

Fish Toxicity Tests for Endocrine Disrupting Chemicals

May 2001

Chemicals Evaluation and Research Institute, Japan

Hiroshi TADOKORO

Ecotoxicology is a research area for adverse effect of chemicals to ecosystem. While toxicology is directly related to human health, relation between ecotoxicology and human health is indirect. In this connection, public awareness of environmental effect has been rather behind that for health effects. Historically the first concern in ecotoxicology was adverse effect of pollution on organisms in waters receiving industrial wastewater. In many cases, toxicity of heavy metals was likely cause of the effects. Then, scientists started to pay attention to fish kill, mostly accidental, occurred by agricultural chemicals. It was the starting of research on toxicity of organic chemicals to environmental organisms. However, most data obtained were regarding acute toxicity to fish. Later data collection has been extending from fish to other organisms such as crustacean and algae, as well as from short-term (acute) to long-term (chronic). At the same time, international guidelines such as OECD guidelines have been developed.

During the last decade, a new serious issue arose, namely some chemicals might have endocrine disrupting effect and the disruption might result in reproductive impairment in human and wild life. Since scientists found that existing test methods were inadequate to detect such activity of chemicals, they started to develop new methods or to modify existing test methods to be able to detect such chemicals.

In the area of ecotoxicology, in the process of development and modification of test method, new and specific end points for endocrine disrupters such as measurement of vitellogenine and hormone levels, sexual behavior, secondary sex characteristics, gonad-somatic index, hepato-somatic index and gonadal histology were incorporated in addition to classical and general endpoints such as growth, maturation, and reproductive performance. So far efforts made in ecotoxicological area are mostly devoted to detection of estrogenic activity as well as to development of fish tests methods in Japan like other countries. Now, we have in vitro methods such as

binding assay using fish receptor as screening (or prescreening) method and in vivo methods such as vitellogenin assay, partial life cycle test (modified or prolonged early life stage toxicity test), reproduction test, and full life cycle test. International collaboration work for validating methods has started under OECD Expert Consultation on Endocrine Disrupters Testing in Fish. Recently, OECD released two new draft guidelines, i.e. 14-day Fish Assay for Identification of Potential Endocrine Disrupting Chemicals and Short-term Reproduction Test with the Fathead Minnow for Identification of Endocrine Disrupting Chemicals. More than 10 laboratories including Japanese laboratories are going to participate ring test using these draft guidelines.

In Japan we have been mainly using medaka (*Oryzias latipes*) to develop a series of test methods because of its advantage for a test. Basic research groups in medaka biology have been supporting test method developing groups in providing special medaka strain, e.g. dr-R and FLF, and information to select test condition and end point.

Now developed test methods are under validation work with designated positive reference substances such as 17β -estradiol, ethinyestradiol and methyltestosterone as well as some antagonist and agonist. Validation work for short-term test methods such as vitellogenin assay is coming to the final stage. For long-term test methods such as a full life cycle test, it may take some more time. At the same time we have started to collect data using applicable methods to obtain information whether candidate chemicals are actually endocrine disrupters according to the priority setting in test chemicals by an expert group under SPEED 98' in Japanese Ministry of the environment.