seconds using a Coleman 295 Spectrophotometer. Duplicate measurements were made on each fish homogenate. At the end of each measurement, 4λ anti-cholinesterase solution was added to check for any non-enzymic hydrolysis. No increase in the yellow colour was recorded after the addition of the anti-cholinesterase solution, indicating that there was no other hydrolysis than that due to the acetylcholinesterase.

Change of absorbance per minute (△A/min) was then calculated and the rates converted to absolute units by the formula:
Umol ASCh hydrolyzed/min/gm fish head weight.

$$= \frac{\triangle A/\text{min} \times \frac{\text{mls soln. in cuvette}}{\text{mls homogenate in cuvette}} \times \frac{100}{\%\text{homog.}} \times \text{head wt.} \times 10^6}{1.36 \times 10^4 \times 10^3}$$

where

1.36×10^4 = extinction coefficient of DTNB
10^3 = conversion mol/l ⟶ mol/ml.
10^6 = conversion mol/ASCh/min/gm. headweight ⟶ Umol ASCh/min/gm. headweight.

RESULTS AND DISCUSSION

The results of the acetylcholinesterase activity measurements are summarized in Tables 1-4. The enzyme activity is expressed as Umol acetylthiocholine (ASCh) hydrolized per minute per gram fish headweight.

Table 1. Acetylcholinesterase (AChE) activity of Tilapia nilotica from the R. White Volta, R. Red Volta and the barrage de Loumbila.

River	No. of fish analysed	Fish hd.wt. gm (mean ± S.D)	Total AChE activity in Umol ASCh/min/gm hd. weight (mean ± S.D)
White Volta (treated)	13	1.06 ± 0.51	7.18 ± 3.09
Red Volta (treated)	11	1.03 ± 0.39	7.12 ± 1.65
barrage de Loumbila (control)	7	1,35 ± 1.03	7.76 ± 4.72

Table 2. Acetylcholinesterase (AChE) activity of Tilapia galilaea from the R. White Volta, R. Red Volta and the barrage de Loumbila.

River	No. of fish analysed	Fish hd.wt. gm (mean ± S.D)	Total AChE activity in Umol ASCh/min/gm hd. weight (mean ± S.D)
White Volta (treated)	13	1.77 ± 0.91	10.63 ± 4.86
Red Volta (treated)	13	1.65 ± 0.83	9.95 ± 2.15
barrage de Loumbila (control)	13	1.59 ± 1.04	9.76 ± 5.26

Table 3. Acetylcholinesterase (AChE) activity of Alestes nurse from the R. Red Volta and the barrage de Loumbila.

River	No. of fish analysed	Fish hd.wt. gm (mean ± S.D)	Total AChE activity in Umol ASCh/min/gm hd. weight (mean ± S.D)
Red Volta (treated)	5	2,51 ± 0.73	10.71 ± 2.19
barrage de Loumbila (control)	17	1.46 ± 0.38	8.51 ± 2.92

Table 4. Acetylcholinesterase (AChE) activity of Schilbe mystus from the R. Red Volta and the barrage de Loumbila.

River	No. of fish analysed	Fish hd.wt. gm (mean ± S.D)	Total AChE activity in Umol ASCh/min/gm hd. weight (mean ± S.D)
River Volta (treated)	8	2.66 ± 0.84	19.96 ± 4.79
barrage de Loumbila (control)	15	3.17 ± 0.54	20.43 ± 3.47

In all the four species studied, namely Tilapia galilaea, Tilapia nilotica, Alestes nurse and Schilbe mystus, there was no significant difference ($P>0.05$) (Student's t-test) between the total AChE activity per whole head of fish from the treated rivers and their respective control groups (Tables 1-4). This finding confirms an earlier observation in a similar study in the Bobo-Dioulasso area in Burkina Faso (Scheringa et al. 1981). However, Gras et al. (1982) have reported that the brain AChE activity of Tilapia guineensis was inhibited when the fish was exposed to 0.05mg/1 temephos for 10 minutes after three consecutive weekly treatments in the laboratory.

Inhibition of AChE activity in fish may cause it to exhibit a number of toxic symptoms. These include a reduction in the ability of the fish to tolerate reduced oxygen tension (Eaton 1970), a slow-down of its reflexes and swimming movements (Pelissier et al. 1982) and a reduction in the feeding activities of the fish (Verma et al. 1970). In a river, fish under these stresses will be prone to easy capture by fishermen and other predators and as a result the fish populations in that river could be greatly reduced.

In the present study, no inhibitory effect was found on the head AChE activity of the four species of fish from the Red and White Voltas which could be attributed to the treatment of the rivers with temephos. It appears that during larviciding, most of the fish in the treated rivers avoid the impact of the larvicide by swimming downstream. Goldfish (Carassius auratus) was observed to avoid water containing fenithrothion (Scherer 1975), while Gambusia species and sheephead minnow avoided water containing chlorpyrifos (Hansen 1969). Abban and Samman (1980) observed erratic movements of fish indicated by splashing within 5 minutes after temephos application in the River Oti in Ghana and attributed this action to the attempts by the fish to avoid the chemical. It has recently been found that during chlorphoxim treatment of the river Marahoue in the Ivory Coast (Antwi 1984), the larvicide induced a reduction in brain AChE of caged Tilapia zilli placed up to a distance of 1.0km downstream from its point

of release and that the larvicide application did not affect the enzyme activity of the caged fish placed at the 3.0km point. Thus, the river waters beyond 3.0km downstream of the treatment site provide a safe place for the fish during temephos application.

In addition to the avoidance reaction of the fish during river treatment, factors which have contributed to the survival of the fish populations in the OCP treated rivers after more than six years of weekly temephos treatment include the non-persistent nature of temephos in the aquatic environment and the great volume of the flowing river water (which ensures rapid dilution of the larvicide from the point of release).

Acknowledgments. I thank T. Sineyogo at the OCP headquarters, Ouagadougou, Burkina Faso, who collected the fish samples. I also thank the Director of the Institute of Aquatic Biology for the opportunity to undertake these studies. Funds for this study were provided by the Onchocerciasis Control Programme of WHO.

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Received June 1, 1985; accepted June 24, 1985