

## Endocrine Disruptive Potentials in Surface Water Samples from Taihu Lake, Yangtze Delta

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**ABSTRACT :** Taihu Lake is a major water source for part of Yangtze Delta, which is one of the most urbanized and economically prosperous areas in China. In last couple of decades, some parts of the lake were highly polluted due to eutrophication. This study analyzed dioxin-like potential and mutagenic potential in surface water samples from Taihu Lake. The samples were prepared by XAD-2 resin procedure. A batch of biological assays, including dioxin-like potential microassay with the rat hepatocyte cell line H411E, and Ames test was employed in the research. Results showed that four water samples have high content of dioxin-like biological potential, the highest activity TEQ to 2,3,7,8-TCDD was 48 pg/l in sample 1. The mutagenic effect with reading-frame shifting mechanism was confirmed in 3 of 4 samples. The effective sewage treatment facilities and reliable monitoring surveillance system are urgently needed for this area.

**Key Words :** Surface water, Dioxin-like potentials, Ames test, Yangtze Delta

### I. INTRODUCTION

Taihu Lake (water surface area: 2334 km<sup>2</sup>) is the third largest freshwater lake in China. Taihu Basin is one of the most developed and urbanized areas in this country. In past few decades, with high-speed economic development and speedup urbanization along the Lake, growing amount of industrial, agricultural and municipal wastewater have been sluiced into the lake. Some parts of the lake have been severely polluted due to eutrophication. Most of discharged organic pollutants are highly hydrophobic, which might pose a long-term hazard to the aquatic biota and to the public health. Some agents might result in a series of harmful consequences, such as tumors, malformation, endocrine disruption and immune deficiency (Kolasa and Pickett, 1992).

Presently in this country only a few of chemicals and environmental parameters are under routine monitoring with a number of analytical chemical approaches. The introduction of different bioassay approaches (Blaise *et al.*, 1988), as an alternation and comple-

ment to present monitoring system might bring about more information for better pollution assessment and for the strategy design of rehabilitation of yet degraded ecosystem.

The aim of this study was to use a batch of short-term bioassays to assess the dioxin-like and genotoxicity potentials of pollutants in surface water from one area in Taihu Lake, Yangtze Delta.

### II. MATERIALS AND METHODS

#### 1. Sample Collection and Preparation

In January, 1999 water samples were collected at the depth of 0 to -0.5 meter from four defined sampling position on one water area (total water surface area: 135 km<sup>2</sup>) in north-west part of the Taihu Lake with the aid of GPS (Global Positioning System) for exact positioning. This area is in the suburb of a medium size industrial city (Population: 4 million). It serves as principal water source for the city, at the same time the city itself discharged most of its industrial and municipal wastewater into the Bay. In last few decades this area was listed to be one of the heavily polluted water bodies in China.

Surface water (0~0.5 m) was collected from every

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Abbreviations: 2,3,7,8-TCDD: 2,3,7,8-tetrachlorodibenzo-p-dioxin, DMSO: dimethyl sulfoxide, TEQ: Toxic Equivalents to 2,3,7,8-tetrachlorodibenzo-p-dioxin

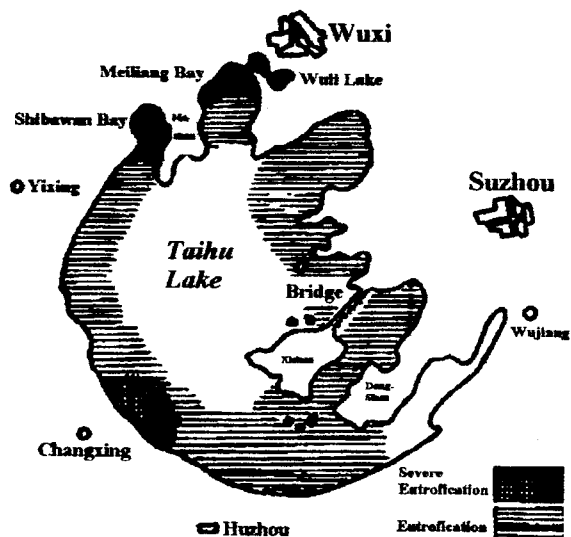


Fig. 1. Taihu Lake and its Surrounding.

sampling site. The concentrating was conducted by passing XAD-2 resin column (30 ml of bed volume) at a flow rate of 50 ml/min or so. Then excess water in the column was blown out with nitrogen flow (99.999%). Adsorbed compounds were eluted with 50 ml acetone. The elute volume was reduced to near dry by evaporation with nitrogen flow at room temperature. Subsequently, these extracts were dissolved with 5ml dimethyl sulfoxide (DMSO, p.a., Merck), for a concentration factor of 5000 with respect to the original water volume (Shen *et al.*, 2000).

## 2. Mutagenicity Assay

The procedure of bacterial culture, the verification of genetic markers and plate incorporation assay were essentially as described by Maron and Ames (1983). *Salmonella typhimurium* TA98 and TA100 were used for the *His*<sup>r</sup> reverse mutation test (Ames test).

## 3. Dioxin-Like Potential Assay

The microassay was according to the methods of Donato *et al.* (1993). The procedure is based on the metabolism by cells cultured in microwells with appropriate substrates at noncytotoxic concentrations. The assay of 2,3,7,8-TCDD activity with the rat hepatocyte cell line H4IIE was based on the induction of ethoxyresorufin-O-deethylase (EROD) activity by

2,3,7,8-TCDD-like effect. EROD activities were determined with intact cells cultured on 96-well plates. Substrates are directly added to the wells and allowed cells to metabolize and the resorufin formed is fluorimetrically measured in microplates. All the dioxin-like potentials are expressed as Toxic Equivalents (TEQ) to 2,3,7,8-TCDD.

## III. RESULTS AND DISCUSSION

### 1. Dioxin-Like Potential Pollution in the Water Samples

Dioxins are of highly hydrophobic and extremely stable in the nature. The potential detected was standardized with 2,3,7,8-TCDD. 1L water of the four samples showed the dioxin-like potential as 48 pg, 34 pg, 36 pg, 28 pg of 2,3,7,8-TCDD could cause, respectively. USEPA National Primary Drinking Water Standards sets MCL (Maximum Contaminant Level) for dioxin at 30 pg/l. A suggestion was proposed that the human body permitted exposure level to TCDD is 10 pg/kg·day (Heuvel and Lucier, 1993).

### 2. Mutagenicity Potentials in the Surface Water Samples

The mutagenicity of the surface water samples collected in January 1999, were examined with the Ames test using *S. typhimurium* TA98 and TA100 strains. Mutagenicity result is regarded as positive only if the observed mutation frequency is more than doubled, compared with spontaneous background of the system. Table 1 shows that significant mutagenic

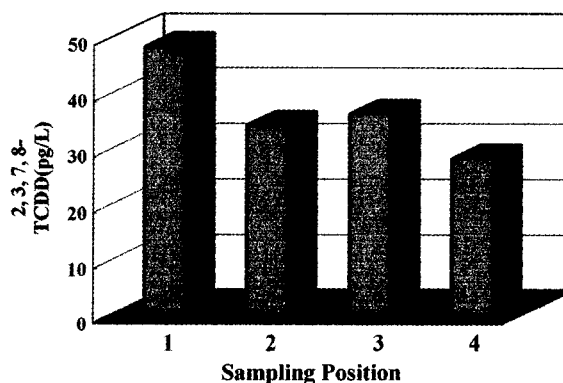


Fig. 2. Dioxin-like potential in surface water samples.

**Table 1.** Mutagenic potential of water samples by Ames test with or without S9

	S9	Water-Equivalent (ml)	Ames test (TA98) Induction factor	Ames test (TA100) Induction factor
Sample 1	-	250	4.6	0.8
	-	500	6.4	1.2
	-	1000	7.8	1.1
	+	250	3.2	1.1
	+	500	4.9	1.0
	+	1000	5.9	1.3
Sample 2	-	250	3.0	1.1
	-	500	5.3	1.2
	-	1000	7.9	1.3
	+	250	3.0	0.9
	+	500	4.4	1.1
	+	1000	5.5	1.3
Sample 3	-	250	3.4	1.0
	-	500	4.6	1.0
	-	1000	5.4	1.4
	+	250	1.6	0.9
	+	500	2.0	1.2
	+	1000	2.5	1.1
Sample 4	-	250	1.0	1.0
	-	500	1.3	1.0
	-	1000	1.1	1.0
	+	250	1.1	0.9
	+	500	1.0	0.9
	+	1000	1.0	1.0
NOPD (mg/pl)	-	5	4.0	
2-AF (mg/pl)	+	5	13.6	
NaN <sub>3</sub> (µg/pl)	-	1		7.5
2-AF (µg/pl)	+	2.5		1.9

activity demonstrated in sample 1,2 and 3 with TA98 under the conditions with or without presence of eucaryotic metabolic activation system (S9). This indicates the predominance of direct-acting genotoxins in water and the genotoxins can be detoxified by certain degrees with S9 mix. Compared the results obtained in August 1998 (Shen *et al.*, 2000), mutation ratios of sample 3 has been lowered. The genotoxicity was found with strain TA98 indicating that frameshifting mutations were the predominating mode of action, nevertheless, base substitution mutation were not recorded with strain TA100 in this study.

Above data were obtained only from surface water samples in the lake. Considering that the pernicious compounds are mostly hydrophobic and show strong

tendency to be enriched in the sediments. Taihu Lake, either its surface water or the sediment, should be kept under even tighter monitoring. The effective sewage treatment facilities and reliable monitoring surveillance system are urgently needed for this area.

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