

## Quadrant II – Transcript and Related Materials

**Programme: Bachelor of Science (Third Year)**

**Subject: Zoology**

**Course Title: Environmental Biology and Toxicology**

**Course Code: ZOC 109**

**Unit: Unit 8- Introduction to Toxicants**

**Module Name: Toxicity tests- Acute and Chronic, LC<sub>50</sub>, LD<sub>50</sub>, and EC<sub>50</sub>**

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### NOTES

#### TOXICITY TESTS: ACUTE AND CHRONIC, LC<sub>50</sub>, LD<sub>50</sub>, EC<sub>50</sub> VALUES

##### Introduction

Toxicology is “the study of the adverse effects of chemical, physical, or biological agents on living organisms and the ecosystem .”

Toxicity: The degree to which a substance (a toxin or poison) can harm humans or animals.

A toxicity test, is designed to generate data concerning the adverse effects of a substance on human or animal health, or the environment.

Toxicity tests are experiments or trials designed to assess or to evaluate the doses/ concentration of xenobiotics and the duration of exposure required to produce a criterion effect. The criteria of the effect may be mortality or death, of the target species, or any other parameter such as histological, physiological, behavioral, immunological, , heamatological, biochemical, etc. Several characteristics such as age, sex and individual susceptibility can influence the degree of poisoning in exposed organism and are to considered while planning the study.

**Toxicity testing** helps to identify possible adverse effects of exposure to environmental agents, to develop dose-response relationships that can elucidate the severity of effects associated with known exposures, to clarify risks to health posed by environmental exposures and provide support for effective risk-management decisions and ultimately to predict the possible and probable effects of exposure in humans.

**Test organisms-** Selection of appropriate test organisms is one of the essential steps for the toxicity test. This is not only necessary for the accuracy of results, but also for the extrapolation of meaningful, and ecologically significant results. Animals used for testing are Daphnids, Midge, Oyster, Mussels, Grass shrimp, Fishes (Zebra fish, Carps), Birds (Pigeons, Domestic fowls, etc.), Mammals (Laboratory rat, Laboratory mouse, Guinea pig, Rabbit, Dogs, Pig, Monkey, etc.), etc.

**Test chemical /toxicant:** Pure chemical or commercial formulation or mixture of chemicals, factory effluent, pesticides, radiations, etc.

**Routs of administration-**The choice of the route of administration depends on the physical and chemical characteristics of the test substance, its intended field of application, and the predominant route of anticipated human exposure

For Non-aquatic forms The Oral route is commonly used. Xenobiotic is given by force feeding. Mixing with food is avoided. A vehicle like solvents, carriers, surfactants, etc. is usually required in dissolving or suspending the xenobiotic to facilitate its administration. Chemicals/ Toxicants can be administered by the oral, dermal or inhalation route in Non-aquatic forms and on aquatic organisms is done in the aquatic medium. During the test period the animal forcibly inhales, ingests or is otherwise exposed to the substance.

A test material is administered by various routes to determine its ability to induce toxicity. Generally the toxicant are administered through the route by which humans would be exposed. Oral, Intranasal, Intravenous, Subcutaneous, Dermal, Intradermal, Intramuscular, Intraperitoneal, Intravaginal, Intraurethral, Ocular, etc.

### **Types of toxicity tests**

I). Based on no. and conditions of species - Single species test, Multispecies test and Ecosystem test.

II) Based on exposure of toxicants - Single dose test & Multiple dose test

III) Based on length of exposure- Acute and Chronic toxicity tests.

Acute toxicity tests are short-term tests that measure the effects of exposure to relatively high concentrations of chemicals.

Chronic toxicity tests generally are longer-term tests that measure the effects of exposure to relatively lower, less toxic concentrations.

**Acute toxicity:** Acute toxicity may be defined as severe effects experienced by the organisms during short-term exposure to toxicants. Acute toxicity tests are principally designed to determine the dose/concentration of a test material producing deleterious effects on a group of test organisms during short-term exposure under controlled laboratory conditions. Acute

toxicity tests are acute lethality tests. Usually 50% response is the most accepted and reproducible measure of toxicity and 96 hour is the standard exposure time.

Acute toxicity tests provide rapid estimates of dose/concentration of test chemicals that cause direct irreversible harm to the organisms. It thus provides a practical means for- estimating the upper limit of xenobiotics producing toxic effects; evaluating relative toxicities of various toxicants; determining relative sensitivity of different organisms to toxicants; evaluating the effects of environmental variables (e.g. water quality and other parameters) on the toxicity of chemicals; understanding the significance of duration of exposures; developing an understanding of dose-response relationship or concentration-response relationship; assessing doses /concentrations for chronic exposures; determining most sensitive species of organisms and indicator species.

Acute Toxicity - damages organisms immediately upon exposure & is distinguished from chronic toxicity, which describes the adverse health effects from repeated exposures, often at lower levels, to a substance over a longer time period (months or years).

**Chronic toxicity:** Chronic toxicity tests are defined as tests that characterize adverse effects following repeated administration of a test substance over a significant portion of the life span of the test species. Establishing the duration of a chronic study is based on the anticipated human or environmental species' exposure. It provides information on the possible health hazards likely to arise from repeated exposure over a considerable part of the lifespan of the species used. The study will provide information on the toxic effects of the substance, indicate target organs and the possibility of accumulation.

**Chronic toxicity tests helps in** -The identification of the chronic toxicity of toxicants; The identification of target organs and to study adverse effect of toxic substances on the structure and functions of organs, tissues and cells after prolonged period of exposure; Characterisation of the dose-response relationship; To evaluate no effect levels or safe levels of toxicants; the identification of the carcinogenic, teratogenic, mutagenic properties of the toxicants; To understand effect of toxicants on Histology, Histopathology, Hematology, cardiac functioning, Body systems and processes, physiological parameters, behavior, genetics, life cycle, etc. on the exposed animals; To understand mechanism of action of toxicants.

### **LD<sub>50</sub> and LC<sub>50</sub>**

**LD<sub>50</sub>** (median lethal dose) & **LC<sub>50</sub>** (median lethal concentration) are important terms in toxicology. These are parameters for different chemical compounds/ xenobiotic which describe the amount of a substance that can cause death.

LD stands for Lethal Dose; LD<sub>50</sub> refers to the dose needed to kill half (50%) of the sample organisms that ingest it.

LC -Lethal Concentration; LC<sub>50</sub> refers to the concentration in air or water that will kill half of the test organisms that are exposed to it.

It is one way to measure the short-term poisoning potential (acute toxicity) of a material.

LD<sub>50</sub> can be found for any route of entry or administration but dermal (applied to the skin) and oral (given by mouth) administration and injection methods are the most common.

LC<sub>50</sub> is the concentrations of the chemical in air or water that kills 50% of the test animals during the observation period .

Observation period may be of minutes, hours (up to 24) or days (up to about 2 weeks) but rarely longer. LD<sub>50</sub> and LC<sub>50</sub> is done to study / compare potency or intensity of different toxicants. It gives a measure of the immediate or acute toxicity of a toxicant in the strain, sex, and age group of a particular animal species being tested. Changing any of these variables (e.g., type animal or age or size) could result in finding a different LD<sub>50</sub> and LC<sub>50</sub> value.

The unit of measurement for LD<sub>50</sub> is milligrams of substance per kilogram body weight of the population whereas the unit for LC<sub>50</sub> is milligrams per cubic meter or ppm (part per million). LD<sub>50</sub> (oral, rat) - 5 mg/kg means 5 g/kg. So, the example "LD<sub>50</sub> (oral, rat) 5 mg/kg" means that 5 milligrams of that chemical for every 1 kg. body weight of the rat, when administered in one dose by mouth, causes the death of 50% of the test group.

- LC<sub>50</sub> value reported state the kind of test animal studied and the duration of the exposure, e.g., LC<sub>50</sub> (rat) - 1000 ppm/ 4 hr or LC<sub>50</sub> (mouse) - 5mg/m<sup>3</sup>/ 2hr.

In general, the smaller the LD<sub>50</sub> or LC<sub>50</sub> value, the more toxic the toxicant is. The larger the LD<sub>50</sub> or LC<sub>50</sub> value, the lower the toxicity.

### **Why these tests?**

To compare the toxic potency or intensity of different toxicant. To carry out lethality testing by measuring how much of a toxicant/ substance is required to cause death. As an aid in establishing occupational exposure limits. To help develop guidelines for the use of appropriate safety clothing and equipment. As an aid in developing emergency procedures in case of a major spill or accident. It is an index determination of medicine and poison's virulence (the severity or harmfulness). The LD<sub>50</sub> / LC<sub>50</sub> value is a base from which other doses could be designed in subacute and chronic toxicity experiments. The LD<sub>50</sub> is important for the prediction of human lethal dose and for the prediction of the symptomatology of poisoning after acute overdosing in humans.

LD<sub>50</sub> are not conducted on humans. Broadly "lethal amount" is calculated from an animal LD<sub>50</sub> by multiplying the animal LD<sub>50</sub> by a number representing average human weight. Example : If the LD<sub>50</sub> is 50 mg/kg: The lethal amount for a child would be 50 mg/kg times 10 kg, which equals 500 mg (about 1/8 tsp.) The lethal amount for an adult would be 50 mg/kg times 70 kg, which is 3,500 mg (about 3/4 tsp.)

### **EC<sub>50</sub>**

## EC<sub>50</sub>(50% Effective Concentration) : Effective Concentration 50

The median effective concentration (**EC<sub>50</sub>**) is the concentration of a substance in an environmental medium expected to produce a certain effect in 50% of test organisms ) in a given population under a defined set of conditions.

EC<sub>50</sub> refers to the concentration of a drug, antibody or toxicant which induces a response halfway between the baseline and maximum after a specified exposure time. In a pharmacological context, it is the concentration of a drug that is necessary to cause half of the maximum possible effect. This value is particularly important in pharmacology, because it serves as an indication of drug potency (the amount of drug required to produce a given effect).

Ex. : Concentration of chemical resulting sterility in 50 percent of test animals.

It is the dose required for an individual to experience 50% of the maximum effect.

The **EC<sub>50</sub>** and the **LC<sub>50</sub>** are often used in ecotoxicology as an indicator of the toxicity of a compound to the environment.

Traditionally, EC<sub>50</sub> is expressed as a molar concentration. The **EC<sub>50</sub>** indicates how much of a drug is needed to achieve 50% of the maximum response. The more potent a drug, the smaller the **EC<sub>50</sub>** will be. This value is obtained from a dose-response curve.

