Environmental Science: Oxford Research Encyclopedias

Pesticides and Human Health

Pierluigi Cocco

Subject: Environment and Human Health Online Publication Date: Aug 2016
DOI: 10.1093/acrefore/9780199389414.013.82

Summary and Keywords

The fight against agricultural and household pests accompanies the history of humanity, and a total ban on the use of pesticides seems unlikely to happen in the foreseeable future. Currently, about 100,000 different chemicals, inorganic and organic, are currently in the market, grouped according to their function as insecticides, herbicides, fungicides, fumigants, rodenticides, fertilizers, growth regulators, etc. against specific pests, such as snails or human parasites, or their chemical structure—organochlorines, organophosphates, pyrethroids, carbamates, dithiocarbamates, organotin compounds, phthalimides, phenoxy acids, heterocyclic azole compounds, coumarins, etc. Runoff from agricultural land and rain precipitation and dry deposition from the atmosphere can extend exposure to the general environment through the transport of pesticides to streams and ground-water. Also, the prolonged bio-persistence of organochlorines generates their accumulation in the food chain, and their atmospheric drift toward remote geographical areas is mentioned as the cause of elevated fat contents in Arctic mammals. Current regulation in the developed world and the phasing out of more toxic pesticides have greatly reduced the frequency of acute intoxications, although less stringent regulations in the developing world contribute to a complex pattern of exposure circumstances worldwide. Nonetheless, evidence is growing about long-term health effects following high-level, long-lasting exposure to specific pesticides, including asthma and other allergic diseases, immunotoxicity, endocrine disruption, cancer, and central and peripheral nervous system effects. Major reasons for uncertainty in interpreting epidemiological findings of pesticide effects include the complex pattern of overlapping exposure due to multiple treatments applied to different crops and their frequent changes over time to overcome pest resistance. Further research will have to address specific agrochemicals with well-characterized exposure patterns.

Keywords: pesticides, environmental health, occupational health, asthma, immunotoxicity, endocrine disruption, cancer, nervous system disorders

date: 29 September 2016

Introduction

> . . . Old woman, bring some sulfur, and make a fire,
>
> so I can purge the hall from this pollution . . .
>
> My child, . . . let me bring you a tunic and cloak to wear.
>
> It would be wrong to stand there in your hall
>
> with your broad shoulders clothed in rags.
>
> Homer, *Odyssey*, Book 22, 611–627

Brief Historical Notes

Methods to fight agricultural and household pests were known and applied as long ago as 4500 BC. The *Ebers Papyrus*, written by Egyptian alchemists in 1550 BC, lists over 800 recipes as pesticides. Homer cites the use of sulfur as a disinfesting agent and the need for protective clothing when using it, even before the Chinese were aware of the insecticidal properties of the arsenicals. Pliny recommended using *marrubium vulgare* (white horehound) extract to treat agricultural pests, and Marcus Terentius Varro suggested using amurca, seeped from olive oil presses, to protect seeds against ants, moles, and weeds (Smith & Secoy, 1975; Taylor, Holley, & Kirk, 2007). However, two major events in Europe pushed toward the discovery of effective pest control agents: the destruction of potato crops by late blight in Ireland in 1845–1848, followed by a famine that killed millions, and the destruction of the French vineyards caused by downy mildew, imported from America in 1878, that subsequently spread all over Europe. In 1882, Pierre-Marie-Alexis Millardet, professor of botanics at the University of Bordeaux, made the serendipitous discovery of the fungicide properties of sulfur and copper and created the mixture known as the “Bordeaux Mixture,” still used to treat mildew in vineyards and other crops (Fishel, 2013; Taylor et al., 2007).

In 1873, Othmar Zeidler, an Austrian chemist, synthesized DDT (dichlorodiphenyltrichloroethane). Its insecticide properties were discovered much later by the Swiss chemist, Paul Hermann Müller, who was awarded the Nobel Prize for Physiology and Medicine in 1948 for the effectiveness of DDT in fighting typhus and vector-borne diseases. In subsequent years, the development of resistance against DDT and the progress of the chemical industry prompted further research into new weapons

date: 29 September 2016

against insects as well as weeds. The carbamate and organophosphate pesticides and the phenoxy acid herbicides appeared on the market, and their use became widespread worldwide. As a result, crop production could be substantially increased, improved, and better preserved in a cheap and effective fashion.

Definition

According to the U.S. Environmental Protection Agency (US EPA) definition, a pesticide is "any substance or mixture of substances intended for preventing, destroying, repelling, regulating, or controlling pests" (U.S. EPA, 1975), which include any unwanted organisms, from insects to rodents to weeds, that affect crops, livestock, foodstuffs, or human health, or damage any human activity or man-made infrastructure. The definition of pesticide covers about 100,000 different chemicals, inorganic and organic, that are grouped according to their function (e.g., insecticides, herbicides, fungicides, fumigants, rodenticides, fertilizers, growth regulators, etc. against specific pests such as snails or human parasites) or their chemical structure (organochlorines, organophosphates, pyrethroids, carbamates, dithiocarbamates, organotin compounds, phthalimides, phenoxy acids, heterocyclic azole compounds, coumarins, etc.). Using chemical agents with different structures is important to prevent insects and fungi from developing resistance, which is the reason why different treatment protocols are applied to different crops, or to the same crop in different seasons, or in different parts of the world. Along with the multiple crops and/or livestock to be treated each season on each farm, the strategy of changing patterns contributes to making it particularly difficult to link nondeterministic effects showing up after a long latency to any specific chemicals. Other conditions contributing to make the assessment of occupational exposure to pesticides especially complex include the multiple routes of absorption, such as inhalation during spraying, and dermal contact. Dermal absorption may occur because of carelessness with wearing personal protective equipment in greenhouses and outdoors, or in its proper dismissal/ disposal, and/or poor personal hygiene, and/or contact with the plants when re-entering too soon the treated area, not respecting the due re-entry time. Moreover, the exposure pattern is also complicated by the changing weather conditions in outdoor spraying. For these reasons, studies of pest control workers using specific chemicals for public health purposes, or agricultural extension agency workers taking accurate notes of treatments applied, are mostly informative.

The Pesticide Market

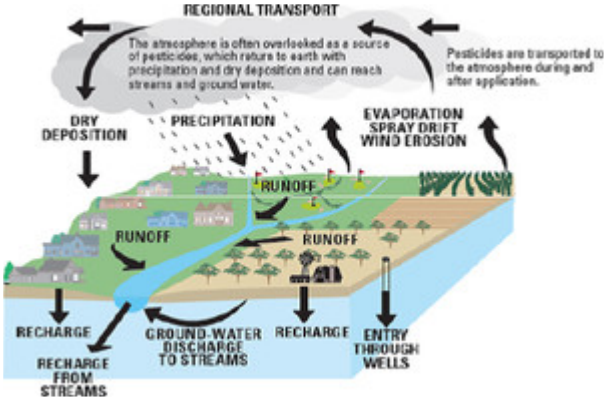
In the last decade, pesticide sales have been roughly stable worldwide with an overall budget of $40 billion, with the U.S. market accounting for 31.6% of the total (Grube,

Donaldson, Kiely, & Wu, 2011). In the last decade, the most significant increase in demand for pesticides has occurred in Central and South America (6.7% annual increase from 2004 to 2014), followed by the Asian market (4% annual increase from 2004 to 2014); the latter is the second largest after North America. Even the small African market, accounting for 3.5% of the global pesticide expenditure in 2004, has shown a sharp 6.4% annual increase in the same period. An annual increase has also been observed in Europe, although less pronounced. Overall and for agricultural uses, the herbicides predominate over the insecticides and fungicides, while household use of insecticides accounts for a larger share than agricultural use, equaling the amount of nonagricultural use of herbicides (Seedquest, 2016). Nonagricultural uses of pesticide include primarily weeds, insects, and other pest control in the household and its premises, parks and golf courses, as well as for public health applications (e.g., mosquito control) and weed control along roads, railroads, and in commercial and industrial areas (Grube et al., 2011). From 2001 to 2007, the two best-selling active ingredients in the U.S. agricultural market were the herbicides glyphosate and atrazine, with another four herbicides and four fumigants (including methyl bromide), ranking among the first 10 active ingredients; chlorpyrifos, the first insecticide, ranked 14 (Grube et al., 2011).

Pesticides in the Environment

Pesticides spread to the environment from specific points of release, such as manufacturing plants, mixing-and-loading facilities, spills, sewage treatment plants, and wastewater and solid waste disposal sites, and from diffuse nonpoint sources, including runoff from agricultural and urban land. Runoff from agricultural and urban land, and rain precipitation and dry deposition from the atmosphere, can transport pesticides to streams and groundwater (Figure 1). For instance, the annual transport to streams of atrazine, a commonly used herbicide, accounts approximately for 1% of the amount applied to their watershed (U.S. Geological Survey, 2006). Chemical and biological reactions in the ground, the atmosphere, and water transform pesticides to new compounds, which may or may not contain the same or new toxicological properties, with a half-life specific for each individual chemical. Depending on their chemical-physical properties, pesticides and their transformed and bio-transformed byproducts drift through the atmosphere and/or move through streams and groundwater to substantial distance from their original point source. The atmospheric drift of long-lived organochlorines, such as chlordane, DDT, and dieldrin, toward remote geographical areas where they were never used, along with their prolonged bio-persistence and accumulation through the food chain, is thought to explain the global burden and their fat content in Arctic mammals (Majewski & Capel, 1995; U.S. Geological Survey, 2006).

date: 29 September 2016



Click to view larger

Figure 1. Distribution of pesticides in the environment. (Modified from U.S. Geological Survey, 2006.)

Exposure to pesticides may occur in the chemical industry, at the manufacturing stage, and in the application in open agricultural fields, but also in greenhouses, warehouses, stables, and in urban and rural environments for public health purposes. Insecticides, fungicides, and rodenticides can be used in libraries and archives to preserve the document collections for future use by humankind. Insects and mold can destroy printed pages, bindings, covers, and valuable documents or damage them with staining. Molds can act particularly quickly after flooding or due to faulty air-handling systems generating high-humidity conditions. Public meeting places (e.g., movie theaters, shopping malls, restaurants, and any commercial activity) also require a pest-free environment; mice, cockroaches, and other insects generate fear and anxiety in the staff and the public, and they can contaminate the food preparation process or make the ambient air unpleasantly smelly or a vehicle of microbial diseases. Herbicides are also extensively used to destroy weeds along railway tracks or highways. Finally, the general public can be exposed to the pesticide residues in the feedstock or through contaminated water.

With regard to the chemical industry, several serious accidents have occurred in different parts of the world. These include Bophal, India, in 1986 in a plant producing carbaryl; Seveso, Italy, in 1976; and Ludwigshafen, Germany, in 1953 in phenoxy herbicide manufacturing plants. These industrial accidents have contributed to public anxiety toward chemical plants, in general, and pesticide manufacturing and use in particular. Inappropriate disposal or use of pesticides has contributed by causing community poisonings, such as the dioxin contamination episode from landfills in the Quail Run Mobile Manor in Gray Summit, Missouri, in 1983, or the use of hexachlorobenzene as a grain fungicide in Turkey in 1955–1959 contaminating bread and causing about 4,000 poisonings and 500 deaths. Also, the easy availability of poorly regulated toxic pesticides in developing countries makes them popular for suicidal uses (Eddleston et al., 2002).

The relevance of such global contamination to human health depends on its geographic extent, the dietary intake through contaminated foodstuffs, the age segment of the affected population, and the often unpredictable interactions with other environmental

contaminants. Exposure to pesticides in pregnant women might have detrimental effects, depending on the specific chemical and the specific phase of the pregnancy, with congenital malformations and growth retardation possible during the organogenesis period (22–50 days since fertilization), and central nervous system effects arising in the subsequent days up to delivery (The European Commission, 2002). Children are mostly sensitive to toxic pesticides, as their organs keep developing quickly after birth, as are the elderly, because of slower metabolism and easier skin absorption. The prevalence of unfavorable metabolic gene polymorphism in the target population also has a significant impact on the number of those affected. However, unless as a consequence of serious industrial accidents, exposure from environmental sources is usually orders of magnitude lower than that occurring in the workplace. Yet, low-level lifetime exposure in vulnerable populations cannot be discarded. For instance, an increased risk on non-Hodgkin lymphoma has been observed in association with concentration of several organochlorines in household carpet dust (Colt et al., 2006), and an elevated risk of infant acute leukemia was reported following maternal exposure to household carbamate insecticides when carrying rearrangements in the MLL gene (Alexander et al., 2001).

Important resources mapping out pesticide uses in the croplands have been developed, such the Web-based Pesticide National Synthesis Project of the U.S. Geological Survey (USGS) of the U.S. Department of the Interior that maps the estimated amount of 482 pesticides used since 1992 at the county level (U.S. Geological Survey, 2016). Such “big data” resources are of major importance, particularly when it becomes possible to cross-link this information with health registries to timely detect time-space variations in disease occurrence.

Toxicology of Pesticides

Toxicological Classification and Labeling of Pesticides

In recent years, the United Nations Economic Commission for Europe (UNECE) Globally Harmonized System (GHS) has replaced the toxicological classification and labeling of chemical products (UNECE, 2009) by providing a basis for harmonizing regulations at the national, regional, and worldwide level to “enhance the protection of human health and the environment during the handling, transport and use of these chemicals.” The GHS classification is based on the acute effects, defined by the lethal dose (by ingestion) or lethal concentration (by inhalation) for 50% of experimental animals (DL_{50} or CL_{50}, respectively), expressed by mg/kg body weight or mg/liter. Based on the DL_{50} or CL_{50},

four classes of acute toxicity are defined (Table 1), each represented by a specific pictogram. The GHS classification is currently implemented in 67 world countries, including most European countries and the United States.

Table 1. Classification of Acute Toxicity According to the Global Harmonized System, UNECE, 2009

Hazard Class	Oral LD_{50} (mg/kg)	Skin LD_{50} (mg/kg)	Inhalation LC_{50} (mg/l)	
			Vapor/gas	Dust
1	≤ 5	≤ 50	≤ 0.5	≤ 0.05
2	50–300	50–200	0.5–2	0.05–0.5
3	300–2000	200–1000	2–10	0.5–1
4	2000–5000	1000–2000	10–20	1–5

Source: UNECE, 2009.

Based on the GHS criteria, the World Health Organization (WHO) International Program on Chemical Safety (IPCS) has reviewed its classification of selected pesticides by hazard (WHO, 2010). As a result, a few of the most hazardous pesticides have been moved between classes, taking into account severe health hazards not just acute toxicity (Table 2). The classification of active compound of selected pesticides is based on the acute oral and dermal LD_{50} to the rat; the most restrictive category is assigned in the case of discordance between the two values as well as adjusting by the acute human health effects not accounted for by LD_{50} assessments. In the case of formulations including two or more active ingredients, the manufacturer should apply the correct toxicological data. In the case of missing information from the manufacturer, the classification of a given formulation is defined by proportionally applying the respective LD_{50} to each ingredient according to the formula:

$$\Sigma \frac{LD_{50}\,\text{active ingredient}}{\%\ \text{active ingredient in formulation}} \times 100$$

Table 2. WHO IPCS Classification Scheme for Pesticides, Based on the LD_{50} for the Rat (mg/kg Body Weight)

Hazard class	**Oral**		**Dermal**	
	Solids	**Liquids**	**Solids**	**Liquids**
Ia. extremely hazardous	≤5	≤20	≤10	≤40
Ib. highly hazardous	5–50	20–200	10–100	40–400
II. moderately hazardous	50–500	200–2000	100–1000	400–4000
III. slightly hazardous	>500	>2000	>1000	>4000

Tables 3 and 4 show the active compounds used as pesticides in each of the four IPCS hazard classes, along with additional information on the chemical type, main uses, GHS classification, and LD_{50} in the rat. As the LD_{50} in the rat is taken as the reference, compounds used as rodenticide might be expected to be included among the upper hazard classes; however, their actual use is primarily for public health purposes, therefore, they are not listed in Tables 3 and 4. Compounds used against specific pests (e.g., mites, lice, and nematodes) are included among the insecticides.

Table 3. Pesticides Currently Used for Agricultural Purposes Listed in the IPCS Hazard Class Ia (Extremely Hazardous)

Common name	**CAS no.**	**Chemical type**	**Physical state**	**GHS hazard class**	**LD_{50} (mg/kg)**
Insecticides					
Aldicarb	116-06-3	Carbamate	Solid	1	0.93
Chlorethoxyfos	54593-83-8	Organophosphate	Liquid	1	1.8
Chlormephos	24934-91-6	Organophosphate	Liquid	2	7.0
Disulfoton	298-04-4	Organophosphate	Liquid	1	2.6
EPN (O-ethyl O-(4-nitrophenyl) phenylphosphonothioate)	2104-64-5	Organophosphate	Solid	2	14.0
Ethoprophos	2104-64-5	Organophosphate	Liquid	2	Dermal 26.0
Mevinphos	26718-65-0	Organophosphate	Liquid	1	Dermal 4.0
Parathion‡	56-38-2	Organophosphate	Liquid	2	13.0
Parathion-methyl‡	298-00-0	Organophosphate	Liquid	2	14.0
Phorate	298-02-2	Organophosphate	Liquid	1	2.0
Phosphamidon‡	13171-21-6	Organophosphate	Liquid	2	7.0
Sulfotep	3689-24-5	Organophosphate	Liquid	1	5.0
Tebupirimfos	96182-53-5	Organophosphate	Liquid	1	1.3

Terbufos	13071-79-9	Organophosphate	Liquid	1	2.0 (broad range)
Fungicides					
Calcium cyanide	592-01-8	Inorganic	Solid	2	39.0
Captafol*,‡	2425-06-1	Phthalimide	Solid	5	5000.0
Hexachlorobenzene*, ‡	118-74-1	Organochlorine	Solid	5	Dermal 10000.0
Mercury chloride‡	7487-94-7	Inorganic	Solid	1	1.0
Phenylmercury acetate‡	62-38-4	Organic mercury	Solid	2	24.0

(*) Captafol is an animal carcinogen (IARC Group 2A); hexachlorobenzene causes porphyria in humans and is included in the list of persistent organic pollutants, the use of which is restricted based on the Stockholm Convention as of May 17, 2004.

(**) Calcium cyanide yields cyanide gas by reacting with water.

(‡) Trade of captafol, hexachlorobenzene, mercury compounds, parathion, parathion-methyl, and phosphamidon is regulated by the Rotterdam Convention on prior informed consent as of February 24, 2004.

Table 4. Pesticides Currently Used for Agricultural Purposes Listed in the IPCS Hazard Class Ib (Highly Hazardous)

Common name	**CAS no**	**Chemical type**	**Physical state**	**GHS hazard class**	**LD_{50} (mg/kg)**
Insecticides					
Azinphos-ethyl	2642-71-9	Organophosphate	Solid	2	12.0
Azinphos-methyl	86-50-0	Organophosphate	Solid	2	16.0
Butocarboxim	34681-10-2	Carbamate	Liquid	3	158.0
Butoxycarboxim	34681-23-7	Carbamate	Liquid	3	Dermal 288.0
Cadusafos	95465-99-9	Organophosphate	liquid	2	37.0
Calcium arsenate	7778-44-1	Inorganic	Solid	2	20.0
Carbofuran‡	1563-66-2	Carbamate	Solid	2	8.0
Chlorfenvinphos	470-90-6	Organophosphate	Liquid	2	31.0
Coumaphos	56-72-4	Organophosphate	Solid	2	7.1
Cyfluthrin	68359-37-5	Pyrethroid	Solid	2	15 (broad range)
Beta-cyfluthrin	68359-37-5	Pyrethroid	Solid	2	11 (broad range)
Zeta-cypermethrin	52315-07-8	Pyrethroid	Liquid	3	86 (broad range)

Demeton-S-methyl	919-86-8	Organophosphate	Liquid	2	40.0
Dichlorvos	62-37-7	Organophosphate	Liquid	3	56.0
Dicrotophos	141-66-2	Organophosphate	Liquid	2	22.0
4,6-Dinitro-*o*-cresol ‡	534-52-1	Nitrophenol	Solid	2	25.0
Ethiofencarb	29973-13-5	Carbamate	Liquid	3	200.0
Famphur	52-85-7	Organophosphate	Solid	2	48.0
Fenamidophos	22224-92-6	Organophosphate	Solid	2	15.0
Flucytrinate	70124-77-5	Pyrethroid	Liquid	3	127.0 (broad range)
Formetanate	22259-30-9	Carbamate	Solid	2	21.0
Furathiocarb	65907-30-4	Carbamate	Liquid	2	42.0
Heptenophos	23560-59-0	Organophosphate	Liquid	3	96.0
Isoxathion	18854-04-8	Organophosphate	Liquid	3	112.0
Lead arsenate	7784-40-9	Inorganic	Solid	2	10.0 (broad range)
Mecarban	2595-54-2	Organophosphate	Oil	2	36.0
Methamidophos‡	10265-92-6	Organophosphate	Solid	2	30.0
Methidathion	950-37-8	Organophosphate	Liquid	2	25.0
Methiocarb	2032-65-7	Carbamate	Solid	2	20.0

Methomyl	16752-77-5	Carbamate	Solid	2	17.0
Monocrotophos‡	6923-22-4	Organophosphate	Solid	2	14
Omethoate	1113-02-6	Organophosphate	Liquid	2	50.0
Oxamyl	23135-22-0	Carbamate	Solid	2	6.0
Oxydemeton-methyl	301-12-2	Organophosphate	Liquid	3	65.0
Paris green (copper arsenate)	12002-03-8	Inorganic	Solid	2	22.0
Pentachlorophenol ‡	87-86-5	Chlorophenol	Solid	2	Dermal 80.0
Propetamphos	31218-83-4	Organophosphate	Liquid	3	106.0
Tefluthrin	79538-32-2	Pyrethroid	Solid	2	22.0 (broad range)
Thiofanox	39196-18-4	Carbamate	Solid	2	8.0
Thiometon	640-15-3	Organophosphate	Oil	3	120.0
Triazophos	24017-47-8	Organophosphate	Liquid	3	82.0
Vamidothion	2275-23-2	Organophosphate	Liquid	3	103.0
Fungicides					
Blasticidin-S	2079-00-7	Aminoacylnucleoside	Solid	2	16.0
Edifenphos	17109-49-8	Organophosphate	Liquid	3	150.0
Herbicides					

Acrolein	107-02-8	Aldehyde	Liquid	2	29.0
Allyl alcohol	107-18-6	Alcohol	Liquid	3	64.0
DInoterb	1420-07-1	Nitrophenol	Solid	2	25.0

(*) Also used as a herbicide.

(‡) Trade of carbofuran, DNOC, methamidophos, monocrotophos, and pentachlorophenol is regulated by the Rotterdam Convention on prior informed consent as of February 24, 2004.

Overall, besides those listed in Tables 3 and 4, another 215 active compounds are listed in IPCS class II (moderately hazardous): 79 insecticides, including popular insecticides such as acephate, carbaryl, chlordane, chlorpyrifos, cypermethrin, DDT, diazinon, dimethoate, endosulfan, lindane, permethrin, and propoxur; 56 fungicides, such as copper sulfate and other copper salts, propiconazole, tebuconazole, thiram, triadimefon, and ziram; 70 herbicides, such as 2,4-D, dicamba, diclofop, diquat, molinate, and paraquat; and a few others with different designations (8 plant growth regulators, 2 bacteriostatics, 1 unspecified). IPCS class III (slightly hazardous) lists another 104 active compounds: among them are the two best-selling herbicides atrazine and glyphosate and one of the most popular organophospates, malathion. Finally, IPCS class IV (pesticides unlikely to present acute hazards in normal use) includes 179 compounds. Upgrades are expected given the recent International Agency for Research on Cancer (IARC) revision of carcinogenicity of lindane (Group 1), DDT, and glyphosate (Group 2A) and others. The IPCS tables also list 291 active compounds known to have been discontinued, based on information from the manufacturers and on the subsequent editions of the Pesticide Manual. Further information on specific compounds can be found at http://www.who.int/ipcs/.

Toxicological Mechanisms

Organochlorines

Organochlorine insecticides result from hydrocarbons, aromatic or aliphatic, covalently bonded to one or more chlorine atoms, replacing hydrogen atoms. DDT was the first of a group of chlorinated aromatic hydrocarbons, including hexachlorobenzene and methoxychlor, most of which have been banned because of their toxicity to humans and wildlife and their persistence in the environment. These compounds affect the ion transport through membranes; in the neuron membrane, the inhibition of ion transport

slows or stops its repolarization following an impulse, resulting in hyperexcitability (Duffus & Worth, 2006). Cyclodienes (including the obsolete chlordane), aldrin, dieldrin, mirex, and the hexachlorocyclohexane isomers (including lindane and endosulfan) are all also discontinued; they act at the central nervous system level, as antagonists of the gamma-aminobutyric acid (GABA) neurotransmitter, which also affects neuronal repolarization, causing uncoordinated hyperexcitability (Duffus & Worth, 2006).

Due to their lipophilicity, the organochlorines are stored in body fat in a biologically inactive form, although they are extremely persistent, with a half-life for DDT reportedly within a 2- to 15-year range (Augustijn-Beckers, Hornsby, & Wauchope, 1994). This prolonged half-life is at once one of the major reason of concern for its environmental effects and one of the reasons for both the continuing effectiveness against malaria, yellow fever, typhus, and other vector-borne diseases and the relatively low cost of DDT.

In 1962, the biologist Rachel Carson published her influential book *Silent Spring* when DDT production was at its highest, with as much as 82 million kg/year being produced in the United States (Carson, 1962). In her book, and in several publications it inspired, adverse reproductive effects of organochlorine pesticides were described in fish and several avian species, including top predators, such as bald eagles, the American national bird. As a result of the growing public concern about its toxicity in wildlife, its widespread contamination, the increasing pest resistance, and the development of alternative insecticides, such as organophosphates, carbamates, and pyrethroids, DDT was banned in the early 1970s in numerous Western countries (U.S. Environmental Protection Agency, 1975). In 2001 the WHO Stockholm Convention on Persistent Organic Pollutants (POPs) made an exception to the worldwide ban on DDT production and use "for disease vector control in accordance with the World Health Organization recommendations and guidelines on the use of DDT and when locally safe, effective and affordable alternatives are not available . . ." In 2011, WHO declared its support for the indoor use of DDT in countries in which malaria is a health problem (World Health Organization, 2002).

Currently, DDT is off patent with only two plants (one in China and another apparently in North Korea) continuing its manufacture, and there are plenty of more expensive, less persistent, and less studied alternatives available from major chemical corporations; therefore, it is understandable why the debate on its complete ban in malaria-ravaged countries is still so controversial.

When administered orally, DDT has a low-to-moderate acute toxicity to mammals, which accounts for the delayed investigation on acute human health effects. These include nausea, diarrhea, increased liver enzyme activity, irritation of the eyes and upper airways, disturbed gait, malaise, and excitability. At high doses, tremors and convulsions

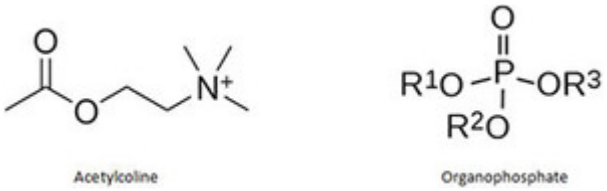
are possible (Extoxnet, 2008). The other organochlorines share with DDT the acute neurologic effects, although only the cyclodienes (aldrin, dieldrin, endrin, endosulfan, and methoxychlor) mirex and lindane are capable of inducing severe seizures and fatalities. Induction of hepatic microsomal drug–metabolizing enzymes may also occur following exposure to DDT and cyclodienes, which would accelerate their own metabolism but also interfere with that of steroid hormones and therapeutic drugs. Other effects include myocardial irritability, which would predispose to cardiac arrhythmia, and porphyria cutanea tarda, which has been described as a consequence of ingestion of hexachlorobenzene-treated wheat (Roberts & Routt Reigart, 2013). Reproductive and neoplastic effects have also been reported, which will be discussed under the specific subheadings.

The organochlorines stored in body fat are at equilibrium with the serum, where they tend to mobilize in the blood with pregnancy, breastfeeding, aging, disease, or fasting. Use of serum or plasma concentration is nowadays the standard method of biomonitoring organochlorine body burden (rather than fat biopsy), because of lesser invasiveness and easier acceptability of the procedure.

Organophosphates

Organophosphates are esters of phosphoric acid, with an oxygen atom linked with a double bond to phosphorus, two lipophilic groups, and a halide also bonded to phosphorus. Organophosphates inhibit acetylcholinesterase (AChE), an enzyme located in the postsynaptic membrane that degrades acetylcholine (ACh) into choline and acetic acid. ACh is the neurotransmitter of cholinergic postsynaptic receptors, including the nicotinic receptors (located in the neuromuscular junction and the autonomic ganglia) and the muscarinic receptors (located in the central nervous system, and at the junction between the postganglionic neuron and the tissue innervated by the parasympathetic autonomic nervous system). By degrading ACh while linked to its postsynaptic membrane receptor, AChE interrupts nervous stimulation; AChE inhibition by organophosphorous compounds allows a prolonged bond between ACh and its receptor, resulting in respiratory paralysis and death of the insect.

The effects in mammals differ in relation to changes in the metabolic pathways. The phosphorus-oxygen (=P=O) active group of organophosphates covalently binds the serine hydroxyl group in the active site of AChE, the same linking ACh (Figure 2). In fact, several thiophosphate insecticides, such as parathion and malathion, have a sulfur atom linked to phosphorus, which is replaced by oxygen by an oxidase in its first metabolic step resulting in the active metabolite. Their toxic action is therefore delayed and they are less toxic to humans compared to other organophosphates such as mevinphos or monocrotophos (Brown, 1999).



Click to view larger

Figure 2. Chemical structures of acetylcholine and organophosphate.

Replacing the oxygen with more stable substitutes makes the bond between the organophosphate and AChE irreversible. Such properties have been used to manufacture chemical weapons such as nerve gases, including sarin and the VX agent.

Organophosphates are metabolized by carboxyl esterases, leading to separation of a leaving group from alkyl phosphates (APs), which are subsequently excreted with urine and can be used as a biomarker of dose (Sudakin & Stone, 2011). When the enzyme activity is reduced more than 50% of the background level (Legaspi & Zenz, 1994) following organophosphate poisoning, AChE inactivation causes ACh accumulation throughout the nervous system; this is responsible for the nicotinic (asthenia, muscle weakness, tremor initially at the upper and lower limbs, headache, cramps, mydriasis, tonic-clonic convulsions, and tachycardia) and muscarinic (nausea, vomit, diarrhea, hypotension, abdominal spasms, bronchospasm, lachrymation, and salivation) clinical effects, due to overstimulation of the respective receptors. Such symptoms might be preceded by cough, phlegm, and headache, thus simulating a trivial flu episode.

Organophosphates can enter the organism by inhalation, dermal contact, or ingestion, with the last mainly associated with suicidal purposes. Biological monitoring of exposure is therefore the preferred method of exposure surveillance to account for multiple possible sources of absorption. The measurement of AChE activity in serum is easily performed, but it does not exactly reflect the true AChE in the nervous system, although both are inhibited by organophosphates. Instead, AChE activity in red blood cells is a more reliable predictor of enzyme activity in the nervous system. Measurement of urinary AP excretion complements the measurement of cholinesterase activity as it reflects recent exposure, while AChE enzyme activity recovers slowly over time. However, the lack of reference values for AP and the other metabolites of organophosphorous pesticides prevents their practical application to biomonitoring exposure in occupational settings (Legaspi & Zenz, 1994). More recently, the detection of organophosphate-protein adducts with cholinesterase in the blood of exposed subjects has been proposed as a biomarker of exposure within weeks from first exposure (Thompson, Prins, & George, 2010).

Carbamates

Carbamates are a group of insecticides deriving from carbamic acid. Their mechanism of action also involves AChE inhibition through carbamylation, which, unlike with organophosphates, is reversible within a few hours, thus preventing its use as a biomarker (Legaspi & Zenz, 1994). As for organophosphates, exposure occurs by inhalation and skin contact, with ingestion involved mainly in suicidal attempts or incidents. Measuring the active molecule itself in the serum or urinary metabolites, such as 1-naphthol for carbaryl and phenol conjugates for propoxur and carbofuran, has been explored (Araoud, 2011), but not widely applied.

Carbamates are usually considered to be of limited acute toxicity. For this reason, carbaryl dust is used against fleas in pets and in typhus control public health programs. However, at high concentration, carbaryl can cause skin irritation and systemic intoxication. Aldicarb is listed in the IPCS group Ia of extremely hazardous pesticides in conditions of normal use, and several other commonly used carbamates, such as carbofuran and methomyl, are listed in the IPCS group Ib of highly toxic pesticides (see Table 4).

Dithiocarbamates

Dithiocarbamates act as metabolic inhibitors, and they are non-AChE inhibiting substances, generally of low toxicity, resulting from dithiocarbamic acid complexes with transition metals, such as zinc or manganese. Zineb, Maneb, Mancozeb, Thiram, and Ziram are among the most popular in this class of pesticides. These compounds are mainly used as fungicides and animal repellents in orchards and to protect harvested crops and seeds during storage and transportation, as well as in the treatment of human scabies, as sunscreens, and as bactericides incorporated into soap or directly applied to the skin (Extoxnet, 2008). Several dithiocarbamates can cause nausea, vomiting, and headache when consumed with alcohol, and for this reason disulfiram has been used in the treatment of alcoholics as a form of voluntary behavior modification. Thiram is an irritant for the upper airways and the conjunctiva, and acute exposure is followed by headaches, dizziness, fatigue, nausea, and diarrhea (Extoxnet, 2008); prolonged exposure can cause skin sensitization with contact dermatitis in the hands, forearms, and feet (Legaspi & Zenz, 1994). Rare cases of mild peripheral neuropathy have been reported following disulfiram treatment in alcoholics (Gessner & Gessner, 1992), and signs of poor coordination, abnormal deep tendon reflexes, and reduced muscular strength have been reported among Ecuadorian pesticide users, who were also exposed to organophosphates and carbamates (Cole, Carpio, Julian, & Leon, 1998). This might be an indirect effect, as the metabolic breakdown of dithiocarbamates can lead to formation of carbon disulfide (used in the past as a solvent in the rubber industry), a powerful neurotoxin of the peripheral nerves.

The urinary excretion of ethylenethiourea (ETU) is used as a biomarker of workplace exposure to dithiocarbamates, but also to monitor the dietary consumption of such chemicals as food contaminants in the general population. Because ETU inhibits the synthesis of thyroid hormones, high-level exposures to the parent fungicides can be associated with possible hypothyroidism, increased thyroid-stimulating hormone (TSH) secretion, and hypofunctional goiter (Colosio & Rubino, 2015).

Pyrethroids

Pyrethrum is a mixture of chemicals including six pyrethrins with active insecticidal properties naturally occurring in chrysanthemum flowers; the pyrethrins are known to have been used in Asia against ticks and various insects since the 19th century. Currently used pyrethroids are structurally similar to the pyrethrins, but they are chemically manufactured to be more active against insects and to last longer in the environment (Bradberry, Cage, Proudfoot & Vale, 2005). Type I carboxylic esters, such as permethrin, block the sodium channels in nerve membrane, thus causing repetitive and prolonged neuronal discharge, but not severe depolarization. Class II cyano esters, such as cypermethrine, cause more persistent membrane depolarization and eventually nerve blockade, resulting in respiratory paralysis in insects (Bradberry et al., 2005). Pyrethroids are rapidly metabolized in humans mostly through hydrolysis of the ester linkage and subsequent oxidation and/or conjugation. Others, such as allethrin, more resistant to hydrolysis, undergo direct oxidation (Miyamoto, 1976).

Effects in humans are mild because of lesser sodium channel sensitivity, greater body size and higher body temperature, and their rapid metabolism. Symptoms include numbness, paresthesias, and, following severe type II poisoning, seizures due to the blockade of the GABA-gated chloride channels. Dermal absorption is the main route with occupational exposure; inhalation exposure is increasing in importance when pyrethroids are used in confined spaces. Despite their extensive use worldwide, including as household insecticides, severe human poisoning has seldom been reported (Bradberry et al., 2005). Pyrethroid metabolites, such as 3-phenoxybenzoic acid (3-PBA), *trans*-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-1-carboxylic acid (*trans*-DCCA), 4-fluoro-3-phenoxybenzoic acid, and *cis*-3-(2,2-dibromovinyl)-2,2-dimethylcyclopropane-1-carboxylic acid, can be monitored in the urine using high-performance liquid chromatography coupled with mass spectrometry (HPLC/MS/MS) (McKelvey et al., 2013)[35].

Chlorophenoxyacetic Acids

Chlorophenoxyacetic acids include several chemicals functionally related to the natural growth hormone indole acetic acid (IAA), that act selectively on broadleaf plants, by inducing a rapid, uncontrolled growth, followed by death, while preserving crops. 2,4-Dichlorophenoxyactic acid (2,4-D) and 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) were

date: 29 September 2016

synthesized in 1941, and methylchlorophenoxyacetic acid (MCPA) was introduced in the market in 1945 (IARC, 1986). Use of 2,4,5-T was discontinued in the United States and numerous other countries in the early 1970s, as inherently contaminated with the 2,3,7,8-tetrachlorodibenzo-*para*-dioxin (TCDD); 2,4-D was a major (50%) component of the Agent Orange defoliant used during the Vietnam War, as well as 2,4,5-T contaminated by TCDD.

Episodes of poisoning among workers and the general population and environmental contamination with dioxin, due to accidents in 2,4,5-T manufacturing plants, have occurred in Germany and in Italy (Schechter, 1994). More recently, manufactured 2,4-D technical acids have been used in crops, pasture and rangeland, forest management, home and garden, and for the control of aquatic vegetation; these are free of dioxin contamination, but 2,4-D amine and ester products may contain TCDD in amounts ranging from 5 to 500 ppb (Extoxnet, 2008). Other widely used phenoxy herbicides include MCPA, not manufactured in the United States, mecoprop, and dichlorprop. Fenoprop, known as Silvex in the United States, was banned in 1985 (U.S. Environmental Protection Agency, 2016).

Phenoxy herbicide can be readily absorbed by inhalation, skin contact, and ingestion and is excreted in urine mostly unchanged within various time lags depending on the individual compound, with half-lives ranging from 10 to 20 hours for 2,4-D, less than 24 hours for 2,4,5-T and MCPA, and somewhat longer for Silvex, dichlorprop, and mecoprop (IARC, 1986). Plasma and urinary levels of the unmodified phenoxy acids can be used for exposure biomonitoring (Legaspi & Zenz, 1994). There is no evidence of accumulation of phenoxy acids in human tissues. With a 375–666 mg/kg LD_{50} in the rat, 2,4-D is classified as moderately hazardous because of producing eye and skin irritation among agricultural workers. Fatigue, nausea, and rare instances of peripheral nerve effects have been described following high-level exposures. Prolonged inhalation can also cause cough, dizziness, and temporary lack of muscle coordination. Nonfatal intoxications with 2,4-D, mostly due to suicidal attempts or ingestion incidents, have resulted in acute parasympathetic symptoms and persistent neurological dysfunction. No long-term consequences were observed in a surviving case of MCPA poisoning (Extoxnet, 2008; IARC, 1986).

Other Pesticides of Common Use

Atrazine

Atrazine is a triazine herbicide used in agricultural crops, including sugarcane, corn, pineapples, rice, and on evergreen tree farms and forests, before and after crop emergence, and against weed growth in highways and railroads, in the spring and summer months (Extoxnet, 2008). Although banned in Europe in 2004 because of groundwater contamination, in the United States it is still the second most widely used

herbicide after glyphosate. It can enter the body through inhalation, ingestion or skin contact. It is listed among the slightly hazardous pesticides (IPCS class III), although it can cause skin rashes, irritation of conjunctiva and mucous membranes, abdominal pain, diarrhea, and vomiting. Several episodes of lethal food poisoning caused by atrazine have been described. About two thirds of the ingested dose is eliminated in the urine within 72 hours, while a 15% residue is retained, mainly in the liver, kidneys, and lungs (Hayes & Laws, 1990). Glutathione conjugation is the major route of biotransformation, resulting in atrazine mercapturate and *N*-dealkylation; urinary concentrations of these metabolites reflect recent exposure.

Glyphosate

Glyphosate is the technical name of *N*-(phosphonomethyl)glycine, initially synthesized for medical applications, but with herbicide properties discovered in 1970 (IARC, 2015). It is an acid, formulated as a salt with isopropylamine, ammonium, or sodium in commercial preparations. As it contains a phosphatidyl functional group, it is commonly considered as an organophosphate, but it is not an organophosphate ester and it does not significantly inhibit cholinesterase activity (Weed Science Society of America, 1994). Because of its structural resemblance to phosphoenol pyruvate, once absorbed by the plant, glyphosate inhibits enolpyruvyl-shikimate-3-phosphate synthase (EPSPS); this enzyme converts the end products of glycolysis and the pentose phosphate pathway to 5-enolpyruvyl-shikimate-3-phosphate (ESP), a precursor for aromatic amino acids, hormones, vitamins and other essential plant metabolites (Glyphosate Task Force, 2013).

Glyphosate is the best-selling herbicide worldwide, for the control of annual and perennial plants including grasses, sedges, broadleaved weeds, and woody plants; it can be used on noncropland as well as on a great variety of crops (Extoxnet, 2008). Experimental studies have not shown significant signs of acute or chronic toxicity, reproductive or mutagenic effects, and therefore it is included in IPCS class III (slightly hazardous). However, IARC has recently evaluated the evidence for human carcinogenicity of glyphosate and classified it in the Group 2A as a “probable human carcinogen,” because of limited evidence of an association with risk of non-Hodgkin’s lymphoma in human studies and sufficient evidence of carcinogenicity in experimental animals (Cole et al., 1998). A few clinical reports of rhabdomyolysis among persons exposed to organophosphates and specifically to glyphosate have been published, and the IPCS states that “rhabdomyolysis is a well known consequence of serious intoxications, and it shows up with relative frequency in association with serious intoxications by organophosphates” (IPCS INCHEM, 1998); however, the association has not been tested with formal epidemiologic studies to date. Glyphosate is poorly absorbed from the digestive tract and is largely excreted unchanged by mammals (Miyamoto, 1976); urinary excretion can be by monitored with HPLC, with post-column reaction and fluorescence

detection, in occupationally and environmentally exposed population groups (Acquavella et al., 2004).

Paraquat

Paraquat (*N*,*N*′-dimethyl-4,4′-bipyridinium dichloride) is a quaternary nitrogen herbicide used for broadleaf weed control, as a crop desiccant and defoliant, and as an aquatic herbicide in numerous countries, while it is regulated in the United States for use by certified applicators only (Extoxnet, 2008). According to the IPCS classification (see below), Paraquat is moderately hazardous (class II); however, a few instances of accidental or suicidal ingestion have been described, with the lung as the target organ of paraquat toxicity, resulting in a typical pulmonary fibrosis (“paraquat lung”) and death from respiratory failure. The lethal dose in humans is 35 mg/kg. Kidney is another target organ of paraquat toxicity; accidental or suicidal ingestion of very high doses has resulted in acute kidney failure due to proximal tubular dysfunction, similar to observations in experimental animals (Legaspi & Zenz, 1994).

Paraquat accumulates in the lung tissue where it is reduced to an unstable free radical by a NADPH-dependent microsomal flavoprotein reductase and subsequently re-oxidized to produce a superoxide anion. Lung toxicity results from cell death by lipid peroxidation or NADPH depletion (Smith, 1987). Skin absorption is normally less important, and it is not followed by systemic toxicity. However, prolonged contact can result in necrosis, white spots, and/or cracking of nails; if extensive skin damage occurs, paraquat can be absorbed and systemic toxicity becomes more likely (Bismuth, Hall & Wong, 1995). Paraquat can be measured in urine samples by enzyme-linked immunosorbent assay (ELISA), a useful workplace exposure biomarker (Park et al., 2008).

Azole Fungicides

Azoles are a group of heterocyclic compounds with a 5-atom ring, including two carbon and three nitrogen atoms, which differentiate by the relative position and the hydrogen bond of nitrogen atoms. They include two major groups, imidazoles (such as thiophanate methyl) and triazoles (such as propiconazole, tebuconazole, and triadimefon); these chemicals are widely used in a number of crops (vegetables, banana, apples, plums, papaya, grapes, citrus fruit, ornamental plants, wheat) as protective agents against fungi. Several azoles, such as ketoconazole and fluconazole, are also used to treat fungal infections in humans (Colosio & Rubino, 2015).

Azoles act by inhibiting lanosterol-14α-demethylase (CYP51), the main component of fungal membranes. However, other P-450 enzymes, including aromatase (CYP19), which is implicated in the transformation of testosterone and androstenedione to estradiol and estrone, respectively, are also inhibited by azole compounds. Also, CYP51 is highly

expressed in male germ cells in early stages of their development, and its inhibition, together with CYP19 inhibition in humans, might result in endocrine disruption (Zarn, Bruschweiler, & Schlatter, 2003).

Inhalation of the azoles can cause irritation of the upper airways and the lungs. Azoles can be easily absorbed by dermal contact or ingestion; the unchanged compound is eliminated in the urine and feces in a few days, although a small amount undergoes biotransformation in the liver by oxidation (CYP3A4). The conjugated metabolites, such as triazolylalanine, triazolylacetic acid, and triazolylpyruvic acid, are common metabolites of triazole fungicides and can be detected in the urine (U.S. Environmental Protection Agency, 2008).

Methyl Bromide

Methyl bromide is the most widely used fumigant (a substance that produces a gas, vapor, fume, or smoke with a wide spectrum of biocide effects) (Legaspi & Zenz, 1994). Fumigants are applied on the soils, prior to sowing, to sterilize them particularly against fungi spores, but their main use is to protect grains, cereals, flour, nuts, and rice in interior buildings (such as warehouses and storage rooms). Methyl bromide, in particular, also has insecticide and rodenticide properties, and therefore it is also used to treat railroad cars or buildings.

Other fumigants include ethylene dibromide, ethylene dichloride, dichlorobenzene, sulfur dioxide, carbon disulfide, hydrogen cyanide, and calcium cyanide. Several of these chemicals (including methyl bromide, ethylene dichloride, and hydrogen cyanide) are highly toxic, but they are not classified by IPCS, although threshold limit values (TLVs) for occupational exposures have been adopted in several countries. The methyl bromide TLV is 5 ppm; it is denser than air, so that it can form clouds in low ventilated areas where it concentrates.

Human exposure to methyl bromide can occur during its use or when entering treated buildings without proper personal protective equipment. Main routes of absorption are inhalation and dermal contact. Because methyl bromide is a strong irritant, it can cause a severe vesicular dermatitis particularly in moist skin area, such as axillae, groin, and abdomen; by inhalation exposure, methyl bromide can cause pulmonary edema, with tremors and convulsions, dizziness, frequently with a delayed onset. Chronic exposure can lead to a peripheral neuropathy, vision and hearing disturbances, central nervous system effects (e.g., confusion, depression, euphoria, hallucination, irritability, and mood changes), and tubular kidney damage. In the cells, methyl bromide inactivates several enzymes, particularly hexokinases and pyruvate kinases, by methylation of their SH groups, thus interfering with pyruvate metabolism (Pillay, 2013).

Once inhaled, a portion of the methyl bromide is eliminated unchanged in the exhaled air, but a more substantial amount is decomposed into a bromide ion and methanol, subsequently detected in blood and urine. The half-life of plasma bromide is about 12 days, which accounts for its delayed and prolonged health effects (Extoxnet, 2008). Although plasma bromide measurement has been used to identify the agent following workplace intoxication, its use for routine monitoring of occupational exposure is not suggested (Legaspi & Zenz, 1994).

Health Effects of Pesticides

Asthma and Other Respiratory Effects

The occurrence of respiratory symptoms, impaired respiratory function, asthma, and chronic bronchitis among agricultural workers exposed to pesticides has been investigated in cross-sectional and longitudinal studies (Mamane, Baldi, Tessier, Raherison, & Bouvier, 2015). A review of the literature reported that, among cross-sectional studies, most studies of respiratory symptoms (12/15), and all studies of asthma (N = 4), respiratory function (N = 4), and chronic bronchitis (N = 3), found an association with occupational exposure to pesticides. Both obstructive and restrictive ventilatory impairments were reported, depending on what chemical class of pesticide was involved. Results from longitudinal studies were less consistent for asthmatic symptoms, while a positive association for chronic bronchitis was consistently reported in three studies. An elevated risk of chronic obstructive pulmonary disease and airways obstruction was also observed in pesticide production workers (Mamane et al., 2015). Most studies were unable to identify the specific chemical agent responsible for the observed associations. However, risk of wheeze, detected by questionnaire in a cross-sectional study, was moderately elevated (12–50%) among applicators of the herbicides, paraquat, S-ethyl-dipropylthiocarbamate (EPTC), and atrazine, and the organophosphate insecticides parathion, malathion, and chlorpyrifos (Hoppin, Umbach, London, Alavanja, & Sandler, 2002). Among pesticide applicators, significant associations with wheezing were also reported for the organophosphates dichlorvos and phorate and the herbicide chlorimuron-ethyl (Hoppin, London, Linch, & Alavanja, 2006). An association with the exacerbation of atopic asthma was noted for the herbicide pendimethalin and the carbamate insecticide aldicarb (Henneberger et al., 2013).

Also, atopic asthma was reported more frequently with exposure to seven insecticides, two herbicides, and one fungicide among farmers participating in the U.S. Agricultural

date: 29 September 2016

Health Study. Risks were mostly elevated for both genders of agricultural workers with parathion and coumaphos, but only for women with metalaxyl and only for men with heptachlor, ethylene dibromide, and carbon disulfide. Pesticides associated with an increased risk of nonatopic asthma were permethrin among women and DDT, malathion, and phorate among men (Hoppin et al., 2008, 2009).

Immunotoxicity

Different groups of pesticides, including organophosphates and organochlorines, can affect numerous steps of the immune response (Li, 2007). For instance, several organophosphates can inhibit natural killer (NK) cell, lymphokine-activated killer (LAK) cell, and cytotoxic T lymphocytes (CTL) activity both in human and in animal studies, which would result in a reduced cellular immunity. T-cell subsets are also affected with a decrease in CD5 cells and, to a lesser extent, in CD4 cells; an increase in CD26 cells; and a moderate decrease in IL-2 production. Macrophages increase in size and phagocytic capability in mice. The humoral response is also affected, with a reduction in antibody production and in neutrophil function and an increase in autoantibodies; the Th1/Th2 cytokine balance is also disrupted, with a mild decrease in the count of CD4 T-helper lymphocytes (Li, 2007). The overall result is a mild immunosuppression.

The mechanisms of organophosphate immunotoxicity include: impairment of the granule exocytosis pathway and the FasL/Fas pathway of NK cells, LAK cells, and CTLs; induction of apoptosis and inhibition of serine hydrolases or other esterases of immune cells; and other yet to be completely elucidated pathways. Carbamates also share some of these immunotoxic effects (Corsini, Sokooti, Galli, Moretto, & Colosio, 2013). Consistent evidence is reported on the immunosuppressive effects of chlorinated compounds, such as pentachlorophenol and hexachlorobenzene. There is evidence for DDT in rodent studies, while human data are scanty, with some suggestion of an inverse correlation between plasma DDE body levels and IgA levels (Corsini et al., 2013). Chlordane seems associated with aberrant peripheral T-cell and B-cell regulation and autoimmunity approximately 10 years after exposure (Corsini et al., 2013). Infants exposed in utero to organochlorines may be more susceptible to their immunosuppressive effects, including a low white blood cell count, particularly lymphocytes, associated with depressed TNFα and IL-10 secretion. On the other hand, dithiocabamates, such as mancozeb, appear to act as immunomodulators and to enhance the immune response; thus, they have been proposed as therapeutic agents and T-cell-specific stimulants against immunodeficiency conditions in adulthood (Corsini et al., 2013).

Endocrine Disruption

An endocrine disrupter is defined as an “exogenous substance or mixture that can alter functions of the endocrine system and consequently causes adverse effects in an intact organism or its progeny or in a subpopulation” (WHO International Program on Chemical Safety, 2002). Concern over potential endocrine disrupting effects of pesticides arose following the experimental finding that organochlorines (particularly DDT isomers and their DDE derivatives) and atrazine bind in vitro to the rat androgen receptor, thus significantly inhibiting the specific binding of [3H]5 α-dihydroxytestosterone (DHT) (Kelce et al., 1995). Various organochlorines also compete in vitro with β-estradiol in linking with the rat estrogen receptor and with the progesterone receptor and the estrogen receptor in alligators (Danzo, 1997; Vonier, Crain, McLachlan, Guillette, & Arnold, 1996). The response of estrogen responsive mammary rat tumor cells to *o,p’*-DDT (a less prevalent DDT isomer present as a contaminant in technical grade DDT) mimics that elicited by the natural estrogen 17β-estradiol (Robison, Sirbasku, & Stancel, 1985); the extent of the response in vitro varies between 70% and 100%, although delayed, and it links to the 8-9S estrogen-binding protein of rat testicular cytosol. In fact, the *o,p’*-DDT estrogen-inducible protein is indistinguishable from that formed after 17β-estradiol. Methoxychlor and β-HCH, at blood concentrations in the µg/L range, were associated with increased uterine and vaginal epithelial thickness compared to control animals (Ulrich, Caperell-Grant, Jung, Hites, & Bigsby, 2000); similar to *o,p*-DDT, these compounds have affected the rate male sexual behavior in adulthood when administered during pregnancy (Vom Saal et al., 1995).

A valuable tool to assess human fertility is to calculate the “time to pregnancy” (TTP) index (i.e., the time before achieving pregnancy with unprotected intercourse (Joffe, 1997)) in relation to pesticide exposure. The analysis is frequently conducted using a modified Cox’s proportional hazard model, cutting the count of events at 12 months, which corresponds to the medical diagnosis of infertility. In this analysis, the hazard ratio between the time-related success in achieving pregnancy in the exposed versus the unexposed group is defined as the fecundability ratio. The exposure of Dutch fruit growers to the fungicide captan was associated with a significant reduction in the fecundability ratio (De Cock, Westveer, Heederik, Te Velde, & Van Kooij, 1994). Dibromochloropropane (DBCP) is another fungicide capable of significantly interfering with human reproduction; it was mostly used as an insecticide in banana plantations, causing azoospermia and oligospermia in the male workers in DBCP- manufacturing plants (Whorton, Millby, Krauss, & Stubbs, 1979) and a decrease in fertility (including sterility) among male Costa Rican banana plantation workers (Potashnik & Porath, 1995).

Although there are claims that DDT is responsible for the worldwide decline in sperm counts, it has not been shown to contribute to an impairment of male human fertility among highly exposed pest control applicators or among the general population (Campagna, Satta, Fadda, Pili, & Cocco, 2015; Cocco, Fadda, & Melis, 2006). Other pesticides for which a reduced human fecundability has been reported include dicamba, 2,4-D, dimethoate, and glyphosate (Cocco, 2002).

Other outcomes indirectly related to human fertility have been investigated in relation to pesticide exposure, including sperm count, sex hormone levels (e.g., plasma follicle-stimulating hormone (FSH), luteinizing hormone (LH), and testosterone, and serum hormone-binding globulin (SHBG) levels), spontaneous abortion, and the male:female ratio at birth (Cocco, 2002) [76]. Table 5 provides a summary overview of the pesticides that have been tested for their endocrine disrupting potential. Overall, apart from specific exceptions, human studies demonstrating endocrine-disrupting effects are infrequent, and when they are available, the results seem inconsistent. A possible explanation for the inconsistent findings might be the effect of interindividual variations in response due to polymorphisms of metabolizing genes, such as cytochrome P4502E1, the glutathione S-transferases m and q, and the paraoxonase genes (Au, Sierra-Torres, Cajas-Salazar, Shipp, & Legator, 1999).

Table 5. Summary Evaluation of Pesticides with Endocrine-Disrupting Potential

Pesticide	**In vitro studies**	**Animal studies**	**Human studies**	**UK-DE group***	**C u**
Organochlorines					
Acetochlor		Thyroid inhibitor			I c
Alachlor	Antiestrogen	No reproductive effects in adult rats			I c a k a s I k g s a s I t
Aldrin	Weak estrogen, anti-androgen				I u 1
Chlordane and oxychlordane	Weak estrogen and anti-androgen	Reduced fertility in rats			I t c I t s

Chlordecone (kepone)	Weak estrogen competitive aPR binding	Reduced fertility	Reduction in sperm quality		
DDT congeners	Anti-androgen, estrogen	Estrogenic effects	No evidence from population studies		
o,p′-DDE *o,p′*-DDE	Estrogen	Thyroid inhibition			
p,p′-DDE*p,p′*-DDE	Anti-androgen				
Dicofol	Weak estrogen and anti-androgen				
Dieldrin	Weak estrogen, weak anti-androgen	No reproductive effects			

Endosulfan	Weak estrogen Competitive aER and aPR binding Impaired steroid synthesis in Leydig cells	Damage to seminiferous tubules in male rats and reproductive organs in female mice			
Endrin	Weak estrogen and anti-androgen				
Fenarimol	Weak estrogen and anti-androgen				
Heptachlor		Reduced fertility in rats			
Pentachlorophenol	Anti-estrogen, weak anti-androgen	No reduced fertility in mice and rats			

Methoxychlor	Estrogen, anti-androgen	Testicular atrophy, decreased sperm production, testosterone levels in rats and mice. Reduced fertility in both genders. Thyroid inhibitor			
Hexachlorocyclohexane		Reduced fertility	Changes in the levels of sex hormones		
α-HCH	Weak anti-androgen				
β-HCH	No effect	Weak estrogen-like effects in mice and rats			
(lindane) γ-HCH	Weak anti-androgen	Testicular atrophy, decreased sperm production, testosterone levels in rats and mink			

	No estrogen effect Impaired steroid synthesis in Leydig cells				
δ-HCH	Weak anti-androgen				
Mirex and photomirex	Weak estrogen. No anti-androgenic effect	Reduced fertility due to testicular degeneration Affects thyroid and parathyroid			
Nonachlor (*cis*- and *trans*-)	Anti-estrogen. Inhibits aER binding of [3H]17 β-estradiol	Sex-reversal in alligator embryos and turtles			
Toxaphene	Weak estrogen Inhibition of ACTH-stimulated corticosterone synthesis in the adrenal cortex	Thyroid inhibitor			
Trans-nonachlor	Weak estrogen Anti-androgen				
Carbamates and Thiocarbamates					

Aldicarb	Weak estrogen	No reproductive effects			
Benomyl (and its breakdown product carbendazim)	Microtubule disruptor Weak estrogen	Decreased sperm production in adult male rats			
Bendiocarb, methomyl, and oxamyl	Weak estrogens	No decrease of fertility in rats			
Carbaryl	Anti-estrogen	Thyroid inhibition and reduced fertility in several animal species	Conflicting results on reduced fecundability		
Carbofuran		Testicular damage in dogs			
Chlorpropham	Anti-androgen				
Mancozeb		Thyroid inhibitor in rats	Goitrogen	B	
Maneb and Metiram		Thyroid inhibitors in several animal species No reproductive effects			
Methiocarb	Weak estrogen and anti-androgen				

Molinate		Reduced fertility	No reproductive effects		
Pirimicarb	Weak estrogen				
Propamocarb	Weak estrogen				
Propoxur	Weak estrogen	Reduced fertility and lactation in female rats			
Thiram		Infertility in male mice, delayed estrous cycle in female mice		A	
Zineb		Thyroid inhibitor in several animal species Reduced fertility in rats			
Ziram		Reduced fertility in female rats ans mice, and in male rats Testicular atrophy			
Organophosphates					
Azinphos-methyl	Anti-androgen				
Chlorpyrifos	Weak estrogen.	No reproductive effects		A	

Dichlorvos	Anti-androgen				
Dimethoate		No reproductive effects	Reduced fecundability	D	
Fenitrothion	Anti-androgen				
Fenthion	Anti-androgen				
Glyphosate		Reproductive changes at very high doses	Reduced fecundability	D	
Methamidophos		Reduced number of deliveries in female rats	Reduced sperm count and viability		
Methyl parathion and malathion	Inhibits catecholamine secretion	No reproductive effects in rats		D	
Parathion		Increased nocturnal synthesis of melatonin Gonadotrophic hormone inhibition			
Pirimiphos-methyl	Anti-androgen				
Tolclofos-methyl	Weak estrogen				

Trichlorfon		Reduced fertility and increased embryonic deaths in rats			
Other Pesticides					
Amitraz		Decreased fertility in rats			
Amitrole		Thyroid inhibitor in several animal species			
Atrazine	Weak estrogen, Weak anti-androgen Inhibits aER binding of [3H]17β-estradiol	Damage to adrenal glands Impairment of steroid hormone metabolism			
Abamectin		Reduced male fertility in several mammalian species Decrease in sperm count		B	
Azadirachtin		Infertility in several animal species			
Bromopropilate	Weak estrogen. Anti-androgen				
Bupirimate		Thyroid effects in rats		C	

Captan		Fetal loss or reduced weight at birth in mice	Reduced fecundability	D	
Chlomethoxyfen	Anti-androgen				
Chlornitrofen	Anti-androgen				
Clofentezine		Thyroid disruptor No reproductive effects			
Cycloprothrin	Anti-estrogen				
Cyfluthrin	Weak estrogen and anti-androgen				
Cyhalothrin	Anti-estrogen, anti-androgen				
Cypermethrin	Weak estrogen				
Cyprodinil	Anti-androgen				

2,4-D		No reduction in fertility Increase in thyroid and ovarian weights.	Reduced fecundability	A	
Deltamethrin	Weak estrogen				
Dibromochloropropane		Infertility in rabbits (atrophy of testes) testicular toxicant	Infertility		
Dicamba		No reduction in fertility in rats and rabbits	Reduced fecundability	D	
Dimethomorph	Anti-androgen				
Dinoseb		Reproductive effects in rats			
Ethylene dibromide		Reduced sperm count in bulls	Reduced sperm count and quality		

Ethoxyquin	Anti-androgen				A u I
Etofenprox	Anti-estrogen				I i
Fenbuconazole	Thyroid inhibitor Weak estrogen				I
Fenhexamid	Anti-androgen				I
Fenvalerate	Weak estrogen and anti-androgen				I i
Fipronil	Thyroid disruptor	Reproductive toxicant in rats			I
Fludioxonil	Anti-androgen				I
Hexachlorobenzene		Decreased male fertility No estrogenic effects			I u s t (c
Imazalil	Anti-androgen				I
Ioxynil	Compete with T4 in binding plasma proteins	Increase in thyroid function No reproductive effects		B	I

Iprodione	Binds the AR receptor; interference with thyroid hormones	Severe effects on prostate, adrenal glands in dogs and male reproductive organs in rats		C	I
Linuron	Androgen receptor agonist	Anti-androgenic No reproductive effects		B	I (c c I c c s a a
Metolachlor	Weak estrogen	Testicular atrophy in rats No reproductive effects in mice			I
Metribuzin	Estrogenic activity	Thyroid effects No reproductive effects		C	I
Myclobutanil	Antiestrogenic	Effects on prostate, adrenal glands, and male reproductive organs in rats		C	I
Nonyl-phenol	Weak estrogen				N
O-Phenyl phenol	Anti-androgen				I
Pentachloronitro benzene (PCNB)	Thyroid inhibitor				I u s t

Permethrin	Weak estrogen Anti-androgen				
Prochloraz	AR antagonism, aromatase inhibition Weak estrogen	Effects on ovaries, prostate and thyroid			
Procymidone	Anti-androgen	Anti-androgen in rats			
Prodiamine		Thyroid disruptor			
Propyzamide		Effects on thyroid and testes			
Pyrazoxyfen	Anti-estrogen				
Pyrimethanil	Anti-androgen	Thyroid inhibitor No reproductive effects			
Quinoxyfen	Anti-androgen				
Simazine		No reproductive effects in rats Distrophy and necrosis of germ cells in sheep			
Spiromesifen		Thyroid disruption, female reproductive toxicity		C	

Tebuconazole	Anti-estrogen and anti-androgen			C	
Tetramethrin	Anti-estrogen				
Thiacloprid	aromatase induction in rat liver	Thyroid and reproductive effects, cancer in reproductive organs		B	
Thiazopyr		Thyroid disruptor No reproductive effects			
Thiophanate-methyl		Thyroid effects		C	
Tribenuron-methyl	Weak estrogen				
Tributyltin		Short-term decrease in the activity of the pituitary-thyroid axis in rats No long-term effects.			
		Pseudoermaphroditism in gastropods			
Triflumizole	Anti-estrogen				
Vinclozolin	Anti-androgen Weak estrogen	Demasculinizing effects in rats No reproductive effects			

(*) Categorization of endocrine disrupters for regulatory purposes proposed by the UK-German Working Group for 98 selected pesticides for which human health endocrine disrupting assessments were conducted.

Sources: Cocco, Fadda, & Melis, 2006; DeCoster & van Larebeke, 2012; Ewence, Rumsby, & Johnson, 2013.

Based on the results of a competitive assay on proliferation of MCF-7 cells (an estrogen-dependent human breast cancer cell line), it has been estimated that the blood concentration of *o*,*p*’-DDE, the most potent xenoestrogen to significantly compete with estradiol in binding the estrogen receptor, would be not less than 30–180 µg/ml in fertile women and 10–70 µg/ml in postmenopausal women and men (Cocco, 2002).

Recently, a UK–German Working Group has proposed a categorization of endocrine disrupters for regulatory purposes for 98 selected pesticides for which human health endocrine-disrupting assessments were conducted, combining information from the ICPS classification scheme of pesticides with the available information on the endocrine effects in human, animal, and experimental studies (Bundesinstitut fur Risikobewertung, 2011). When endocrine-disrupting activities appear at or below the dose threshold with the application of a GHS-CLP category 1 or category 2 after repeated exposure either oral, dermal, or by inhalation, in short-term, medium-term, of chronic animal studies, the substance should be considered to pose a human health risk requiring consideration for regulatory action within the European Community (Ewence, Brescia, Johnson, & Rumsby, 2015). The UK–German Working Group assessments assigned each substance to one of four groups based on the mammalian toxicology or ecotoxicology data: Group A includes substances requiring additional information; Group B, endocrine disrupters more likely to pose a human health risk; Group C, endocrine disrupters less likely to pose a human health risk; and Group D, substances not considered to be an endocrine disrupter based on the mammalian toxicology data. The categorization resulting from such assessment is also reported in Table 5.

The ability of the so-called xenoestrogens to interfere with natural hormones by interacting with the same receptors depends on the dose and the receptor affinity with reference to the natural estrogen. The E-screen assay (based on the dose-related estrogen-dependent proliferation of MCF-7 cells, an estrogen-sensitive human breast cancer cell line) measures the relative potency of xenoestrogens (Soto et al., 1995). Outcomes of the E-screen assay are twofold: (a) the relative proliferative potency (RPP; the ratio between the estradiol concentration inducing maximal proliferation and the concentration of the test xenoestrogen required to achieve the same effect); and (b) the relative proliferative effect (RPE; 100 times the ratio between maximal cell yield achieved

with the xenoestrogen and that obtained with estradiol). Table 6 shows that the estradiol concentration required to induce maximal cell yields ranges between 10 and 100 picomoles; on the other hand, xenoestrogens may achieve a comparable effect for concentrations one million times greater, in the range of micromoles.

Table 6. The Estrogenic Potential of Some Pesticides and Therapeutic Hormones (with Reference to Estradiol) Based on the In Culture "E. Screen" Assay

Compound	**Concentration**	**RPE (%)**	**RPP (%)**
17β-Estradiol	10pM	100.00	100.00
DDT (technical grade)	10μM	79.61	0.0001
o,p'-DDT	10μM	86.14	0.0001
p,p'-DDT	10μM	71.00	0.0001
Dieldrin	10μM	54.89	0.0001
Endosulfan (technical grade)	10μM	81.25	0.0001
1-Hydroxychlordane	10μM	40.00	0.0001
Kepone	10μM	84.00	0.0001
Methoxychlor	10μM	57.00	0.0001
Toxaphene	10μM	51.90	0.0001

Source: Soto et al., 1995.

In the real world of pesticide manufacture and use, multiple pesticide exposures occur within a short time span, with different types of pseudo-endocrine effects which can interact among them in unpredictable ways either synergistically or antagonistically. Endocrine disruptors can act through nuclear receptors and through bound estrogen receptors; they can interact with cytoplasmic targets, modulate nitric oxide, and interfere with hormone metabolism; and they can cause changes in DNA methylation or histone modifications or genomic instability. Therefore, it is quite difficult to predict a threshold

concentration for the endocrine-disrupting effects of one pesticide only based exclusively on the relative potency of this single pesticide once linked to the estrogen receptor. Nevertheless, based upon the possible range of circulating estradiol levels, men, postmenopausal women, and children are expected to be more sensitive to xenoestrogens (Cocco, 2002).

Cancer

In 1991, IARC classified “occupational exposures in spraying and application of non-arsenical insecticides” as a group as “probable human carcinogens” (Group 2A) (IARC, 1991). This group is composed of 61 pesticides (including 26 insecticides, 12 fungicides, 17 herbicides, and 6 fumigants) examined in 8 different IARC monographs, among the 985 agents and groups of agents examined in 43 years of IARC Monographs. Some of these pesticides have been banned or abandoned for some decades, although a few of these keep being used in developing countries (Table 7). Arsenic and arsenical pesticides, 1,2-dichloropropane and lindane, are Group 1 human carcinogens. Group 2A, probable human carcinogens, includes the fungicide captafol (restricted in the United States and most countries since 1999), the herbicide glyphosate, and the insecticides malathion, diazinon, DDT (still allowed only for public health purposes), and ethylene dibromide (used as a grain fumigant). As for the rest, the inadequate or not available evidence from human studies is coupled with the limited or inadequate evidence from experimental animal studies.

Table 7. Evidence of Carcinogenicity and Summary Evaluation of Human Carcinogenicity of Pesticide Compounds examined in the IARC Monographs N. 1-114 (S= sufficient; I = inadequate; ND = no data available as of February 2016).

Compounds	**Year**	**Man**	**Animal**	**Summary evaluation of human carcinogenicity**
Insecticides				
Aldicarb	1991	ND	I	3
Aldrin	1987	ND	I	3
Aramite	1987	ND	S	2B
Arsenic and arsenical compounds	1987	S	L	1
Carbaryl	1987	ND	I	3
Chlordhane/eptachlor	2001	I	S	2B
Chlordecone	1987	ND	S	2B
Chlorobenzilate	1987	ND	L	3
DDT	2015	L	S	2A
Deltamethrin	1991	ND	I	3
Diazinon	2015	L	L	2A
Dichlorvos	1991	I	S	2B
Dicofol	1987	ND	L	3
Endrin	1987	ND	I	3
Fenvalerate	1991	ND	I	3

γ-Hexaxchlorocyclohexane (lindane)	2015	S	S	1
Malathion	2015	L	S	2A
Methoxychlor	1987	ND	I	3
Methyl-parathion	1987	ND	I	3
Mirex	1987	ND	S	2B
Parathion	2015	I	S	2B
Permethrin	1991	ND	I	3
Terpene polychlorinates (Strobane)	1987	ND	I	3
Tetrachlorvinphos	2015	I	S	2B
Toxaphene	2001	ND	S	2B
Trichlorfon	1987	ND	I	3
Fungicides				
Captafol	1991	ND	S	2A
Captan	1987	ND	L	3
Chlorothalonil	1999	ND	L	2B
Ferbam	1987	ND	I	3
Hexachlorobenzene	2001	I	S	2B
Hexachlorophene	1987	ND	I	3
Maneb	1987	ND	L	3

Pentachlorophenol and polychlorophenols as a class	1999	L	S	2B
Quintozene	1987	ND	I	3
Insecticides				
Sodium ortho-phenylphenate	1999	ND	S	2B
Thiram	1991	I	I	3
Ziram	1991	ND	L	3
Herbicides				
Amitrole	1987	I	S	2B
2,4-D	2015	I	L	2B
Atrazine	1999	I	S	3
Chlordimeform	1987	ND	I	3
Chloropropham	1987	ND	I	3
Diallate	1987	ND	L	3
Fluometuron	1987	ND	I	3
Glyphosate	2015	L	S	2A
MCPA	1987	I	L	2B
Monuron	1991	ND	L	3
Nitrofen	1987	ND	S	2B
Picloram	1991	ND	L	3

Propham	1987	ND	I	3
Simazine	1999	I	L	3
Sulfallate	1987	ND	S	2B
2,4,5-Trichlorophenoxyacetic acid	1987	I	I	2B
Trifluralin	1991	I	L	3
Fumigants				
1,2-Dibromo-3-chloropropane	1987	I	S	2B
1,3-Dichloropropene	1999	ND	S	2B
1,2-Dichloropropane	2014	S	L	1
Ethylene dibromide	1987	I	S	2A
Methyl bromide	1999	I	L	3
Piperonyl butoxide	1987	ND	I	3

(S= sufficient; I = inadequate; ND = no data available as of February 2016).

Thousands of chemicals are now available to farmers to treat plant diseases and protect their crops; their use changes year by year, across countries and within each country, by type of crop and by type of phytopathology. Therefore, the difficulty of conducting epidemiological studies of the long-term effects of agrochemicals is reflected in the inadequate information on their human carcinogenicity and the absence of evaluation by international scientific and regulatory agencies (Cocco et al., 2013).

The U.S. Agricultural Health Study is an extensive prospective longitudinal study of cancer and other health outcomes in a cohort of licensed pesticide applicators (including farmers) and their spouses in the states of Iowa and North Carolina to assess health risks associated with exposure to specific agricultural chemicals, with the support of a detailed retrospective exposure assessment and with proper consideration of lifestyle and genetic

factors (Alavanja et al., 1996). The study was started in 1993 as a collaborative effort involving investigators from National Cancer Institute, the National Institute of Environmental Health Sciences, the Environmental Protection Agency, and the National Institute for Occupational Safety and Health. The study began by collecting baseline information from participants at enrollment (1993–1997). Overall, 20,235 pesticide applicators and 1683 spouses were recruited. In two subsequent phases, 1999–2003 and 2005–2010, follow-up telephone interviews were conducted in 64% of private applicators, 59% of commercial applicators, and 74% of spouses. Questionnaire information included farming practices, lifestyle, and health and diet; a DNA sample from a buccal swab was also acquired. The interview was repeated in 2005–2010 by 46% of private applicators and 62% of spouses who participated in the baseline phase. An additional follow-up was initiated in 2013 with a questionnaire by mail, telephone, or through the Internet, this time including also those who had not participated in the previous phases. Thus far, the Agricultural Health Study has greatly contributed to the identification of cancer risks in relation to exposure to individual chemicals not previously evaluated. Table 8 lists the cancer outcomes associated with exposure to pesticides, including the compounds classified by IARC for which epidemiological studies were conducted, and those not yet examined for which suggestions emerged from the Agricultural Health Study or other studies. Only active compounds are listed in Table 8; contaminants (e.g., tetrachlorodibenzodioxin [TCDD], a group 1 human carcinogen according to IARC, which contaminated Agent Orange and other phenoxy herbicide preparations and caused environmental contaminations following industrial accidents) are not included.

Table 8. Individual Pesticide Compounds for Which Associations with Cancer Risk Have Emerged from The Agricultural Health Study and Other Studies.

Pesticide	IARC classification	Cancer site	Epidemiological evidence	Ref.
Alachlor	Not evaluated	Leukemia	Positive association in the highest exposure category	Alavanja, Ross, & Bonner, 2013
Aldicarb	3	Colorectum	Significant upward trend with exposure level (small numbers)	Christensen et al., 2010
Aldrin	3	Prostate	Increased risk in subjects with positive family history of PC.	Alavanja, Ross, & Bonner, 2013
Atrazine	3	NHL, thyroid	Positive associations with NHL in subjects with t(14:18). Suggestive increase in thyoid cancer (small numbers)	Alavanja, Ross, & Bonner, 2013; Guyton et al., 2015
Butylate	Not evaluated	Prostate, NHL	High risk in long duration exposed; interaction with family history (nonsignificant)	Freeman et al., 2011
Captan	3	Multiple myeloma	Positive association with MM risk	Alavanja, Ross, & Bonner, 2013

Carbaryl	3	Multiple myeloma, skin melanoma	Positive association with MM, and melanoma (long duration)	Alavanja, Ros,s & Bonner, 2013; Koutros et al., 2015
Chlordane	2B	Leukemia, NHL (inconsistent)	Positive association with leukemia risk; inconsistent results on risk of NHL	Alavanja, Ross, & Bonner, 2013
Chlordecone	2B	Prostate	Positive association in the highest exposure category	Alavanja, Ross, & Bonner, 2013
Chlorpyrifos	Not evaluated	Colorectum, lung	Significant upward trend with exposure	Christensen et al., 2010; Kang et al., 2008
Coumaphos	Not evaluated	Prostate	Increased risk in subjects with positive family history of PC.	Barry et al., 2012
2,4-D	2B	NHL	Inadequate evidence for inconsistent findings.	Loomis et al., 2015

DDT	2A	Liver, NHL, testis	Limited evidence for Inconsistent findings with serum DDE levels. Inadequate evidence for prostate and breast	Loomis et al., 2015
Diazinon	2A	NHL, leukemia and lung cancer	Increasing trends in risk after adjustment for other pesticides and confounders. Limited evidence.	Guyton et al., 2015
Dicamba	Not evaluated	Lung, colon	Significant upward trends	Samanic et al., 2006
Dieldrin	3	NHL, prostate cancer	Inadequate evidence for inconsistent findings for NHL. Suggestions for prostate cancer.	Alavanja, Ross, & Bonner, 2013
S-Ethyl dipropyl carbamothioate (EPTC)	Not evaluated	Leukemia, colon and pancreatic cancer	Relatively small number of exposed in the high exposure category, only. Significant trend for pancreatic cancer	Alavanja, Ross, & Bonner, 2013; Andreotti et al., 2009

Fonofos	Not evaluated	Prostate cancer, leukemia.	Trend in risk of prostate cancer. Positive association with leukemia; interaction with genetic variants in 8q24, base excision repair, nucleotide excision repair.	Alavanja, Ross, & Bonner, 2013
Glyphosate	2A	NHL	Limited evidence of a positive association with NHL risk	Guyton et al., 2015
Hexachloro benzene	2B	NHL, prostate	Positive associations with plasma level of HCH. Inadequate evidence. Inconsistent association with prostate cancer	Alavanja, Ross, & Bonner, 2013
Imazaquin	Not evaluated	Bladder	Positive association	Koutros et al., 2015
Imazethapyr	Not evaluated	Bladder, colon	Association observed only among nonsmokers	Koutros et al., 2015
Lindane	1	NHL	Consistent association for NHL	Loomis et al., 2015

Malathion	2A	NHL, prostate	Limited evidence for NHL and prostate cancer. Genotoxicity, oxidative stress, Inflammation, receptor-mediated effects, and cell proliferation or death	Guyton et al., 2015
Maneb/ mancozeb	3	Skin melanoma	Association with high duration exposure	Dennis, Lynch, Sandler & Alavanja, 2010
Metolachlor	Not evaluated	Liver, follicular lymphoma, lung	Significant upward trends with exposure	Alavanja et al., 2004; Silver et al., 2015
Methyl bromide	3	Prostate, stomach	Nonsignificant association among subject with positive family history of PC. Significant trend for stomach cancer.	Alavanja, Ross, & Bonner, 2013; Barry et al., 2012
Methyl-chloro phenoxyacetic acid (MCPA)	2B	NHL	Positive association among subjects with asthma or hay fever.	Alavanja, Ross, & Bonner, 2013

Metribuzin	Not evaluated	Leukemia, NHL	Significant upward trend for lymphohemopoietic malignancies, nonsignificant for leukemia and NHL	Alavanja, Ross, & Bonner, 2013
Mirex	3	NHL	Inconsistent epidemiological results	Alavanja, Ross, & Bonner, 2013
Parathion	2B	Skin melanoma	Association with high duration exposure	Dennis et al., 2010
Pendimethalin	Not evaluated	Pancreas, lung	Significant trend with exposure level	Alavanja et al, 2004; Andreotti et al., 2009
Permethrin	3	Multiple myeloma	Significant interaction with genetic variants in 8q24	Alavanja, Ross & Bonner, 2013
Phorate	Not evaluated	Prostate	Increased risk in subjects with positive family history of PC.	Mahajan, Bonner, Hoppin, & Alavanja, 2006
Simazine	3	Prostate	Inconsistent epidemiological results	Alavanja, Ross, & Bonner, 2013

Terbufos	Not evaluated	Prostate, lung, leukemia, NHL	Nonsignificant trends in risk. Significant interaction with genetic variants in 8q24	Alavanja, Ross, & Bonner, 2013; Bonner et al, 2010
Toxaphene	2B	NHL	Positive association with cases with t(14:18)	Alavanja, Ross, & Bonner, 2013
Trifluralin	3	Colon, kidney	Elevated risk for colon cancer in the highest exposure category. Nonsignificant association with kidney cancer	Kang et al., 2008

Other Health Effects

Central Nervous System and Peripheral Nervous System Effects

Acute high-level exposure to organophosphates, carbamate, and organochlorine pesticides is a well-established cause of neurotoxic effects, while the association of these effects with chronic exposure to low-moderate levels is more controversial (Kamel & Hoppin, 2004). The neurotoxicity of organophosphates is related to the inhibition of acetylcholinesterase and the resulting hyperstimulation of postsynaptic receptors. In less severe cases, symptoms (e.g., headache, dizziness, nausea, vomiting, pupillary constriction, and excessive sweating, tearing, and salivation) occur within minutes. More severe intoxications present with muscular weakness, bronchospasm, and twitching, with possible progression to convulsions and coma (Kamel & Hoppin, 2004). A delayed neuropathy due to axonal death can follow as a result of the acute organophosphate inhibition of an esterase and can be irreversible (Keifer & Mahurin, 1997). The long-term consequences of organophosphate poisoning include cognitive and psychomotor deficits, decreased vibration sensitivity, and motor dysfunction, observed as long as 10 years after an acute episode even with mild intoxications by carbamates or organophosphates not requiring hospitalization (Kamel & Hoppin, 2004; Wesseling et al., 2002). Results from

studies of mild acute intoxication by organophosphates vary by country, with positive findings mainly in developing countries where exposure was presumably higher.

Chronic low-level exposure to organophosphates is also accompanied by a higher frequency of self-reported symptoms, such as headache, dizziness, fatigue, insomnia, nausea, chest tightness, and breathing difficulty; impaired neurobehavioral performance, such as confusion, memory loss, and difficulty in concentrating, confirmed by batteries of neurobehavioral tests; mood disorders; and sensory and motor dysfunction, such as weakness, tremors, numbness, tingling, and visual and hearing disturbances (Crawford et al., 2008; Kamel & Hoppin, 2004; Starks et al., 2012A). Such effects are not necessarily related to acetylcholinesterase inhibition (Pope, 1999). The same symptoms have been described in association with exposure to DDT and fumigants, although negative findings have also been reported. Direct measurement of motor or sensory nerve function (e.g., vibration sensitivity, smell, sense of balance from damage to the vestibular function, or visual contrast sensitivity) resulted in inconsistent findings in relation to exposure to organophosphates, DDT, or fumigants (Kamel & Hoppin, 2004). In the Agricultural Health Study, abnormal toe proprioception was associated with six organophosphates, while null associations were observed with electrophysiological tests, hand strength, sway speed, and vibro-tactile thresholds (Starks et al., 2012C).

An extensive body of literature suggests an increased incidence of Parkinson’s disease among pesticide-exposed workers. However, inconsistent findings have also been published, and most studies were unable to identify specific pesticides. Paraquat is known to induce selective damage of the dopaminergic neurons involved in Parkinson’s disease, and experimental animal models and several case reports have described Parkinson’s disease cases in exposed workers. However, exposure to organochlorines, organophospates, carbamates, and dithiocarbamates has also been associated with the risk of Parkinson’s, as has exposure to the herbicide glyphosate and the fungicides diquat and maneb (Kamel & Hoppin, 2004).

A meta-analysis was suggestive for an association of amyotrophic lateral sclerosis (ALS) with pesticide exposure in general; nonsignificantly increased risks were found in association with organochlorine insecticides, pyrethroids, herbicides, and fumigants, and specific compounds such as aldrin, dieldrin, DDT, and toxaphene (Kamel et al., 2012).

The risk of Alzheimer’s disease was also associated with exposure to pesticides in general in two case-control studies. However, no specific compounds have been identified. Such results are also difficult to interpret, as Alzheimer’s disease is characterized by the loss of cholinergic neurons, which is why cholinesterase inhibitors are used in its treatment (Kamel & Hoppin, 2004). Interestingly, in a neurobehavioral study of participants in the Agricultural Health Study, exposure to chlorpyrifos, coumaphos, parathion, phorate, and

tetrachlorvinphos were associated with improvements in verbal learning and memory; coumaphos was associated with better motor speed and visual scanning performance; and parathion with better sustained attention (Starks et al., 2012B).

Liver, Kidney, and Skin Disorders

Acute paraquat poisoning by ingestion is relatively frequently followed by toxic hepatitis after about one week, and about three quarters of patients subsequently develop kidney failure, but no fatalities have been reported (Yang et al., 2012). Half of the patients of intentional or accidental endosulfan poisoning also developed acute toxic hepatitis, along with other complications (such as rhabdomyolisis and refractory epilepsy) (Moon & Chun, 2009); mortality among these latter subjects was around 30%. At high doses, 2,4-D poisoning also damages the liver and kidney and irritates mucous membranes (Garabrant & Philbert, 2002).

Under the conditions of regular occupational exposure, indoor residual spraying with DDT lasting six months was not followed by changes in the serum level of liver enzymes (Bimenya et al., 2010). Among Brazilian residents in an area heavily contaminated with the organochlorine beta-hexachlorocyclohexane (β-HCH), *p,p'*-DDE, and hexachlorobenzene (HCB), serum bilirubin levels were the only parameter significantly associated with β-HCH blood level but not with the blood levels of the other organochlorine pesticides (Freire, Koifman, & Koifman, 2015).

The chlorophenoxy herbicides may cause dermatitis following prolonged skin contact, and prolonged high-level exposure to TCDD causes chloracne (i.e. inclusion cysts, comedones, and pustules) in the face and other parts of the body (Legaspi & Zenz, 1994). Also, numerous compounds have been described as responsible for contact or allergic forms of dermatitis, frequently due to the presence of solvents as co-formulants (Colosio & Rubino, 2015).

Prevention

In the industrialized world, the concern following industrial and community pesticide poisoning events has generated more careful monitoring systems of pesticide production and use, as well as regular monitoring programs of their residual concentration in foodstuffs along the chain of market distribution. Several compounds are simply not allowed at any concentration; therefore, products imported from other geographic areas must be certified as exempt from residuals of these prohibited compounds, and their detection leads to the withdrawal of the contaminated products (Colosio & Rubino, 2015).

date: 29 September 2016

For authorized products, an acceptable daily intake (ADI) and a safety level expressed as acute reference dose (ARD) have been established based on toxicological tests in experimental animals. ADI values concern the regular condition of commercialized foodstuffs, and they are published by the U.S. Department of Agriculture and by DG Sanco in Europe.

In pesticide manufacturing and packaging, basic industrial hygiene principles apply, involving the recognition, constant evaluation, and control of potential health hazards, taking into account all possible routes of exposure. Dermal absorption can be more relevant than inhalation, and therefore proper clothing and gloves need to be worn and cleaned at the facility. Routine washing of hands, face, and neck and a shower at the end of the work shift are advised. To prevent oral consumption, smoking, eating or storing food, cigarettes, or cosmetics in areas with potential pesticide contamination should not be allowed. To prevent inhalation, the use of properly filtered masks while handling pesticides should be enforced, and air-supplied respirator helmets should be provided when necessary to protect against particularly toxic chemicals. Also, routine periodic decontamination should be scheduled for both the work areas and the communal work areas (e.g., such as cafeterias, locker rooms, toilets, offices, and laboratories). Finally, access to potentially contaminated areas in the plant should be limited to trained employees only.

When applying pesticides in a farm, or whatever other use, a written registry should be kept indicating the date, the product used, its amount, crop, and purpose, as well as the names of the applicators and a checklist of the necessary protective equipment.

Preplacement medical examinations are provided to all workers to identify pre-existing illnesses or conditions that might limit fitness to work and to provide a baseline for any subsequent adverse health outcomes. With regard to periodic testing, when organophosphate or carbamate use is expected to occur, the acetyl cholinesterase activity in the serum and the red blood cells should be measured at baseline and periodically reassessed, as well as any other suitable biological monitoring. Such surveillance programs need to be accompanied by an appropriate educational program on the work procedures necessary to minimize exposure, as well as highlighting the importance of wearing personal protective equipment even in unfavorable weather conditions; there should also be discussion of the potential acute and long-term health effects, including infertility and cancer, to be expected if these precautionary measures are not effectively implemented (Legaspi & Zenz, 1994). These educational messages should be updated and re-introduced to the workers at regular intervals.

Implementing preventive action in developing countries is quite another story. In developing countries, farmers rely on information directly from the chemical companies,

regulatory controls are frequently inadequate, and educational programs on the hazards posed by misuse of pesticides are seldom implemented. Wearing protective gear in subequatorial countries is less tolerable because of high temperature and humidity; along with lack of proper education, poverty and the natural environment might contribute to what observed in some surveys in these countries: pesticide applicators seldom use personal protective equipment, and women often participate with their babies attached to their backs (Naidoo et al., 2010; Oesterlund et al., 2014). Consensus exists among international organizations, including the Food and Agriculture Organization of the United Nations (FAO), the World Health Organization, and the World Bank, that hazardous pesticides should not be accessible to farmers unless they are educated about their proper management and storage and the use of protective gear. Therefore, FAO recommends withdrawal of hazardous pesticides from the market in developing countries (FAO, 2013).

A total ban on the use of pesticides is unlikely to happen in the foreseeable future. However, the shift of primary production and the manufacturing industries from developed toward developing countries in the globalized world has generated a differential pattern of exposure circumstances, with a phasing out of persistent organochlorine pesticides and more toxic pesticides in Europe (where stricter regulations are applied) and less restrictive attitudes toward occupational and environmental exposures in the developing world. Organic farming is growing in some parts of the United States and Europe, but it seems unlikely that it will become productive enough to comply with the increasing demand for good quality and safe agricultural products. Specific guidelines to limit the human health and environmental effects of improper handling and disposal of pesticides have been issued, and they are outlined in Table 9.

Table 9. Exposure Control Actions in the Workplace and the General Environment

Subject	Route of exposure	Preventive or corrective action
Applicator	Inhalation	**1.** Mix or load pesticides outdoors, in a well-ventilated area. **2.** Wear appropriate respiratory protective equipment according to pesticide label instructions.
	Ingestion	**1.** Do not eat, drink, or smoke during pesticide handling or application.
	Dermal	**1.** Use personal protective equipment including chemically resistant gloves. **2.** Remove all labor clothes after work; if pesticide-soiled, remove them as soon as possible. **3.** Have a shower immediately after application.
Bystanders and children guardians	Inhalation	**1.** Do not stockpile pesticides. Purchase only what is needed for immediate application. **2.** Dispose of pesticides properly, according to the label directions. **3.** Report any symptoms possibly related to pesticide exposure to your doctor. Remind of the name of the product, the ingredients, and the first aid instructions on the product label. **4.** When someone is applying pesticides outdoors near your home, stay indoors with your children and pets, and keep windows and doors closed.

	Ingestion	**1.** Never store pesticides in cabinets with or near food. **2.** Always store pesticides in their original containers, complete with labels that list ingredients, directions for use, and first aid in case of accidental exposure. **3.** Never transfer pesticides to soft drink bottles or other containers. **4.** Rinse fruits and vegetables with water and, if possible, peel them before eating.
	Dermal	**1.** Do not enter and do not allow children to enter fields, lawns, or confined spaces after pesticides have been applied for the period specified on label instructions. **2.** Encourage family members to remove shoes and labor clothes outside the home or as soon as possible after entering the home. **3.** Vacuum rug and/or clean floors if soiled with pesticides. **4.** Do not store pesticides in living areas or anywhere within the reach of children. Keep all pesticides in a locked cabinet in a well-ventilated utility area or garden shed. **5.** Encourage family members exposed to pesticides to shower as soon as possible after exposure. **6.** When soiled with pesticides, do not allow pets to enter the living areas of the home until cleaned. **7.** Wash clothing soiled with pesticides separately from other laundry.

Regulatory agencies, scientific community and chemical manufacturers	All	**1.** Identify human carcinogens and remove them from the marketplace or greatly limit their use. **2.** Identity the persistence and accumulation potential of pesticides and reduce the use of long-lived pesticides wherever possible. **3.** Identify good pesticide work practices and educate the public in these practices. **4.** Train properly certified professional applicators to handle pesticides in the fields and in major household disinfestations. **5.** Design more effective pesticide containers and application equipment that minimizes pesticide exposure to the applicator and contact to children.

Source: Alavanja, Ross, & Bonner, 2013.

As mentioned in the introduction, research on the health effects of pesticides is particularly complicated because of the complex pattern of applications to different crops and their frequent changes over time to prevent pest resistance. Apart from professional pest control operators, exposures are usually intermittent and poorly documented. Besides, using exposure biomarkers to characterize exposure to the most popular pesticides is difficult, as they tend to have short half-life metabolites, and these might be not specific enough. Therefore, efforts must be made to extend educational programs to the agricultural workers in developing countries, starting from the proper registration of treatments applied, and including a checklist of the precautionary measures to be taken, date, type, and amount of the pesticide to be applied, and pest to be treated. Apart from the educational effectiveness of these procedures in raising concern about the potential health effects of pesticide use, the data gathered would be of utmost importance in replicating findings and confirming associations suggested from small-scale and poorly documented studies. Questions that remain to be solved include the specific long-term effects, including cancer and the neurodegenerative diseases, and the endocrine-disrupting and trans-generational effects associated with specific farm practices and specific pesticides. Education of pesticide applicators and proper data collection for future, better documented studies would be most effective in preventing acute effects and known long-term effects.

References

Acquavella, J. F., Alexander, B. H., Mandel, J. S., Gustin, C., Baker, B., Chapman, P., et al. (2004). Glyphosate biomonitoring for farmers and their families: Results from the farm family exposure study. *Environmental Health Perspectives*, *112*, 321–326.

Alavanja, M. C., Dosemeci, M., Samanic, C., Lubin, J., Lynch, C. F., Knott, C., et al. (2004). Pesticides and lung cancer risk in the agricultural health study cohort. *American Journal of Epidemiology*, *160*, 876–885.

Alavanja, M. C. R., Ross, M. K., & Bonner, M. R. (2013). Increased Cancer Burden Among Pesticide Applicators and Others Due to Pesticide Exposure. *CA-A Cancer Journal for Clinicians*, *63*, 120–142.

Alavanja, M. C. R., Sandler, D. P., McMaster, S. B., Zham, S. H., Mc Donnell, C. J., Lynch, C. F., et al. (1996). The Agricultural Health Study. *Environmental Health Perspectives*, *104*, 362–369.

Alexander, F. E., Patheal, S. L., Biondi, A., Brandalise, S., Cabrera, M. E., Chan, L. C., et al. (2001). Transplacental chemical exposure and risk of infant leukemia with MLL gene fusion. *Cancer Research*, *61*, 2542–2546.

Andreotti, G., Freeman, L. E., Hou, L., Coble, J., Rusiecki, J., Hoppin, J. A., et al. (2009.). Agricultural pesticide use and pancreatic cancer risk in the Agricultural Health Study cohort. *International Journal of Cancer*, *124*, 2495–2500.

Araoud, M. (2011). Biological markers of human exposure to pesticides. In: Stoycheva M. Pesticides in the modern World—Pests Control and Pesticides exposure and toxicity assessment. Rijeka, Croatia: InTech open access publisher. Retrieved from .

Au, W. W., Sierra-Torres, C. H., Cajas-Salazar, N., Shipp, B. K., & Legator, M. S. (1999). Cytogenetic effects from exposure to mixed pesticides and the influence from genetic susceptibility. *Environmental Health Perspectives*, *107*, 501–505.

Augustijn-Beckers, P. W. M., Hornsby, A. G., & Wauchope, R. D. (1994). SCS/ARS/CES pesticide properties database for environmental decision making II. Additional properties. *Reviews of Environmental Contamination and Toxicology*, *137*, 1–82.

Barry, K. H., Koutros, S., Lubin, J. H., Coble, J. B., Barone-Adesi, F., Beane Freeman, L. E., et al. (2012). Methyl bromide exposure and cancer risk in the Agricultural Health Study. *Cancer Causes & Control*, *23*, 807–818.

Bimenya, G. S., Harabulema, M., Okot, J. P., Francis, O., Lugemwa, M., & Okwi, A. L. (2010). Plasma levels of DDT/DDE and liver function in malaria control personnel 6 months after indoor residual spraying with DDT in northern Uganda, 2008. *Sout African Medical Journal*, *100*, 118–121.

Bismuth, C., Hall, A. H., & Wong, A. (1995). Paraquat ingestion exposure. In C. Bismuth & A. H. Hall (Eds.), *Paraquat poisoning: Mechanisms, Prevention, Treatment* (pp. 195–210). New York: Marcel Dekker.

Bonner, M. R., Williams, B. A., Rusiecki, J. A., Blair, A., Beane Freeman, L. E., Hoppin, J. A., et al. (2010). Occupational exposure to terbufos and the incidence of cancer in the Agricultural Health Study. *Cancer Causes & Control*, *21*, 871–877.

Bradberry, S. M., Cage, S. A., Proudfoot, A. T., & Vale, J. A. (2005). Poisoning due to pyrethroids. *Toxicological Reviews*, *24*, 93–106.

Brown, I. (1999). Agriculture (pesticides). In S. S. Sadhra & K. G. Rampal (Eds.), *Occupational health: Risk assessment and management* (pp. 361–368). Oxford: Blackwell Science.

Campagna, M., Satta, G., Fadda, D., Pili, S., & Cocco, P. (2015). Male fertility following occupational exposure to dichlorodiphenyltrichloroethane (DDT). *Environment International*, *77C*, 42–47.

Carson, R. (1962). *Silent spring*. Boston: Houghton Mifflin.

Christensen, C. H., Platz, E. A., Andreotti, G., Blair, A., Hoppin, J. A., Koutros, S., et al. (2010). Coumaphos exposure and incident cancer among male participants in the Agricultural Health Study (AHS). *Environmental Health Perspectives*, *118*, 92–96.

Cocco, P. (2002). On the rumors about the silent spring. Review of the scientific evidence linking occupational and environmental pesticide exposure to endocrine disruption health effects. *Cadernos de Saúde Pública*, *18*, 379–402.

Cocco, P., Fadda, D., & Melis, M. (2006). Reproductive outcomes following environmental exposure to DDT. *Reproductive Toxicology*, *22*, 5–7.

Cocco, P., Satta, G., Dubois, S., Pili, C., Pilleri, M., Zucca, M., et al. (2013). Lymphoma risk and occupational exposure to pesticides: results of the Epilymph study. *Occupational and Environmental Medicine*, *70*, 91–98.

Cole, D. C., Carpio, F., Julian, J., & Léon, N. (1998). Assessment of peripheral nerve function in an Ecuadorian rural population exposed to pesticides. *Journal of Toxicology and Environmental Health, Part A*, *55*, 77–91.

Colosio, C., & Rubino, F. M. (2015). Composti organici: Prodotti fitosanitari. In L. Alessio, G. Franco, & F. Tomei (Eds.), *Trattato di Medicina del Lavoro* (pp. 965–985). Padua, Italy: Piccin Nuova Libraria.

Colt, J. S., Davis, S., Severson, R. K., Lynch, C. F., Cozen, W., Camann, D., et al. (2006). Residential insecticide use and risk of non-Hodgkin's lymphoma. *Cancer Epidemiology Biomarkers & Prevention*, *15*, 251–257.

Corsini, E., Sokooti, M., Galli, C. L., Moretto, A., & Colosio, C. (2013). Pesticide induced immunotoxicity in humans: A comprehensive review of the existing evidence. *Toxicology*, *307*, 123–135

Crawford, J. M., Hoppin, J. A., Alavanja, M. C., Blair, A., Sandler, D. P., & Kamel, F. (2008). Hearing loss among licensed pesticide applicators in the agricultural health study. *Journal of Occupational and Environmental Medicine*, *50*, 817–826.

Danzo, B. J. (1997). Environmental xenobiotics may disrupt normal endocrine function by interfering with the binding of physiological ligands to steroid receptors and binding proteins. *Environmental Health Perspectives*, *105*, 294–301.

De Cock, J., Westveer, K., Heederik, D., Te Velde, E., & Van Kooij, R. (1994). Time to pregnancy and occupational exposure to pesticides in fruit growers in The Netherlands. *Journal of Occupational and Environmental Medicine*, 51, 693–699.

DeCoster, S., & van Larebeke, N. (2012). Endocrine-disrupting chemicals: Associated disorders and mechanisms of action. *Journal of Environmental and Public Health*, 2012, 1–52.

Dennis, L. K., Lynch, C. F., Sandler, D. P., & Alavanja, M. C. (2010). Pesticide use and cutaneous melanoma in pesticide applicators in the agricultural heath study. *Environmental Health Perspectives*, *118*, 812–817.

Bundesinstitut fur Risikobewertung. (2011). Joint DE-UK position paper. Regulatory definition of an endocrine disrupter in relation to potential threat to human health. Proposal applicable in the regulatory context of plant protection products, biocidial products, and chemicals targeted within REACH. Retrieved from .

Duffus, J. H., & Worth, H. G. J. (Eds). (2006). *Fundamental toxicology*. Cambridge, U.K.: The Royal Society of Chemistry.

Eddleston, M., Karalliedde, L., Buckley, N., Fernando, R., Hutchinson, G., Isbister, G., et al. (2002). Pesticide poisoning in the developing world—a minimum pesticides list. *Lancet*, *360*, 1163–1167.

U.S. Environmental Protection Agency. (2016). *Pesticides*. Retrieved from .

The European Commission. (2002). Effects of in utero exposure to ionising radiation during the early phases of pregnancy. Proceedings of a scientific seminar, Luxembourg, 5.11.2001. Luxembourg: Office for Official Publications of the European Communities.

Ewence, A., Brescia, S., Johnson, I., & Rumsby, P. C. (2015). An approach to the identification and regulation of endocrine disrupting pesticides. *Food and Chemical Toxicology*, *78*, 214–220.

Ewence, A., Rumsby, P., & Johnson, I. (2013). *Extended impact assessment study of the human health and environmental criteria for endocrine disrupting substances proposed by HSE, CRD*. Swindon, Wiltshire, U.K.: WRc plc.

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: Atrazine. Retrieved from .

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: 2,4 D. Retrieved from .

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: DDT (dichlorodiphenyl trichloroethane). Retrieved from .

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: Glyphosate. Retrieved from .

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: Paraquat. Retrieved from .

Extoxnet, the Extension Toxicology Network. (2008). Pesticide information profile: Thiram. Retrieved from .

Food and Agriculture Organization of the United Nations. (2013). *Highly hazardous pesticides should be phased out in developing countries*. Retrieved from .

Fishel, F. M. (2013). *Pest management and pesticides: A historical perspective* (revised version). Gainesville: Institute of Food and Agricultural Sciences, Agronomy Department, Florida Cooperative Extension Service University of Florida.

Freeman, L. E., Rusiecki, J. A., Hoppin, J. A., Lubin, J. H., Koutros, S., Andreotti, G., et al. (2011). Atrazine and cancer incidence among pesticide applicators in the agricultural health study (1994–2007). *Environmental Health Perspectives*, *119*, 1253–1259.

Freire, C., Koifman, R. J., & Koifman, S. (2015). Hematological and hepatic alterations in Brazilian population heavily exposed to organochlorine pesticides. *Journal of Toxicology and Environmental Health, Part A*, *78*, 534–548.

Garabrant, D. H., & Philbert, M. A. (2002). Review of 2,4-dichlorophenoxyacetic acid (2,4-D) epidemiology and toxicology. *Critical Reviews in Toxicology*, *32*, 233–257.

Gessner, P. K., & Gessner, T. (1992). *Disulfiram and its metabolite diethyldithiocarbamate. Pharmacology and status in the treatment of alcoholism, HIV infections, AIDS and heavy metal toxicity*. Padstow, Cornwall, U.K.: TJ Press.

Glyphosate Task Force. (2013). Glyphosate mechanism of action. Retrieved from .

Grube, A., Donaldson, D., Kiely, T., & Wu, L. (2011). Pesticides industry sales and usage. 2006 and 2007 market estimates. Washington, DC: U.S. Environmental Protection Agency.

Guyton, K. Z., Loomis, D., Grosse, Y., El Ghissassi, F., Benbrahim-Tallaa, L., Guha, N., et al. (2015). Carcinogenicity of tetrachlorvinphos, parathion, malathion, diazinon, and glyphosate. *Lancet Oncology*, *16*, 490–491.

Hayes, W. J., & Laws, E. R. (Eds.). (1990). *Handbook of pesticide toxicology*, Vol. 3, *Classes of pesticides*. New York: Academic Press.

Henneberger, P. K., Liang, X., London, S. J., Umbach, D. M., Sandler, D. P., & Hoppin, J. A. (2013). Exacerbation of symptoms in agricultural pesticide applicators with asthma. *International Archives of Occupational and Environmental Health*, 87, 423–432.

Hoppin, J. A., London, S. J., Linch, C. F., & Alavanja, M. C. (2006). Pesticides associated with wheeze among commercial pesticide applicators in the Agricultural Health Study. *American Journal of Epidemiology*, *163*, 1129–1137.

Hoppin, J. A., Umbach, D. M., London, S. J., Alavanja, M. C., & Sandler, D. P. (2002). Chemical predictors of wheeze among farmer pesticide applicators in the Agricultural Health Study. *American Journal of Respiratory and Ciritical Care Medicine*, *165*, 683–689.

Hoppin, J. A., Umbach, D. M., London, S. J., Henneberger, P. K., Kullman, G. J., Alavanja, M. C., et al. (2008). Pesticides and atopic and nonatopic asthma among farm women in

the Agricultural Health Study. *American Journal of Respiratory and Ciritical Care Medicine*, *177*, 11–18.

Hoppin, J. A., Umbach, D. M., London, S. J., Henneberger, P. K., Kullman, G. J., Coble, J., et al. (2009). Pesticide use and adult-onset asthma among male farmers in the Agricultural Health Study. *European Respiratory Journal*, *34*, 1296–1303.

IARC. (1986). Some halogenated hydrocarbons and pesticide exposures. In *Chlorophenoxy herbicides (occupational exposure to)*. Vol. 41. IARC Monographs on the Evaluation of the Carcinogenic Risk to Humans (pp. 357–406). Lyon, France: International Agency for Research on Cancer.

IARC. (1991). Some halogenated hydrocarbons and pesticide exposures. In *Occupational exposures in insecticide application and some pesticides* (Vol. 53) (pp. 45–535). IARC Monographs on the Evaluation of the Carcinogenic Risk to Humans. Lyon, France: International Agency for Research on Cancer.

IARC. (2015). Some halogenated hydrocarbons and pesticide exposures. Vol. 112. Some organophosphate insecticides and herbicides: diazinon, glyphosate, malathion, parathion, and tetrachlorvinphos. IARC Monographs on the Evaluation of the Carcinogenic Risk to Humans. Lyon, France: International Agency for Research on Cancer.

International Program on Chemical Safety. (1998). Chemical Safety Information from Intergovernmental Organizations. Poisons Information Monograph (Group Monograph) G001 Chemicals. Retrieved from .

Joffe, M. (1997). Time to pregnancy: A measure of reproductive function in either sex. *Occupational and Environmental Medicine*, *54*, 289–295.

Kamel, F., & Hoppin, J. A. (2004). Association of pesticide exposure with neurologic dysfunction and disease. *Environmental Health Perspectives*, *112*, 950–958.

Kamel, F., Umbach, D. M., Bedlack, R. S., Richards, M., Watson, M., Alavanja, M. C., et al. (2012). Pesticide exposure and amyotrophic lateral sclerosis. *Neurotoxicology*, *33*, 457–462.

Kang, D., Park, S. K., Beane-Freeman, L., Lynch, C. F., Knott, C. E., Sandler, D. P., et al. (2008). Cancer incidence among pesticide applicators exposed to trifluralin in the Agricultural Health Study. *Environmental Research*, *107*, 271–276.

Keifer, M., & Mahurin, R. (1997). Chronic neurologic effects of pesticide overexposure. *Occupational Medicine*, *12*, 291–304.

Kelce, W. R., Stone, C. R., Laws, S. C., Earl-Gray, L., Kemppainen, J. A., & Wilson, E. M. (1995). Persistent DDT metabolite p,p’-DDE is a potent androgen receptor antagonist. *Nature*, *375*, 581–585.

Koutros, S., Silverman, D. T., Alavanja, M. C., Andreotti, G., Lerro, C. C., Heltshe, S., et al. (2015). Occupational exposure to pesticides and bladder cancer risk. *International Journal of Epidemiology, pii*, dyv195.

Legaspi, J. A., & Zenz, C. (1994). Occupational health aspects of pesticides. Clinical and hygienic principles. In C. Zenz, O. B. Dickerson & E.P. Horvath (Eds.), *Occupational medicine* (3d ed.) (pp. 617–653). St. Louis, MO: Mosby Year Book.

Li, Q. (2007). New mechanism of organophosphorus pesticide-induced immunotoxicity. *Journal of Nippon Medical School*, *74*, 92–105.

Lee, W. J., Blair, A., Hoppin, J. A., Lubin, J. H., Rusiecki, J. A., Sandler, D. P., et al. (2004). Cancer incidence among pesticide applicators exposed to chlorpyrifos in the Agricultural Health Study. *Journal of the National Cancer Institute*, *96*, 1781–1789.

Lee, W. J., Sandler, D. P., Blair, A., Samanic, C., Cross, A. J., & Alavanja, M. C. (2007). Pesticide use and colorectal cancer risk in the Agricultural Health Study. *International Journal of Cancer*, *121*, 339–346.

Loomis, D., Guyton, K., Grosse, Y., El Ghissassi, F., Bouvard, V., Benbrahim-Tallaa, L., et al. (2015). Carcinogenicity of lindane, DDT, and 2,4-dichlorophenoxyacetic acid. *Lancet Oncology*, *16*, 891–892.

Lynch, S. M., Mahajan, R., Beane Freeman, L. E., Hoppin, J. A., & Alavanja, M. C. (2009). Cancer incidence among pesticide applicators exposed to butylate in the Agricultural Health Study (AHS). *Environmental Research*, *109*, 860–868.

Mahajan, R., Bonner, M. R., Hoppin, J. A., & Alavanja, M. C. (2006). Phorate exposure and incidence of cancer in the agricultural health study. *Environmental Health Perspectives*, *114*, 1205–1209.

Majewski, M. S., & Capel, P. D. (1995). *Pesticides in the atmosphere; distribution, trends, and governing factors.* Chelsea, MI: Ann Arbor Press.

Mamane, A., Baldi, I., Tessier, J.-F., Raherison, C., & Bouvier, G. (2015). Occupational exposure to pesticides and respiratory health. *European Respiratory Review*, *24*, 306–319.

McKelvey, W., Jacobson, J. B., Kass, D., Barr, D. B., Davis, M., Calafat, A. M., et al. (2013). Population-based biomonitoring of exposure to organophosphate and pyrethroid pesticides in New York City. *Environmental Health Perspectives*, *121*, 1349–1356.

Majewski, M. S., & Capel, P. D. (1995). *Pesticides in the atmosphere; distribution, trends, and governing factors.* Chelsea, MI: Ann Arbor Press.

Miyamoto, J. (1976). Degradation, metabolism and toxicity of synthetic pyrethroids. *Environmental Health Perspectives*, *14*, 15–28.

Moon, J. M., & Chun, B. J. (2009). Acute endosulfan poisoning: A retrospective study. *Human & Experimental Toxicology*, *28*, 309–316.

Naidoo, S., London, L., Rother, H. A., Burdorf, A., Naidoo, R. N., Kromhout, H. (2010). Pesticide safety training and practices in women working in small-scale agriculture in South Africa. *Occupational and Environmental Medicine*, *67*, 823–828.

Oesterlund, A. H., Thomsen, J. F., Sekimpi, D. K., Maziina, J., Racheal, A., Jørs, E. (2014). Pesticide knowledge, practice and attitude and how it affects the health of small-scale farmers in Uganda: A cross-sectional study. *African Health Sciences*, *14*, 420–433.

Roberts, J. R., & Routt Reigart, J. (2013). Recognition and management of pesticide poisoning (6th ed.). Washington, DC: U.S. Environmental Protection Agency.

Robison, A. K., Sirbasku, D. A., & Stancel, G. M. (1985). DDT supports the growth of an estrogen-responsive tumor. *Toxicology Letters*, *27*, 109–113.

Park, E. K., Duarte Tagles, H., Gee, S. J., Hammock, B. D., Lee, K., & Schenker, M. B. (2008). Recruiting strategy and 24-hour biomonitoring of paraquat in agricultural workers. *Joutnal of Agromedicine*, *13*, 207–217.

Pillay, V. V. (2013). *Modern medical toxicology* (4th ed.). New Delhi: Jaypee Brothers Medical Publishers.

Pope, C. N. (1999). Organophosphorus pesticides: Do they all have the same mechanism of toxicity? *Journal of Toxicology and Environmental Health, Part B Crit Rev*, *2*, 161–181.

Potashnik, G., & Porath, A. (1995). Dibromochloropropane (DBCP): A 17-year reassessment of testicular function and reproductive performance. *Journal of Occupational and Environmental Medicine*, 37, 1287–1292.

Samanic, C., Rusiecki, J., Dosemeci, M., Hou, L., Hoppin, J. A., Sandler, D. P., et al. (2006). Cancer incidence among pesticide applicators exposed to dicamba in the agricultural health study. *Environmental Health Perspectives*, *114*, 1521–1526.

Schechter, A. (Ed.). (1994). *Dioxins and health*. New York: Plenum Press.

SeedQuest—Global Information Services for Seed Professionals. (2016). Market data and statistics. Retrieved from .

Silver, S. R., Bertke, S. J., Hines, C. J., Alavanja, M. C., Hoppin, J. A., Lubin, J. H., et al. (2015). Cancer incidence and metolachlor use in the Agricultural Health Study: An update. *International Journal of Cancer*, 137, 2630–2643.

Smith, A. E., & Secoy, D. M. (1975). Forerunners of pesticides in classical Greece and Rome. *Journal of Agricultural and Food Chemistry 23*, 1050–1055.

Smith, L. L. (1987). Mechanism of paraquat toxicity in lung and its relevance to treatment. *Human Toxicology*, *6*, 31–36.

Soto, A. M., Sonnenschein, C., Chung, K. L., Fernandez, M. F., Olea, N., & Serrano, F. O. (1995). The E-screen assay as a tool to identify estrogens: An update on estrogenic environmental pollutants. *Environmental Health Perspectives*, *103*, 113–122.

Starks, S. E., Gerr, F., Kamel, F., Lynch, C. F., Alavanja, M. C., Sandler, D. P., et al. (2012a). High pesticide exposure events and central nervous system function among pesticide applicators in the Agricultural Health Study. *International Archives of Occupational and Environmental Health*, *85*, 505–515.

Starks, S. E., Gerr, F., Kamel, F., Lynch, C. F., Jones, M. P., Alavanja, M. C., et al. (2012b). Neurobehavioral function and organophosphate insecticide use among pesticide applicators in the Agricultural Health Study. *Neurotoxicology and Teratology*, *34*, 168–176.

Starks, S. E., Hoppin, J. A., Kamel, F., Lynch, C. F., Jones, M. P., Alavanja, M. C., et al. (2012c). Peripheral nervous system function and organophosphate pesticide use among licensed pesticide applicators in the Agricultural Health Study. *Environmental Health Perspectives*, *120*, 515–520.

Sudakin, D. L., & Stone, D. L. (2011). Dialkyl phosphates as biomarkers of organophosphates: The current divide between epidemiology and clinical toxicology. *Clinical Toxicology (Philadelphia)*, *49*, 771–781.

Taylor, E. L., Holley, A. G., & Kirk, M. (2007). Pesticide development: a brief look at the history. Southern Regional Extension Forestry. Retrieved from .

Thompson, C. M., Prins, J. M., & George, K. M. (2010). Mass spectrometric analyses of organophosphate insecticide oxon protein adducts. *Environmental Health Perspectives*, *118*, 11–19.

United Nations Economic Commission for Europe. (2009). Globally harmonized system of Classification and Labelling of Chemicals (GHS). Retrieved from .

U.S. Geological Survey. (2006). Pesticides in nation’s streams and groundwater, 1992–2001. Sacramento, CA: U.S. Department of the Interior.

U.S. Environmental Protection Agency. (1975). Regulatory history. A brief survey (to 1975). Retrieved from

U.S. Environmental Protection Agency. (2008). Triazole metabolites. Dietary exposure and risk assessment. Retrieved from .

U.S. Geological Survey. (2016). Pesticide National Synthesis Project. Pesticides use maps. Retrieved from .

Ulrich, E. M., Caperell-Grant, A., Jung, S.-H., Hites, R. A., & Bigsby, R. M. (2000). Environmentally relevant xenoestrogen tissue concentrations correlated to biological responses in mice. *Environmental Health Perspectives*, *108*, 973–977.

Vom Saal, F. S., Nagel, S., Palanza, P., Boechler, M., Parmigiani, S., & Welshons, W. V. (1995). Estrogenic pesticides: binding relative to estradiol in MCF-7 cells and effects of exposure during fetal life on subsequent territorial behavior in male mice. *Toxicology Letters*, *77*, 343–350.

Vonier, P. M., Crain, D. A., McLachlan, J. A., Guillette, L. J., Jr., & Arnold, S. F. (1996). Interaction of environmental chemicals with estrogen and progesterone receptors from the oviduct of the American alligator. *Environmental Health Perspectives*, *104*, 1328–1322.

Weed Science Society of America. (1994). *Herbicide Handbook*. 7th ed. Champaign, IL: WSSA.

Wesseling, C., Keifer, M., Ahlbom, A., McConnell, R., Moon, J., Rosenstock, L., et al. (2002). Long-term neurobehavioral effects of mild poisonings with organophosphate and nmethyl carbamate pesticides among banana workers. *International Journal of Occupational and Environmental Health*, *8*, 27–34.

WHO International Program on Chemical Safety. (2002). *Global assessment of the state-of-the-science of endocrine disruptors.* Geneva, Switzerland: World Health Organization.

WHO International Program on Chemical Safety. (2010). The WHO recommended classification of pesticides by hazard and guidelines to classification: 2009. Geneva, Switzerland: World Health Organization.

Whorton, D., Millby, T. H., Krauss, R. M., & Stubbs, H. A. (1979). Testicular function in DBCP exposed pesticide workers. *Journal of Occupational Medicine*, *21*, 161–166.

Yang, C. J., Lin, J. L., Lin-Tan, D. T., Weng, C. H., Hsu, C. W., Lee, S. Y., et al. (2012). Spectrum of toxic hepatitis following intentional paraquat ingestion: analysis of 187 cases. *Liver International*, *32*, 1400–1406.

Zarn, J. A., Bruschweiler, B. J., & Schlatter, J. R. (2003). Azole fungicides affect mammalian steroidogenesis by inhibiting sterol 14 alpha-demethylase and aromatase. *Environmental Health Perspectives*, 111, 255–262.

Pierluigi Cocco

Public Health and Clinical Molecular Medicine, University of Cagliari