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Ecological Risks of Pesticides

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2013 One Medicine Symposium

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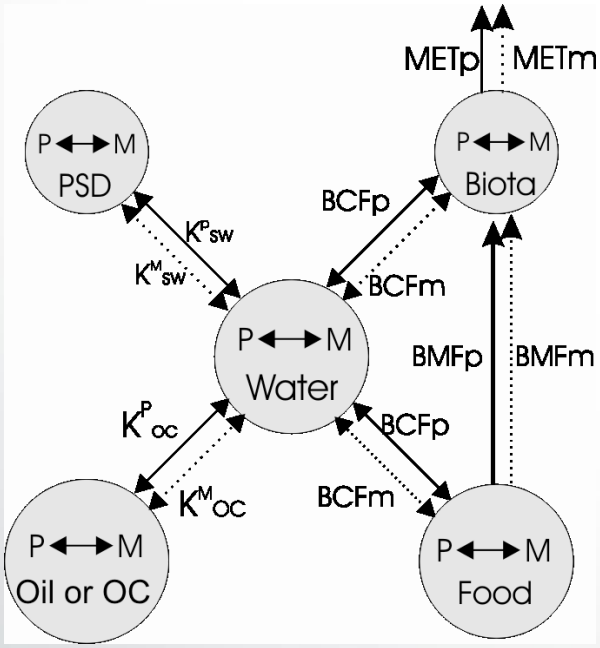
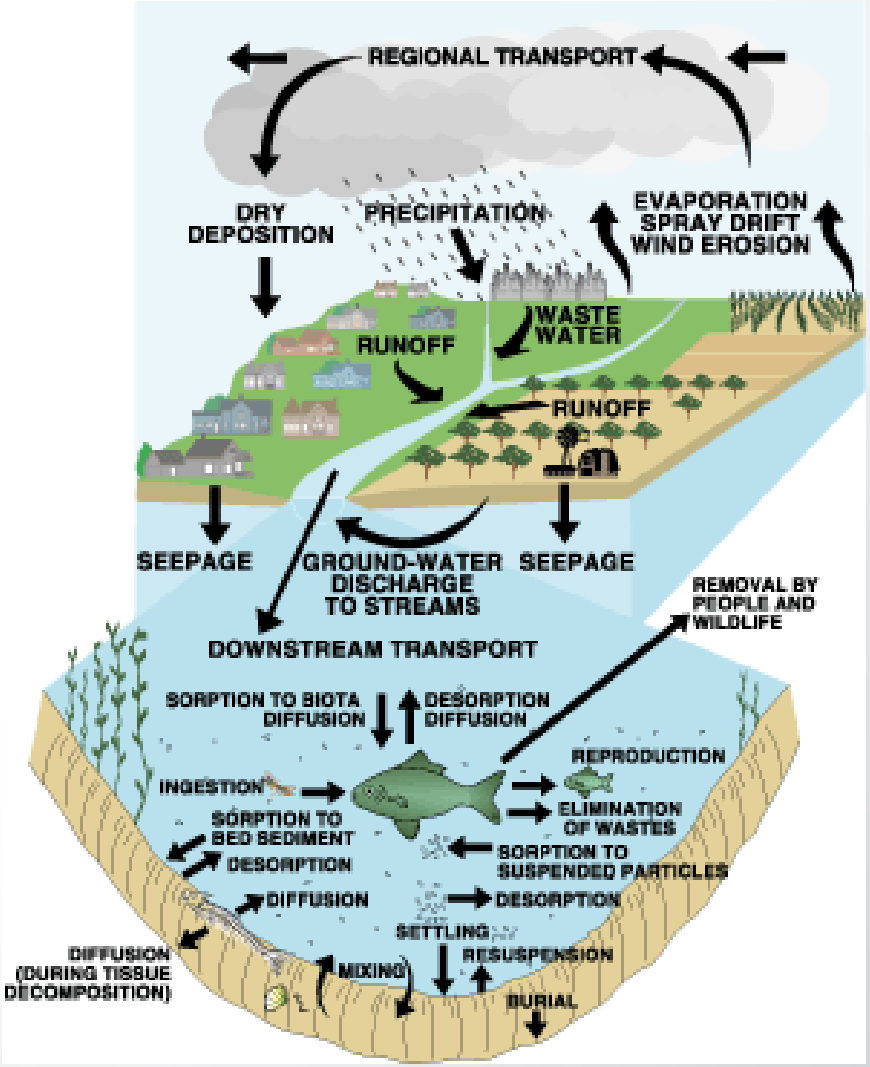
Outline of Presentation

- Overview of pesticide use and potential for adverse effects on ecosystems
- Case studies:
  - Very brief summary of DDT
  - Pyrethroid insecticides
  - Is atrazine causing frog deformities in nature?

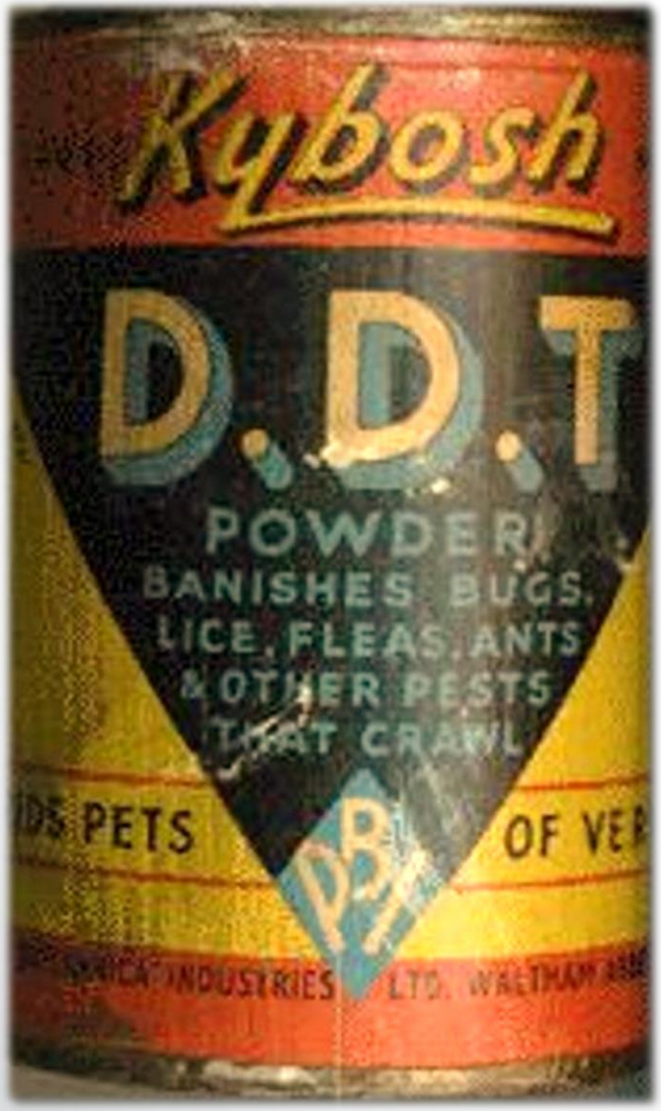
Sources and Fate of Pesticides

Risk is a function of toxicity and exposure to the bioavailable fraction of the chemical



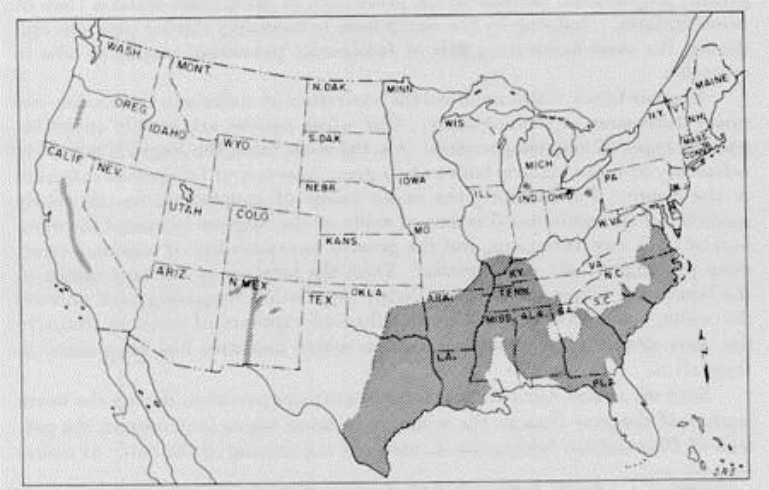


DDT

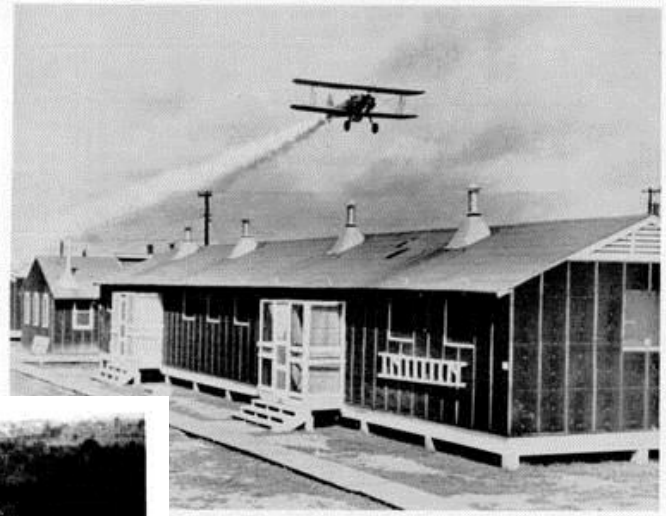

Kybosh
D.D.T
POWDER
BANISHES BUGS,
LICE, FLEAS, ANTS
& OTHER PESTS
THAT CRAWL
PETS
OF VE
INDUSTRIES
LTD. WALTHAM

D.D.T.
Powerful Insecticide
Harmless to Humans
TODD

DDT:
Rural Areas of US



MAP 4.—Areas of the continental United States believed to be malarious in 1934–35.

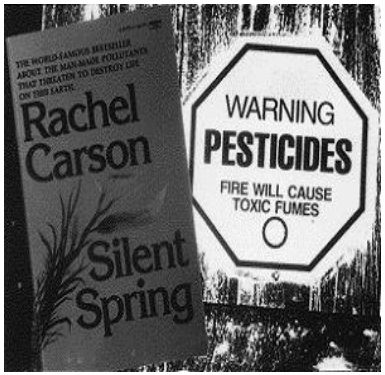


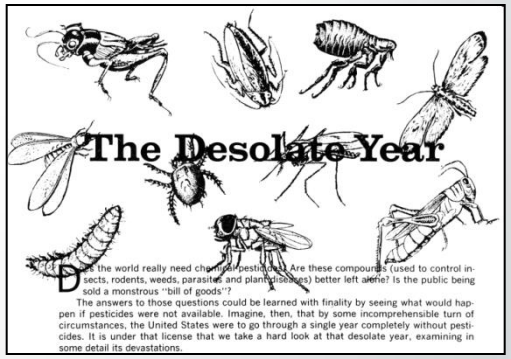
biplane applying DDT to control anophelines over Stuttgart, Ark., Army Air Base.

FIGURE 20.—Airplane application of larvicide over a waterchestnut infested area of the Potomac River.

DDT: What Happened?

- Rachel Carson's *Silent Spring* (1962)
- Peregrine Falcons near extinction in Eastern U.S.
  - Reproductive success damaged by eggshell thinning
- Monsanto's "The Desolate Year" (1962)




The Desolate Year

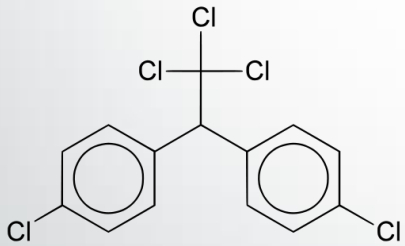
Does the world really need chemical pesticides? Are these compounds (used to control insects, rodents, weeds, parasites and plant diseases) better left alone? Is the public being sold a monstrous "bill of goods"?

The answers to those questions could be learned with finality by seeing what would happen if pesticides were not available. Imagine, then, that by some incomprehensible turn of circumstances, the United States were to go through a single year completely without pesticides. It is under that license that we take a hard look at that desolate year, examining in some detail its devastations.

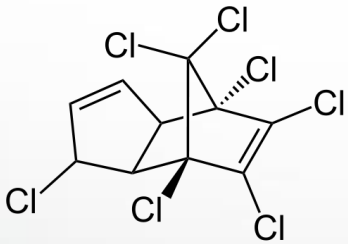
OC Insecticides: Physical Chemical Properties

Chemical	MW	Solubility (ug/L)	Log K_{ow}	VP (mm Hg)	K_H (atm-m^3/mol)	$T_{1/2}$ (soil - years)
DDT	354.5	25	**6.91**	1.6 e-7	8.3 e-6	**2 – 15**
Heptachlor	373.3	50	6.10	3.0 e-4	2.9 e-4	0.5 – 3.5
Dieldrin	380.9	110	6.20	5.9 e-6	5.2 e-6	2.5
Lindane	290.8	7300	3.80	4.5 e-5	6.6 e-6	0.1 – 0.5

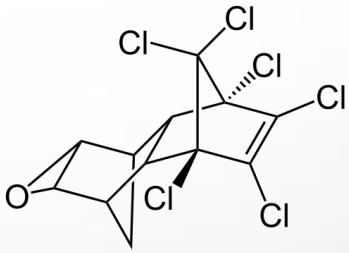
From ATSDR



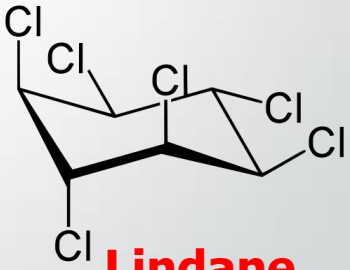
DDT



Heptachlor



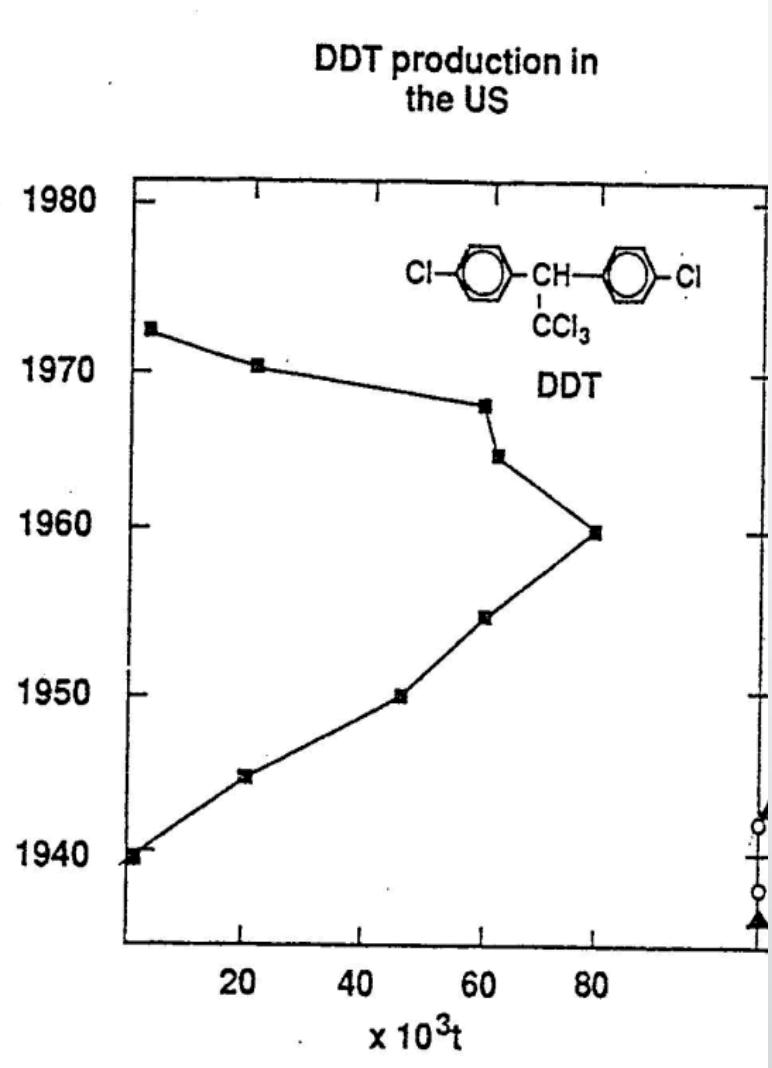
Dieldrin



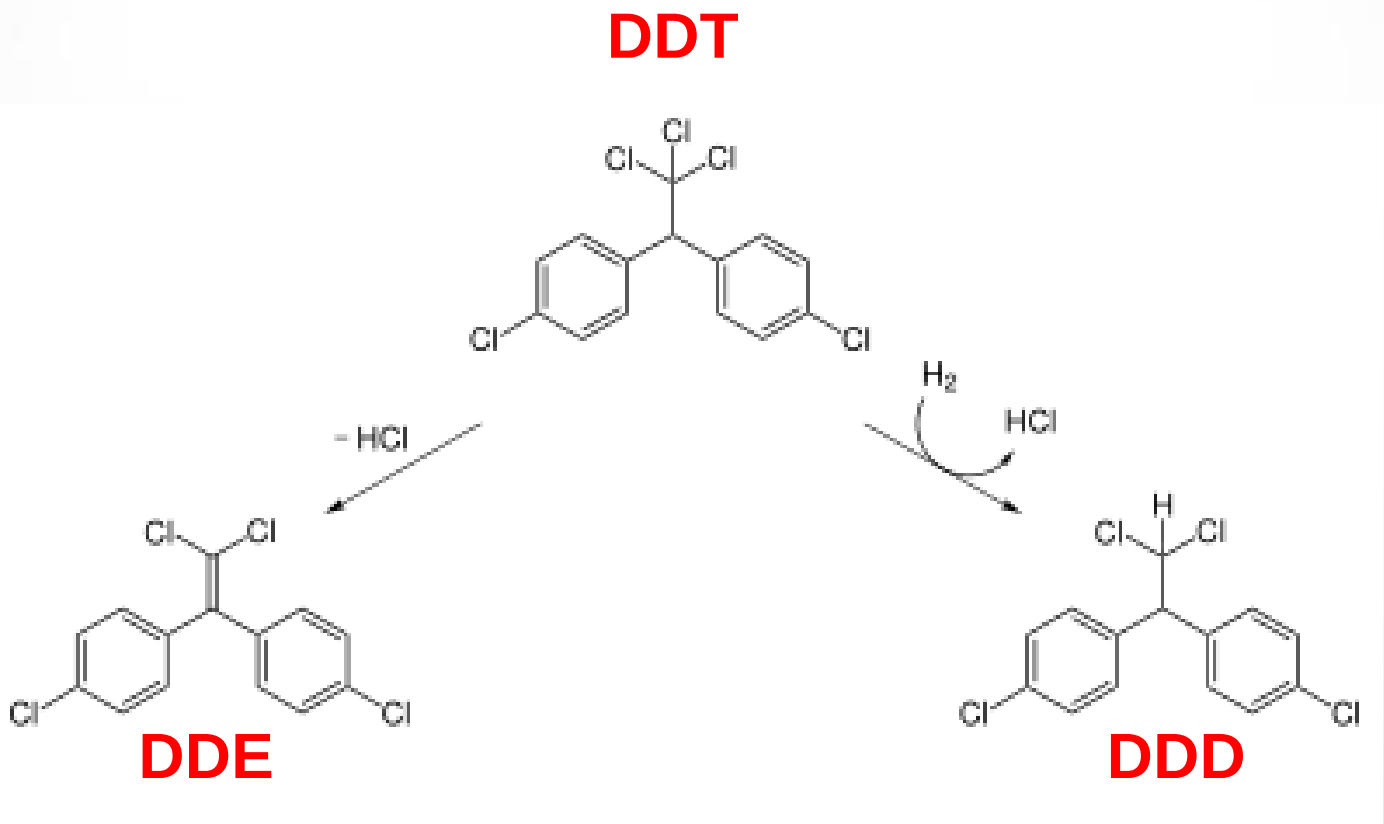
Lindane

DDT Timeline

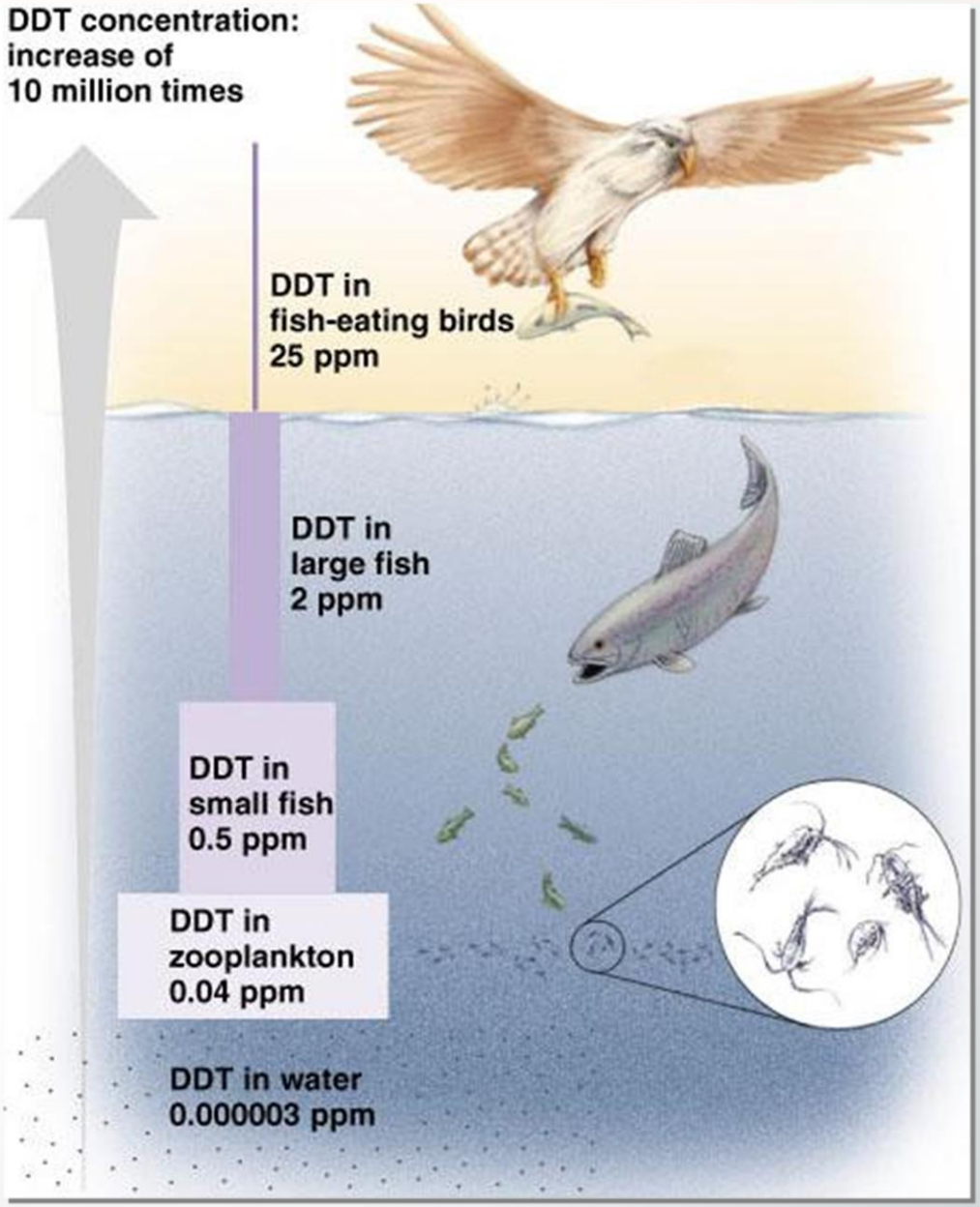
- Discovered in 1939
- U.S. used in WWII
  - Control vectors of malaria and typhus
- 1945 - Agricultural pesticide for 30 years
  - 1.3 billion pounds used
- 1959 – use declined greatly
- 1972 – banned in US
- Still used in Africa & Asia



Degradation Pathway



Biomagnification



DDT and DDE are readily stored in fats of vertebrates

Toxic Effects

- Insects
  - Death
- Fish
  - Reproductive effects
  - Neurotoxin effects
- Birds
  - Eggshell thinning
- Mammals
  - Localized liver and kidney damage
  - Neurotoxin effects
  - Teratogenic effects (mice)
  - Carcinogenic effects are likely

Molecular Interactions

- How it kills insects
  - Na^+ ion channels stay open, resulting in random neuron fire and death “DDT Jitters”
  - Those with a mutation or up-regulated P-450 genes are resistant
- DDE mimics the action of estradiol causing a decline in androgen synthesis in some vertebrates
- DDE inhibits the Ca^{2+} -ATPase or Ca^{2+}-Mg^{2+} ATPase activity in the eggshell gland

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SCIENTIFIC AMERICAN™

Banned Pesticide DDT Is Still Killing California Condors

John R. Platt

September 20, 2013

The Mystery of the Vanishing DDT in the Ocean Near Los Angeles

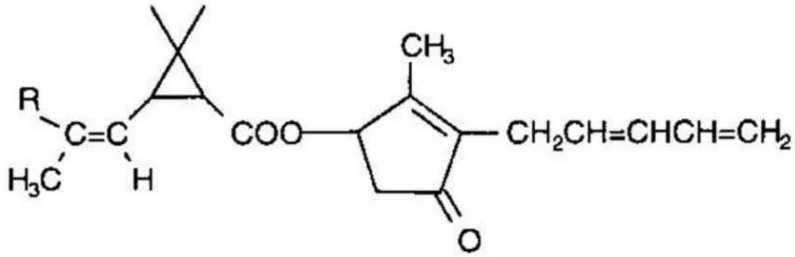
A deposit of 110 tons of the banned pesticide has rapidly shrunk on the ocean floor and cast doubt on the need to mount an expensive cleanup

Maria Cone, March 13, 2013

Pyrethroids

Pyrethrins

- Pyrethrins (6 acids and esters)
  - extracted from chrysanthemums
- Low mammalian toxicity
- Nonpersistent, degraded by heat/light

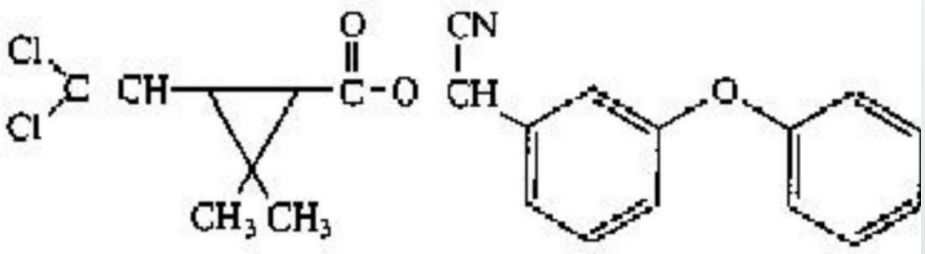


Pyrethrin I (R=CH_3) LD50 800–1500
Pyrethrin II (R=$COOCH_3$)

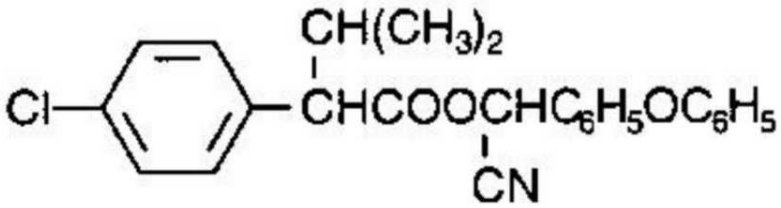
Pyrethroid Insecticides

- Designed from pyrethrin structure for added photostability
- Two basic structures
  - Cyclopropane ring
    - cypermethrin
  - No cyclopropane ring
    - fenvalerate

CYPERMETHRIN



FENVALERATE



LD50 450

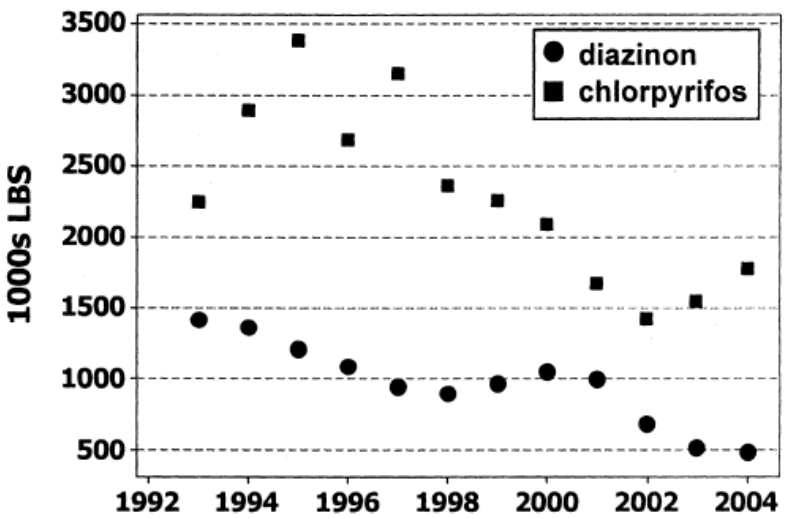
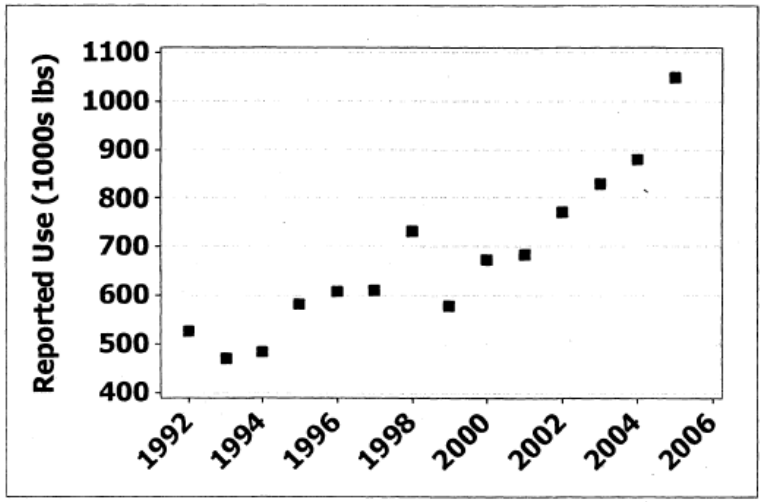


Figure 1. Overall trend in California reported use (16) of diazinon and chlorpyrifos. Includes agricultural, commercial structural, and landscape maintenance applications.

Use of OP insecticides has decreased markedly since the mid-1990s for two main reasons:

1. **The USEPA agreements with registrants to phase-out all residential uses in the early 2000's**
1. **Widespread recognition of OP surface water quality problems**

One consequence of decreased OP use has been a corresponding increase in use of synthetic pyrethroids



2005 TOP 7	-	lbs
permethrin	-	550k
cypermethrin	-	208k
bifenthrin	-	63k
cyfluthrin	-	50k
fenpropathrin	-	40k
λ-cyhalothrin	-	37k
esfenvalerate	-	33k

Figure 2. Overall trend in California reported use of 25 synthetic pyrethroids (20). Includes agricultural, commercial structura,l and landscape maintenance applications.

Source: In Synethetic Pyrethroids; Gan, J., et al; ACS Symposium Series; American Chemical Society: Washington DC, 2008

Advantages of Pyrethroids

- Highly lipophilic (hydrophobic)

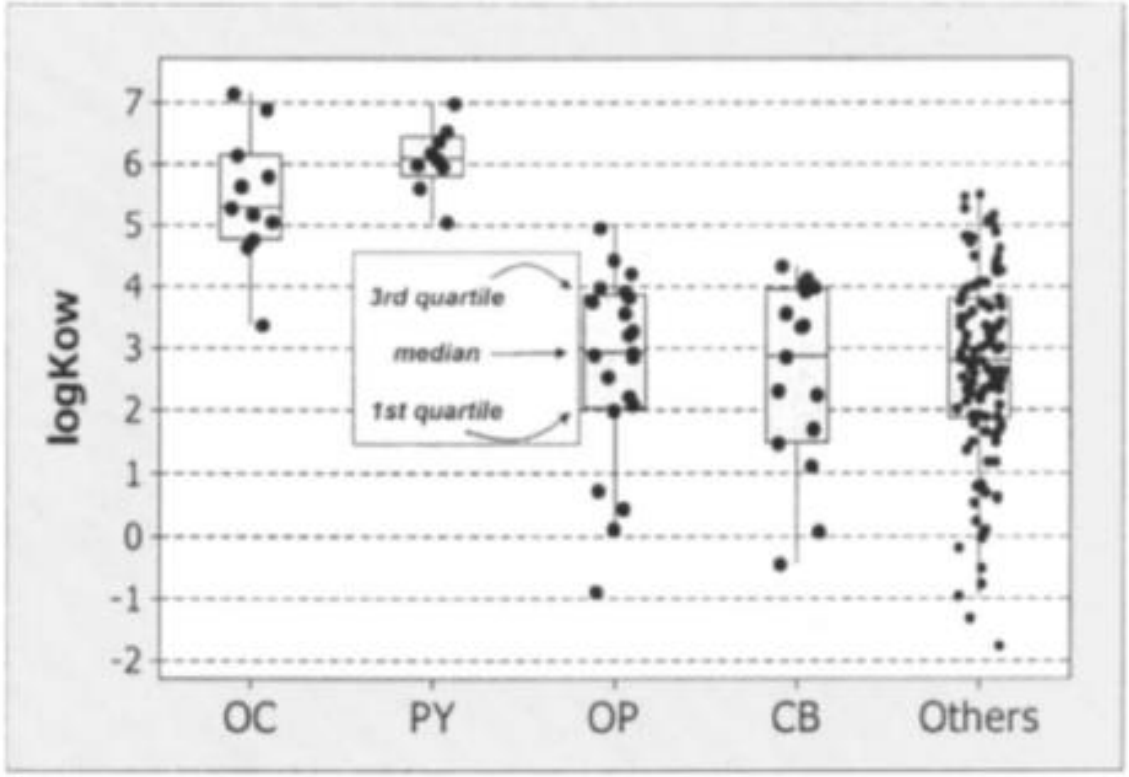


Figure 4. Comparison of log octanol/water partition coefficients for organochlorines (OC), pyrethroids (PY), organophosphates (OP), carbamates (CB) and other miscellaneous pesticides.

Source: In Synethetic Pyrethroids; Gan, J., et al; ACS Symposium Series; American Chemical Society: Washington DC, 2008

Advantages of Pyrethroids

- Short half-life in the environment

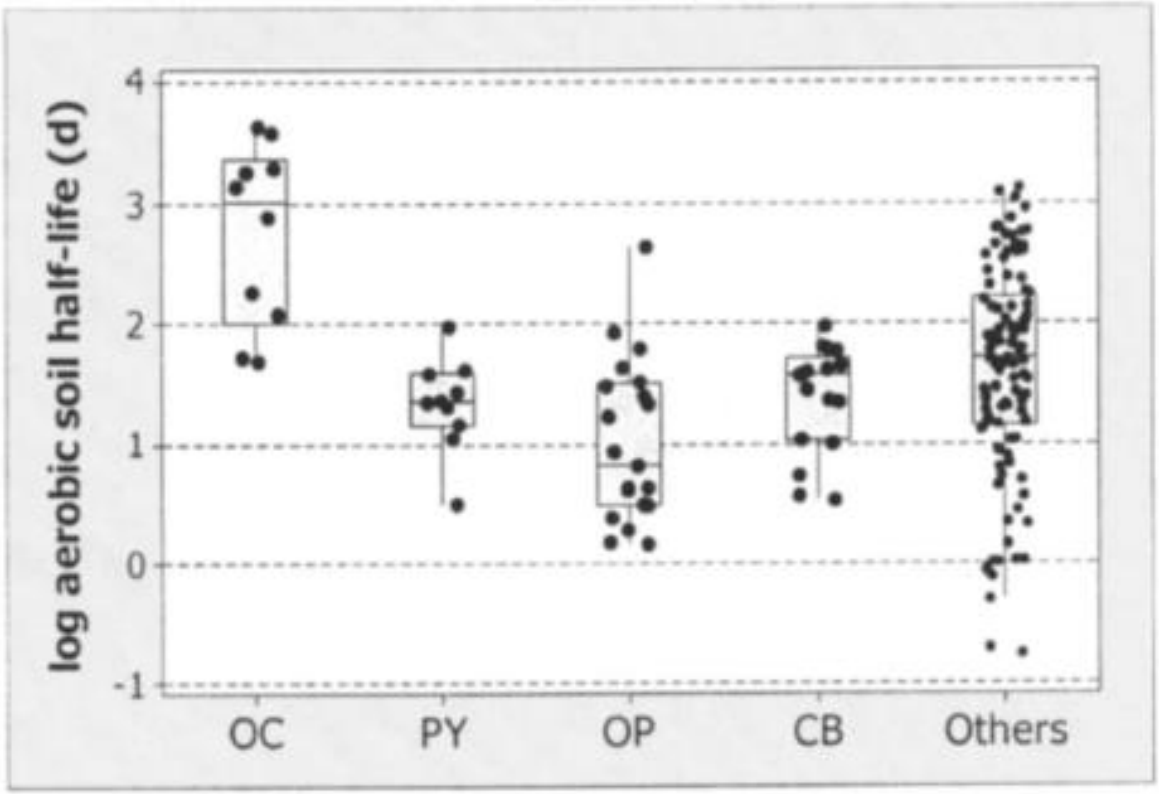


Figure 5. Comparison of laboratory aerobic soil half-lives for organochlorines (OC), pyrethroids (PY), organophosphates (OP), carbamates (CB) and other miscellaneous pesticides.

Source: In Synethetic Pyrethroids; Gan, J., et al; ACS Symposium Series; American Chemical Society: Washington DC, 2008

Advantages of Pyrethroids

- Low toxicity to terrestrial vertebrates

Table 3.2 LC_{50} values of pyrethrins, type I (allethrins, permethrin and resmethrin), II (cypermethrin and deltamethrin) and pseudopyrethroid (etofenprox) for mallard duck (*Anas platyrhynchos*), rat (*Rattus norvegicus*) and rainbow trout (*Oncorhynchus mykiss*).

Compound	*Mallard Duck*[a]	*Rat*[a]	*Rainbow Trout*[b]	*Source*
Pyrethrins	> 5620	700	5.1	USEPA[118]
Allethrins	> 2000	720	9.7	WHO[222]
Permethrin	> 10000	8900	6.43	USEPA[10]; Kumaraguru and Beamish[223]
Resmethrin	> 5000	4639	0.28	USEPA[68]
Cypermethrin	> 2634	247	0.39	USEPA[10]
Deltamethrin	> 4640	128	1.97	WHO[224]
Etofenprox	> 2000	> 5000	13	USEPA[25]

[a]Acute oral LC_{50} ($mg\,kg^{-1}$).
[b]96-h LC_{50} ($\mu g\,l^{-1}$).

Source: Schleirer, J. and Peterson, R.; Pyrethrins and Pyrethroid Insecticides; Green Trends in Insect Control; Royal Society of Chemistry, 2011

Advantages of Pyrethroids

- Do not biomagnify like older chemical classes

Table 3.1 Bioconcentration factors (BCF) for type I and II pyrethroids and DDT for rainbow trout (*Oncorhynchus mykiss*) from Muir *et al.*[221]

Compound	*BCF*
Cypermethrin	832
Permethrin	1940
Deltamethrin	502
Fenvalerate	403
DDT	72500

Source: Schleirer, J. and Peterson, R.; Pyrethrins and Pyrethroid Insecticides; Green Trends in Insect Control; Royal Society of Chemistry, 2011

Potential Concerns?

- Toxicity to aquatic invertebrates

96-h median lethal concentration values for permethrin obtained from the USEPA's Ecotox database					
Species	**Common name**	**LC50 (ug/L)**	**Species**	**Common name**	**LC50 (ug/L)**
Menippe mercenaria	Florida stone crab	0.018	Uca pugilator	Sand fiddler crab	2.39
Hyalella azteca		**0.021**	Etheostoma lepidum	Greenthroat darter	2.71
Palaemonetes pugio	Grass shrimp	0.05	Chironomus riparius	Harlequin fly	2.89
Chironomus dilutes		0.059	Oncorhynchus mykiss	Rainbow trout	2.9
Americamysis bahia	Mysid shrimp	0.075	Pimephales promelas	Fathead minnow	3
Crangon septemspinosa	Sand shrimp	0.13	Etheostoma fonticola	Fountain darter	3.34
Gammarus pseudolimnaeus		0.17	Notropis mekistocholas	Cape Fear shiner	4.16
Penaeus duorarum	Pink shrimp	0.22	Xyrauchen texanus	Razorback sucker	5.95
Procambarus clarkii	Freshwater crayfish	0.28	Ictalurus punctatus	Channel catfish	7.2
Daphnia magna	Daphnia magna	0.3	Micropterus salmoides	Largemouth bass	8.5
Penaeus aztecus	Brown shrimp	0.34	Micropterus sp.	Black bass	8.5
Gammarus pulex	Freshwater amphipod	0.44	Chironomus tentans	Chironomus tentans	10.45
Ceriodaphnia dubia		0.57	Gambusia affinis	Mosquitofish	12
Nitocra spinipes	Copepod	0.6	Lepomis macrochirus	Bluegill	13
Homarus americanus	American lobster	0.73	Cyprinodon variegates	Sheepshead minnow	17
Salmo salar	Atlantic salmon	1.5	Cyprinodon bovines	Leon spring pupfish	21
Oncorhynchus clarki ssp. Henshawi	Lahonton cutthroat trout	1.58	Ptychocheilus lucius	Colorado pikeminnow	24
Erimonax monachus	Spotfin chub	1.7	Atherinops affinis	Topsmelt	25.3
Oncorhynchus gilae ssp. Apache	Apache trout	1.71	Menidia beryllina	Inland silverside	27.5
Salvelinus fontinalis	Brook trout	2.3			

Potential Concerns?

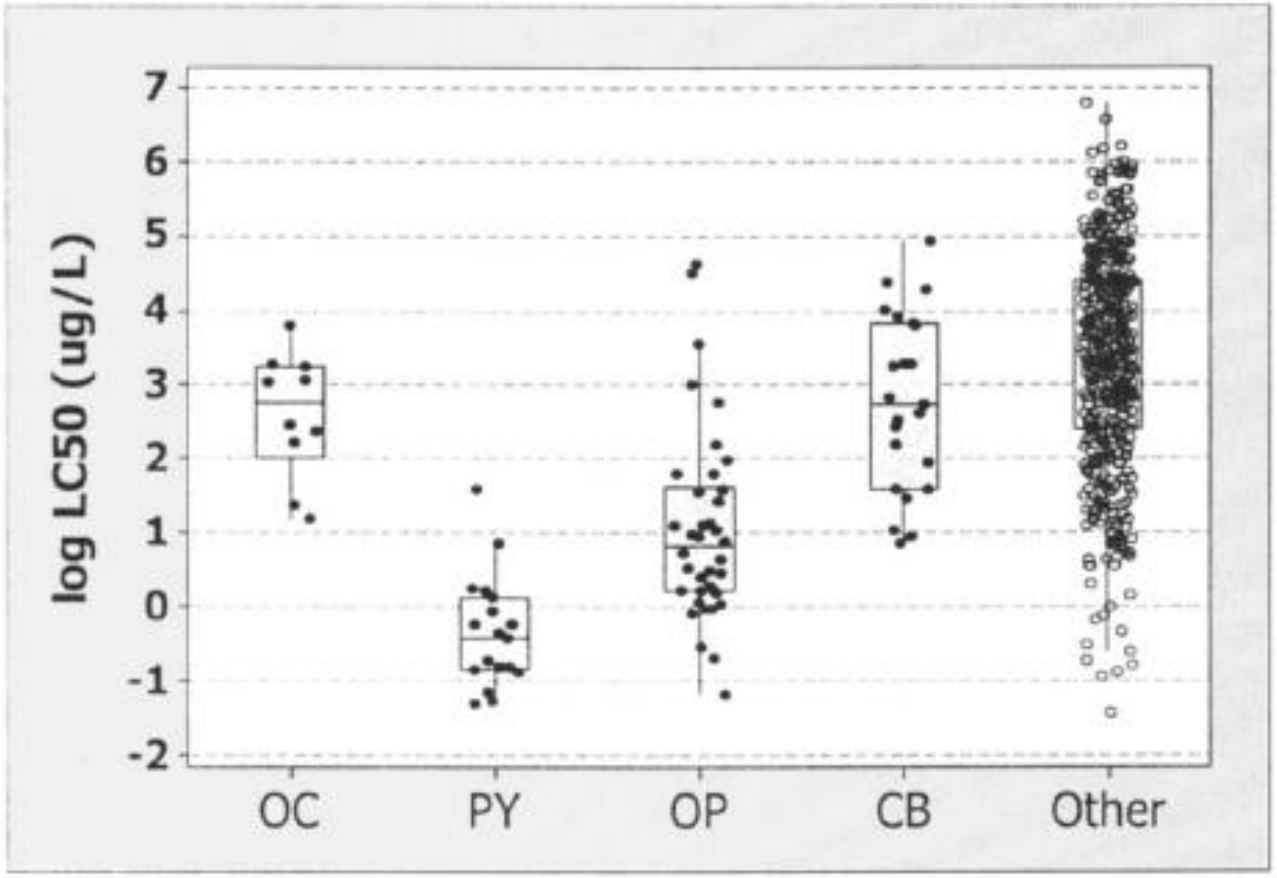


Figure 6. Comparison of Dapnia magna median lethal concentrations (LC50) for organochlorines (OC), pyrethroids (PY), organophosphates (OP), carbamates (CB, and other miscellaneous pesticides. Data from laboratory aqueous studies.

Source: In Synethetic Pyrethroids; Gan, J., et al; ACS Symposium Series; American Chemical Society: Washington DC, 2008

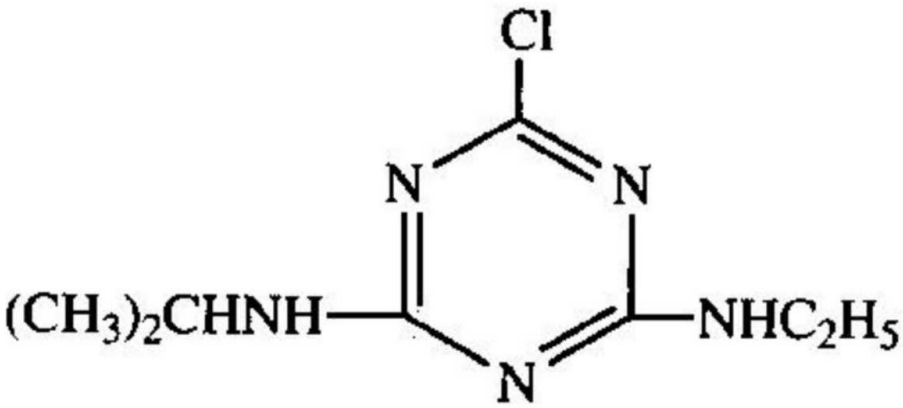
Potential Concerns?

- Pyrethroids are hydrophobic (log K_{ow} ~6.0) so primary concern is with benthic invertebrates
- High toxicity seen in laboratory sediment toxicity testing while lower toxicity observed in the streams with measured pyrethroid concentrations
- Natural populations seem much less affected
- Are pyrethroids bioavailable in sediment?
- Are we using right test organism?
- This is an area of ongoing research

Atrazine

Atrazine

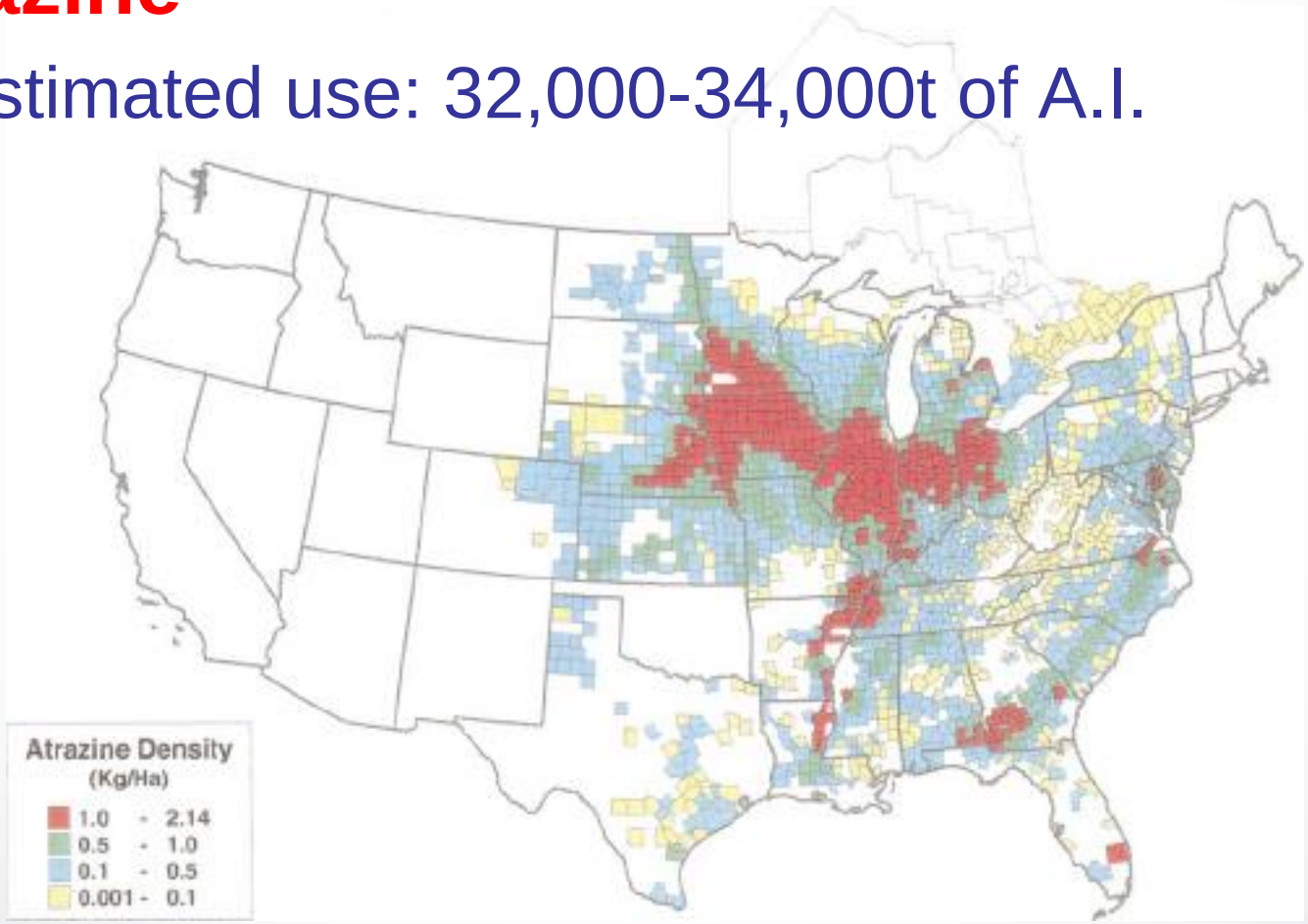
- Triazine herbicide
- One of the most used pesticides in the U.S.
- Used to control annual broadleaf and grass weeds, primarily in corn
- 1959: registered for commercial use



Atrazine

Atrazine

- Estimated use: 32,000-34,000t of A.I.



Atrazine usage on corn per county, 1985–1988.

Source: Solomon, K. et al; Ecological Risk Assessment of Atrazine in North American Surface Waters. ET&C. Vol. 15, No. 1, pp. 31–76, 1996

Atrazine: Physical & Chemical Properties

- Water solubility: 33 ug/mL at 22°C
- Vapor Pressure: 2.89×10^{-7} mmHg at 25°C
- Henry's Law constant: 2.48×10^{-9} atm m^3 mol^{-1}
- **Log K_{ow}: 2.68** at 25°C
- K_d: 0.19 to 2.46
- K_{oc}: 25 to 155
- Hydrolysis: stable for 30d at pH 5-9 at 25°C
- Aq photolysis: Natural light – $t_{1/2}$ 335d at pH 7

Source: Solomon, K. et al; Ecological Risk Assessment of Atrazine in North American Surface Waters. ET&C. Vol. 15, No. 1, pp. 31–76, 1996

Toxicity of Atrazine

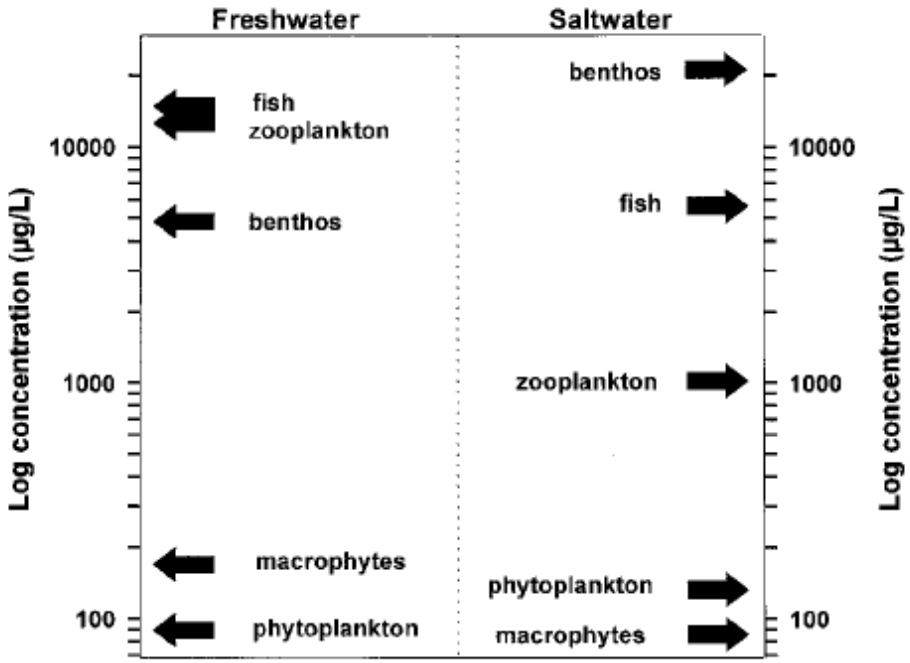


Fig. 16. Geometric means for acute atrazine toxicity data in freshwater and saltwater organisms.

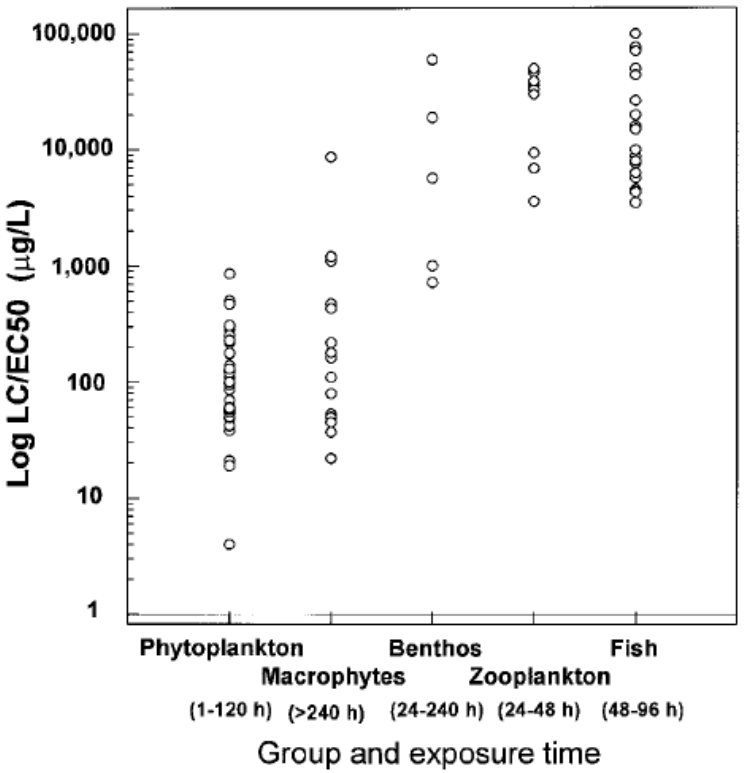


Fig. 17. Range of acute toxicity data for a number of groups of freshwater organisms.

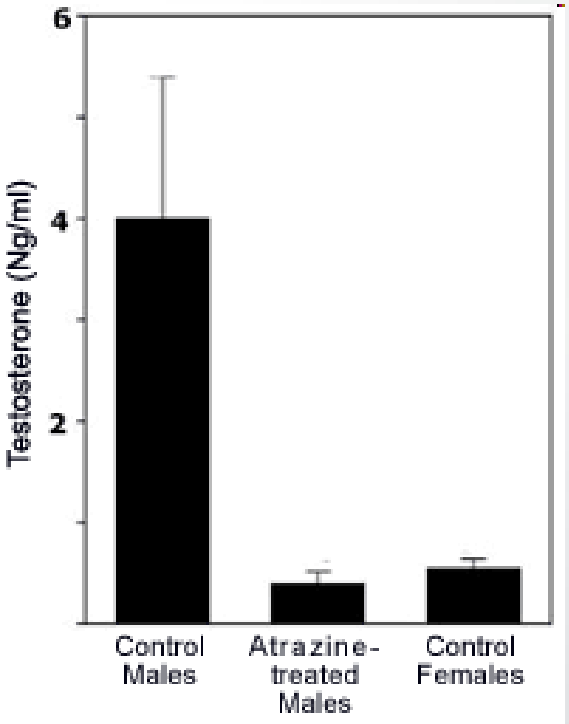
Source: Solomon, K. et al; Ecological Risk Assessment of Atrazine in North American Surface Waters. ET&C. Vol. 15, No. 1, pp. 31–76, 1996

Atrazine and Amphibians

In 2002, Tyrone Hayes, a scientist at UC Berkeley, found evidence that atrazine is a tetratogen, causing demasculinization in male frogs even at low concentrations, and an estrogen disruptor.

Male frogs affected by atrazine had testosterone levels below females.

In 2003, Dr. Hayes found that atrazine induced hermaphroditism at 0.1 ppb in American leopard frogs.



ScienceDaily

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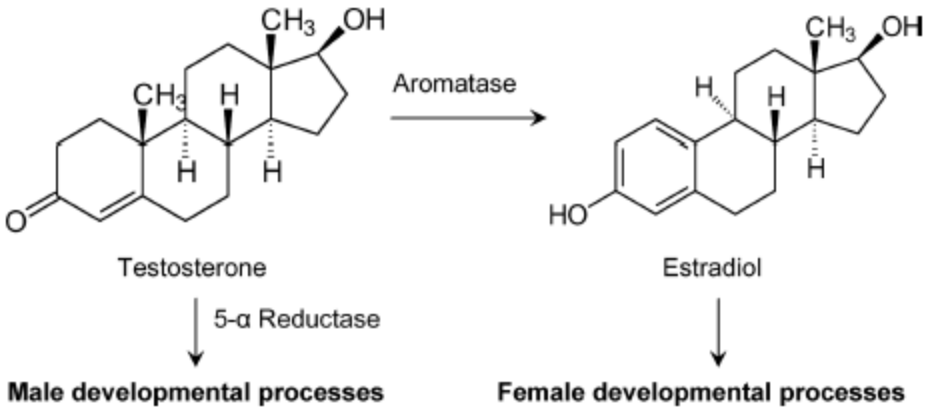
Pesticide Atrazine Can Turn Male Frogs Into Females

Atrazine and Amphibians

1. Estrogen-mediated mechanisms?
2. Adverse effects on gonadal development in amphibians?
3. Adverse effects at the population level in exposed amphibians?

Estrogen-mediated mechanisms?

- Hayes and coworkers have hypothesized that upregulation of *CYP19* is a cause for adverse effects in frogs (2002), however they have never reported any measurements of *CYP19* mRNA or aromatase enzyme activity.
- However, when others have measured both *CYP19* mRNA expression and aromatase activity, no effects of atrazine have ever been observed



Estrogen-mediated mechanisms?

A study with *P. promelas* showed that exposure of adults to measured concentrations 25 and 224 μg atrazine/L for 21 d had no effect on a range of parameters in females, including body weight, gonadal somatic index (ratio of gonad mass to body mass), stage of ovarian development, proportion of atretic follicles or postovulatory follicles, number of eggs produced, or the number of eggs hatched (U.S. EPA, 2005).

Estrogen-mediated mechanisms?

- A study on frogs exposed to atrazine under field conditions in South Africa, where *X. laevis* is native, found no effects on aromatase activity (Hecker et al., 2004)
- Neither did a similar field study on *R. clamitans* in Michigan (Murphy et al., 2006a)
- These studies included concentrations in excess of the 0.1 μg/L that has been suggested by Hayes et al. (2002) as the threshold above which this effect causes adverse effects in frogs.

Adverse effects on gonadal development in amphibians?

- Laboratory studies on *R. clamitans* exposed to atrazine at concentrations as great as 25 *µ*g/L (Coady et al., 2004) and *X. laevis* at concentrations as great as 100 *µ*g/L did not reveal any effects on sex ratio (Coady et al., 2005; Kloas et al., 2008).
- Studies in wild populations of *X. laevis* in areas of corn production and atrazine use and in reference areas did not reveal effects on sex ratios of adults or metamorphs (Du Preez et al., 2005b
- No differences in the absolute or relative numbers of testicular cell types were observed in *X. laevis* from corn and non-corn growing areas in South Africa where atrazine concentrations ranged from 0 to 9 *µ*g/L (Smith et al., 2005).
- In a study of *R. pipiens* metamorphs exposed to 15 *µ*g atrazine/L throughout metamorphosis, Orton et al. (2006) reported no difference in the total number of spermatogenic cells.

Adverse effects on gonadal development in amphibians?

- The first study to report on sex reversal in genetic males stated that atrazine concentrations ranging from 0.1 to200 *µ*g atrazine/L produced gonadal abnormalities in developing *X. laevis* exposed from hatching until completion of metamorphosis (Hayes et al., 2002).
- The authors reported that 16–20% of the exposed animals had multiple gonads or were“hermaphrodites” with multiple testes and ovaries
- However, the incidence of gonadal abnormalities at each test concentration was not reported, making it impossible to determine if these effects were concentration-related
- A concentration-related increase in the incidence of segmented and anomalous gonads was observed in *X. laevis* exposed to atrazine from <24 h after fertilization for 70 d (Carr et al., 2003). The incidence of abnormal gonads based on gross morphology and categorized as intersex individuals differed significantly from controls only in animals exposed to 25*µ*g atrazine/L.
- However, subsequent histological evaluation of the gonads from the 25- *µ*g/L group, by pathologists from EPL Laboratories (Wolf, 2007), confirmed that gonads identified as intersex based upon their outward appearance showed no evidence of mixed ovarian and testicular tissue.

Adverse effects on populations in exposed amphibians?

- There has been a loss of diverse amphibian populations from several continents. (U.S. Fish and Wildlife Service, 1984; Fellers and Drost, 1993, Ingram and McDonald, 1993; Hines et al., 1999; Houlahan et al., 2000; Stuart et al., 2004; Mendelson et al., 2006)
- No published population studies to suggest that atrazine is associated with declines in populations of amphibians.
- Limited data on the potential relationship between atrazine and amphibian population dynamics
  - Amphibians inhabiting ponds on agricultural land in Minnesota and exposed to atrazine (0.1–0.5 μg/L) and de-ethyl atrazine (0.1– 0.3 μg/L) concentrations 5-fold greater than those reported to produce gonadal effects (Hayes et al., 2003) exhibited no differences in species richness or reproductive success (Knutson et al., 2004).

Conclusions

- Majority of observations do **NOT** support the theory that environmentally relevant concentrations of Atrazine affect reproductions and/or reproductive development in amphibians
- They also do **NOT** support the theory that atrazine induces aromatase, the enzyme that converts testosterone to estradiol.
- Environmentally relevant concentrations of Atrazine do not affect amphibian growth, sexual development, reproduction, and survival. And although few data are available, the same conclusions apply to fish and reptiles
- **In 2006 the U.S. EPA considered re-registration of Atrazine final when it issued a cumulative risk assessments on the triazine herbicides, and concluded that they posed "no harm that would result to the general U.S. population, infants, children or other...fish and wildlife…consumers."**

Critical Reviews in Toxicology, 38:721–772, 2008

Effects of Atrazine on Fish, Amphibians, and Aquatic Reptiles: A Critical Review

"A number of reports on the effects of atrazine on aquatic vertebrates, mostly amphibians, have been published, yet there is inconsistency in the effects reported, and inconsistency between studies in different laboratories. Based on a weight of evidence analysis of all of the data, the central theory that environmentally relevant concentrations of atrazine affect reproduction and/or reproductive development in fish, amphibians, and reptiles is not supported by the vast majority of observations. The same conclusions also hold for the supporting theories such as induction of aromatase, the enzyme that converts testosterone to estradiol. For other responses, such as immune function, stress endocrinology, parasitism, or population-level effects, there are no indications of effects or there is such a paucity of good data that definitive conclusions cannot be made."

Summary

- Many pesticides used prior to 1970s (e.g., DDT) were
  - Very persistent in soil, water, air, and biota
  - Very hydrophobic so they bioaccumulated in food chain
  - Moderately to highly toxic (incl. some metabolites, DDE)
  - Resulted in significant ecological harm that persists in some areas
- The evolution of pesticides since then has seen
  - Less persistence in soil, water, air, and biota
  - Less hydrophobic – they accumulate much less in food chain
  - Generally much less toxic, with some exceptions
  - Resulting in much less ecological harm
- Although some problems and uncertainty remain, this is largely a "good news" story where we have learned our lesson regarding pesticide impacts on ecosystems

Twenty tips for interpreting scientific claims

WJ Sutherland, D Spiegelhalter & M Burgman

***Nature* 20 November 2013**

This list will help non-scientists (and scientists too!) to better grasp the limitations of evidence and critically evaluate scientific claims