Time-dependent toxicity of pesticides and other toxicants: implications for a new approach to risk assessment

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Introduction

A fundamental goal of toxicology is to determine safe levels of exposure to potentially poisonous substances for humans and the environment. Traditionally, safe levels of a chemical have been derived from the non-observable effect level (NOEL) estimated in laboratory toxicity bioassays with organisms which are representative of certain taxa. There are fundamental problems with the validity of this approach, both conceptual and statistical in nature (Landis & Chapman 2011).

The outdated NOEL concept can be replaced by the no-effect concentration (NEC) level (Kooijman et al. 1996), which assumes that toxic chemicals do not have any effect on a population of organisms at very low concentrations. Recent developments in ecotoxicology, however, suggest that some toxicants can produce effects at any concentration level provided their exposure time is sufficiently long (Sánchez-Bayo 2009; Tennekes 2010). Consequently, risk assessment of these chemicals, which includes neonicotinoid insecticides, some carcinogenic substances and certain metallic compounds, may require entirely new approaches.

Methodology

Time-to-event (TTE) approaches measure the times to respond for all individuals in a toxicity bioassay, and provides information on the acquired doses as well as the exposure times needed for a toxic compound to produce any level of effect on the organisms tested (Newman & McCloskey 1996). Consequently, extrapolations and predictions of toxic effects for any combination of concentration and time are now made possible (Fig. 1).

Sound toxicological models that consider the response of organisms to chemicals with time have demonstrated the simple relationship that exists between concentration (C) and time of exposure (t), usually measured by the median time to effect (T_{50}) (Tennekes & Sánchez-Bayo 2012). In essence, the total dose of toxicant is the product of concentration and time, and three types of responses can be defined based on the relative velocities of association and dissociation of the target receptor with the toxic chemical.

$$Dose = C \bullet T_{50}^{\,n} \quad \text{where} \quad n = 1/b$$

Chemical	C (µg/L)	T50 (days)	D = C * T50	n
Imidacloprid	60000	3	197500	2.40
	6700	18	121158	
	2220	14	31080	
	750	33	24594	
	250	52	13000	
Selenium	2000	0.25	500	1.03
	1600	0.38	600	
	1200	0.50	600	
	800	0.71	567	
	158	3.25	514	
Zinc	750	0.96	719	0.47
	500	1.3	667	
	250	1.9	490	
	125	32	4000	
	59	163	9617	

$$\ln T_{50} = a - b \bullet \ln C$$

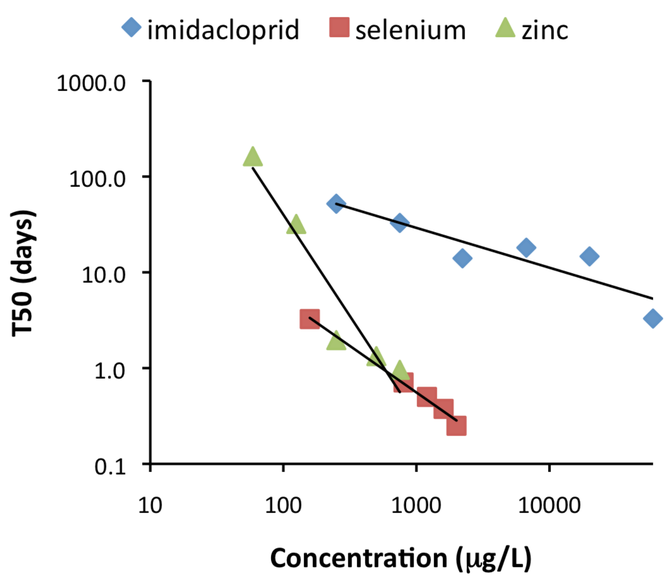


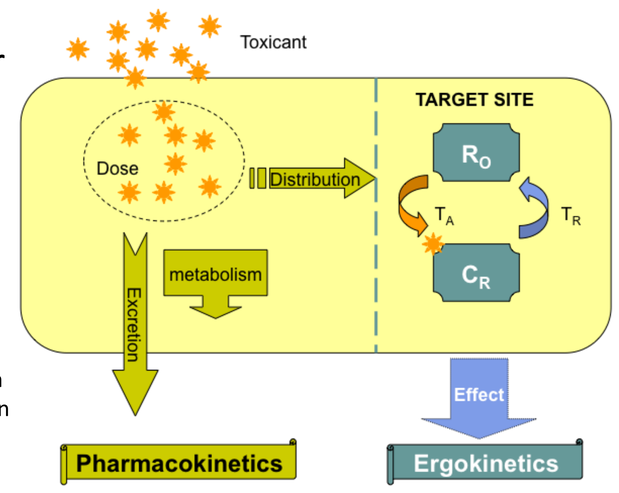
Fig 1. Response of *Daphnia magna* to three toxicants with different mode of action (Sánchez-Bayo, 2009).

Druckrey-Küpfmüller model

for $C_R << R_o$

$$\frac{C_R}{R_O} = \frac{C}{R_O} \times \frac{T_R}{T_A}$$

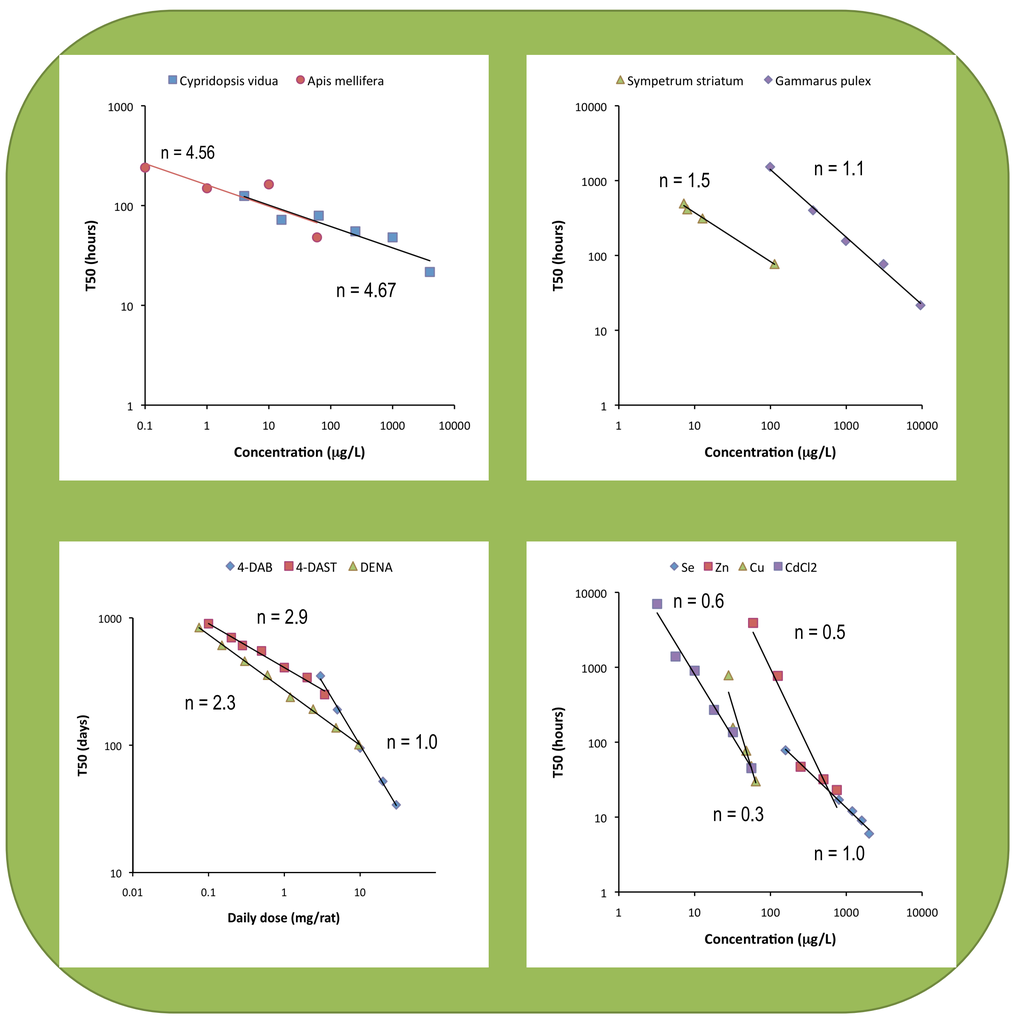
C concentration at target site
R_O initial receptor's concentration
C_R bound receptor's concentration
T_A time constant for association
T_R time constant for dissociation



neonicotinoids carcinogens → n > 1 → **effect ~ ∫ ∫ C dt** → Irreversible Cumulative effect

thiacloprid carcinogens selenium → n = 1 → **effect = C x t** → Irreversible

metals, metaloids → n < 1 → **effect ~ C & t** → Elimination process



Examples

Above left: imidacloprid effects on a freshwater ostracod (*Cypridopsis vidua*) and the honeybee (*Apis mellifera*), showing irreversible effects reinforced by exposure time [Data sources: Sánchez-Bayo 2009; Suchail et al. 2001]

Above right: thiacloprid effects on dragonfly larvae (*Sympetrum striatum*) and a freshwater amphipod (*Gammarus pulex*), showing irreversible effects [Data source: Beketov & Liess 2008].

Below left: median tumor induction period in rats exposed to three carcinogens: 4-dimethylaminobenzene (4-DAB), 4-dimethylaminostilbene (4-DAST) and diethylnitrosamine (DENA) showing irreversible effects, which in the case of 4-DAT and DENA are reinforced by exposure time [Data source: Tennekes 2010].

Below right: toxic effect of metals, metalloids and metallic compounds on *Daphnia magna*. Toxic effects of Se are irreversible, whereas Zn, Cu and $CdCl_2$ are eliminated over time [Data sources: Hoang et al. 2007; Kooijman, 1981].

Risk assessment

The Colony Collapse Disorder in honeybees (*Apis mellifera*) is explained by the chronic toxicity of neonicotinoids. Based on data by Suchail et al. (2001), predicted times to 50% mortality after ingestion of nectar or pollen contaminated with imidacloprid are shown below

LnT50 = 5.11 – 0.22 lnC (r^2 =0.75)

Residues in	Imidacloprid (µg/L or kg)	Predicted T_{50} (days)	HQ = PEC/NOEL
nectar	1	6.9	0.05
	3	5.4	0.15
pollen	0.7	7.5	0.035
	10	4.2	0.5

By contrast, hazard quotients based on NOEL of 20 µg/L suggest that imidacloprid poses no danger to honeybees. Standard risk assessments, therefore, fail to acknowledge the chronic toxicity of systemic insecticides.

Pollen contamination with insecticides reaches levels of 40-70% in France and the USA, with 11% of pollen containing imidacloprid (Bonmatin et al, 2007; Mullin et al, 2010). Given the worldwide use of this insecticide in agriculture and forestry, it is not surprising that honeybees and wild bee pollinators are declining rapidly in many countries.

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Conclusions

Time-to-event bioassays are essential tools that allow prediction of toxic effects for any concentration of chemical in the environment. Understanding the patterns of toxicity over time should become the rule in ecotoxicology, replacing NOELs and LC/LD_{50} at fixed times.

Three patterns of toxicity can be identified based on $C\ T_{50}^{\,n}$ = constant

- Toxicity effects reinforced by time, when n > 1
- Effects = concentration x time, when n = 1 (Haber rule)
- Elimination with time, when n < 1

Chemicals with time-dependent toxicity, such as neoncotinoids and some carcinogenics, require different methods of risk assessment, for these toxicants have no safe levels – time determines their effects.