



# Pesticide use in Rice cultivation in Tarapoto, Peru

Pesticide residues in blood of farmers, usage behaviour, and health care practices



Master's thesis 20 p (Examensarbete)

Gun Lange

Department of Environmental Assessment Swedish University of Agricultural Sciences Box 7050 SE-750 07 Uppsala Sweden

Supervisor: Henrik Kylin

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#### Resumen

Se realizó un estudio sobre el uso de plaguicidas en los cultivos de arroz en la ciudad de, Tarapoto, perteneciente al Departamento de San Martín, Perú. Durante los 80'se construyó en Tarapoto un canal de irrigación y fue el punto de partida para cultivar arroz bajo riego. De la mano con el riego fueron introducidos los plaguicidas y el uso de estos es necesario para conseguir un rendimiento rentables.

Grandes cantidades de plaugicidas han sido usados y entre ellos son varías classificados según la Organización Mundial de la Salud (WHO), como Extremadamente (Ia) y Altamente peligrosos (Ib). Durante el estudio de investigación no había ningún signo del uso de COP's en la región.

El objetivo del estudio fue investigar el uso de plaugicidas e identificar las vías en el medio ambiente de diferente tipos de plagucidas. Este estudio es una parte de un proyecto interdiciplinario mayor, en que aquel estudio está orientado en medir concentraciónes de diferentes tipos de plaguicidas en la sangre de agricultores expuestos.

Fueron tomados muestras de sangre de 24 agricultores. Despues extracción y pasos de limpieza se analizó la sangre a través de GC-ECD (Cromatografía de Gases/ Detector de Captura de Electrones). 21 plaguicidas diferentes incluido sus metabolitos fueron las referencias. El residuo de la plaguicida más detectado fue p,p-DDE. Concentraciónes bajos de  $\alpha$ -HCH,  $\gamma$ -HCH, HCB y restos de Halogenated biphenyls (PCB) y DDT fueron detectados en varías de las muestras. Una sustancia desconocida fue detectado en todas las muestras y fue identificada en MS (espectros de masas) como Pentachloroanisole (PCA). Las muestras de sangre tienen concentraciónes más altas de p,p-DDE comparado con estudios hechos en Europa, pero las concentraciónes son levemente menores comparado con estudios realizados en Nicaragua y México. Las concentraciónes de HCB son equivalentes o menores comparado con Europa y America Latina, pero las concentraciones de  $\gamma$ -HCH son levemente mayores comparado con Nicaragua y México pero más alto comparado con Europa.

Durante la toma de muestras de sangre, se realizó una entrevista corta con los agricultores, el objetivo fue conseguir información sobre su uso y manejo de los plaguicidas. Como resultado de esta se obtuvo que la mayoria de las plaguicidas son guardados en la casa, los agricultores fumugan con una mochila motofumigadora con poco o nada de equipo de protección, l. La mayoría de los agricultores han tenido síntomas de intoxicación por plaugicidasa lo menos en una oportunidad, pero pocos de ellos habían concurrido al hospital por tratamiento, en su reemplazo el "tratamiento" más común es beber diferentes liquidos mezclados como leche con gaseosa.

A su vez, 7 entrevistas más profundas con agricultores fueron realizadas. El objetivo fue tener más conocimiento sobre el proceso del cultivo del arroz y entender el uso de plaguicidas. La mayoría de las entrevistas fueron realizadas en el campo, que dio la oportunidad de observar mientras los agricultores estaban trabajando.

Un encuesta fue distrubuido entre 3 Centros de salud y fue llenado por 15 empleados. Los objetivos fueron tener información sobre los tratamientos en los Centros de salud entre agricultores y otras personas intoxicadas por plaguicidas, y tambien saber el conocimiento entre los empleados del hospital como tratar síntomas. Según la encuesta los empleados del hospital tienen experiencia de tratar pacientes intoxicados por plaugicidas, pero pocos de ellos habían tenido entrenamiento profesional para hacerlo. Parece que hay un procedimiento de norma para el tratamiento de casos de intoxicación por plaguicidas en uno de los Centros de salud, aunque no todos los empleados encuestados tenian conocimiento de aquello.

Palabras claves: Residuos de plaguicidas, Sangre humana, GC-ECD, p,p-DDE, Pentachloro-

anisole, arroz, mochila motofumigadora, manejo de p plaguicidas, Tarapoto, Perú	laguicidas, tratamiento de intoxicaciones por

#### **Abstract**

A study of the pesticide use in the rice cultivation in Tarapoto, the largest town in the department of San Martin situated in the northeast part of Peru, was performed. In Tarapoto, an irrigation canal was built in the 1980s, which gave the farmers the prerequisite to cultivate irrigated rice. The pesticides were introduced around the same time and have become a necessity to obtain a good and profitable yield.

Large quantities of pesticides are used and many of them are classified as Extremely hazardous (Ia) or Highly hazardous (Ib) according to the WHO. During the study no traces of the use of POPs was found nor observed in the region.

The aim of the study was to investigate the pesticide use, and identify the environmental pathways of different pesticides. It was part of a larger interdisciplinary project where this study in particular focused on the concentrations of different pesticides in blood from exposed people.

Blood samples from 24 farmers were analysed on GC-ECD after extraction and clean-up. A screening of 21 different pesticides or metabolites was made and the most abundant pesticide residue detected was p,p-DDE. Lower concentrations of  $\alpha$ -HCH,  $\gamma$ -HCH, HCB and traces of halogenated biphenyls (PCB) and DDT were detected in many of the samples. An unknown substance detected in all the samples was identified as Pentachloroanisole (PCA) on the MS. The samples show higher concentrations of p,p-DDE than in some European studies but slightly lower than studies from Nicaragua and Mexico. The concentrations of HCB are equivalent or even lower than in the studies from Europe and Latin America whereas the concentrations of  $\gamma$ -HCH seemed to be somewhat higher than in the studies from Nicaragua and Mexico and higher than in a European study.

During the blood sampling, short interviews were made with the farmers to obtain some knowledge about how they handle the pesticides. The pesticides were mainly stored at home. The farmers apply the pesticides using back pack sprayers wearing little or no protective clothing at all. Many of the farmers had felt symptoms of poisoning due to pesticides, but they seldom visited the hospitals with them. Instead the most common cure was to drink different blends, like milk with soda.

Another 7 deep interviews with farmers were made to obtain a better understanding of the whole cultivation process and thus the reason for the pesticide use. Often the interviews were made out on the field, which gave the possibility of observing the farmers while they were working.

A questionnaire was distributed at three different health care centres and filled out by 15 employees. The purpose was to see how farmers and other people with poisoning due to pesticides were treated and also to look at the knowledge and training amongst the staff in how to treat them. Even though many of the personnel had experience in treating patients with poisoning due to pesticides, they were hardly given any professional training in treating them. There did seem to be a standard working procedure for treating these patients in one of the three hospitals, however not all of the personnel working there were aware of this matter.

**Keywords:** Pesticide residues, Human blood, GC-ECD, p,p-DDE, Pentachloroanisole, Rice, Bag-pack sprayer, Handle of pesticides, Treating poisoning due to pesticides, Tarapoto, Peru

## **Abbreviations**

a.i. Active ingredient

**b.p.** Blood plasma

C The river of Cumbaza

**CARITAS** Catholic aid organization

CIP Centro Internacional de la Papa/International potato center

**DDA** 2.2-bis(4-chlorophenyl)acetic acid (degradation product of DDT)

**DDD** 1.1-dichloro-2.2-bis(p-chlorophenyl)ethylene (degradation product of DDT)

**DDE** 1,1-dichloro-2,2-bis(p-chlorophenyl)ethane (degradation product to DDT)

**DDT** 1,1,1-trichloro-2,2-bis(p-chlorophenyl)ethane (organochlorine insecticide)

**EA** Environmental Agency

FAO Food and Agriculture Organization of the United Nations

GC-ECD Gas chromatograph with electron-capture detector

**GDP** Gross Domestic Product

**HCB** Hexachlorbenzene

**HCH** Hexachlorocyclohexane

**IAAP** Instituto de Investigaciónes de la Amazonia Peruana/Institute for research of the Amazonian Peru

IMA Institutionen för Miljöanalys/Department of Environmental Assessment, SLU

IMK Institutionen för Miljökemi/Department of Environmental chemistry, SU

**INEI** Instituto Nacional de Estadística e Informática/National Institute for statistics and information

INIA Instituto Nacional de Investigación Agraria/National Institute for Agricultural research

MFS Minor Field Study

**OSPAR** Oslo and Paris commission (OSPAR commison for the protection of the Marine environment of the North-East Atlantic)

**PAN** Pesticide Action Network

**PCP** Pentachlorphenole

**PCA** Pentachloroanisole

POP/COP Persistent Organic Pollutant/Contaminantes Orgánicos Persistentes

**RAAA** Red de acción en Alternatives al uso de Agroquimicos/Action network for alternatives to the use of Agrochemicals

SENASA Servicio Nacional de Sanidad Agraria/National Agricultural Ministery

**SIDA/ASDI** Swedish International Development Cooperation Agency/Agencia Sueca de Cooperación para el Desarollo

SLU Sveriges Lantbruksuniversitet/Swedish University of Agricultural Sciences

SU Stockholms universitet/Stockholm University

UNSM Universidad Naciónal de San Martin/University of San Martin

# Spanish expressions

Almácigo nursery for the rice plants

**Agricultor** Farmer

Arroz rice

Bomba mochila backpack sprayer, manually operated

**Cabrilla** Cultivation practice, were the farmer receive a second yield, by letting the rice regrow after harvesting

Campaña cultivation season

Chakra The farmer's piece of farmland

Motofumigadora bagpack sprayer, motor operated

**Peon** Farmhand

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# 1. Aim and objectives

The aim of this Minor Field Study, MFS, was to investigate the use and identify the environmental pathways of different pesticides in the rice production in the district of Morales and Cacatachi, Tarapoto, San Martin, Peru.

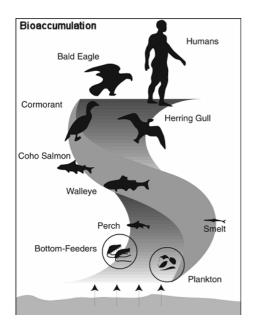
This study was a part of a larger interdisciplinary project that was performed together with two other students from the disciplines of soil science and business economics from SLU and Södertörn University. The goal was, to achieve a more reliable picture of the actual situation for all the parties involved and try to show the complexity of the problem concerning pesticide use. This study focuses in particular on the concentrations of different pesticides in blood from exposed people.

## 2. Introduction

## 2.1. Ecotoxicological concepts

A substance has to be bioavailable, be able to be absorbed by an organism, to be toxic to it. This means that it has to have a chemical form that is suitable for uptake. Other parameters that influence the bioavailability are pH, temperature, concentration, time and frequency of exposure (Fent, 1998).

If a substance is bioavailable it can also accumulate in an organism and hence provide a higher concentration in the animal then in its surroundings, one then talks about bioaccumulation (Bernes, 1998). A substance is said to achieve a biomagnification if it is concentrated in the food chain. This is differential for chemicals with high  $K_{ow}$  that are stored in the fatty tissue of the organism.



The toxicity depends on to what extent an organism is exposed to a substance. The acute toxicity is the toxicity after a single exposure or a very short period of exposure and where you often can see the effects of the substance at once. Chronic toxicity, on the other hand, occurs when the organism is exposed over a longer period of time and where the effects develop slowly. Some examples of chronic toxicity can be carcinogenicity, mutagenicity, teratogenicity, neurotoxicity and so on. Acute and chronic toxicity are both dose-dependent, but the effects can, as mentioned above, be quite different. It is important to consider them both when doing a risk assessment (Fent 1998).

**Fig. 1.** Bioaccumulation in the food chain<sup>1</sup>.

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<sup>&</sup>lt;sup>1</sup> Department of natural resources of Wisconsin http://www.dnr.state.wi.us/org/water/wm/foxriver/whatarepcbs.html

**Table 1.** Important concepts in the Ecotoxicology (Fent, 1998)

LD <sub>50</sub> / LC <sub>50</sub>	Median Lethal Dose resp. Concentration indicates at which concentration half of the animals in the toxicity test die. $LD_{50}$ is a measure for the acute
	toxicity for mammals and humans whereas LC <sub>50</sub> is a measure for aquatic
	organisms.
Kow (Log Kow	The partition coefficient between the concentrations of a substance in
/Log P)	octanol divided by the concentration in water. The higher the $K_{ow}$ the
	more the substance tends to bioaccumulate. This means that a high $K_{ow}$
	leads to low water solubility. Often Log K <sub>ow</sub> is used since it is direct
	proportional to the bioconcentration factor, BCF.
Bioconcentration	This is the ratio between the concentrations of a chemical substance in an
factor, BCF	organism in comparison to the ambient medium.
NOEL	No Observed Effect Level, the highest dose at which no effect is
	observed.
LOEL	Lowest Observed Effect Level.
EC <sub>50</sub>	Concentration where the effect is 50 %.

Different substances are often used in combination and they can have different effects in combination, additive, synergetic or antagonistic. An additive effect is simply if the effects of the individual substances work together so that the total effect is the sum of the effects of the individual of the individual substances. If the effect is synergistic, the effects of the two substances are greater than the sum of the individual effects. And if the effects are antagonistic the total effect is less than what is expected from the individual effects (Bernes, 1998).

Another concept amongst substances and their effects is the potency. This is a measure of the strength of a substance compared to other substances. The more potent the substance, the lower concentration it takes to give an effect; the less potent the substance, the higher concentration it takes to achieve an effect.

There is also a possibility that a substance can be bioactivated. This means that a substance that from the beginning might be harmless can become toxic inside the body after it has been transformed due to metabolism (Fent, 1998).

#### 2.2 Pesticides in the environment

Pesticides are chemicals designed to kill or control unwanted organisms. These can be roughly divided based on the organisms they are intended to kill. The three biggest groups are insecticides for insects, herbicides for weeds, and fungicides for fungal deceases. Many different chemicals are used and there are over 800 active ingredients in over ten thousands of formulations that are sold over the world (PAN 2005).

Agricultural pesticides reach the environment after application by the farmers. The proportion that is not sorbed by the crop may be washed away by rain or irrigation water, transported by the wind, or adsorbed by the soil. From the soil a pesticide may leak to the groundwater if it is not bound to tightly to the soil particles. Depending on the physico-chemical properties of the pesticide, it is preferentially partitioned into different compartments in the environment. As examples the herbicide 2,4-D, which is a salt of carboxylic acid, is highly water-soluble whereas the insecticide DDT, which is poorly water soluble, sorbs to the organic matter in the soil, and Paraquat, although ionized, strongly binds to the clay mineral fraction of the soil.

Fat-soluble organochlorines are, if taken up into animals stored in the fatty tissue (Wheeler 2002).

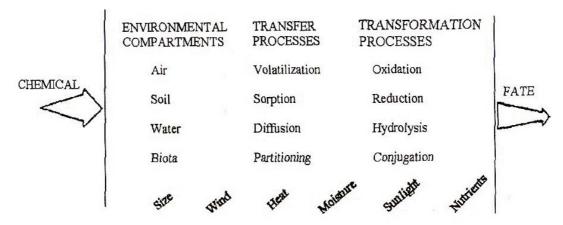


Fig. 2 A schematic of the components of the fate of chemicals in the environment (Wheeler, 2002).

## 2.3 Health effects of pesticides

#### 2.3.1 General

Pesticides can enter humans through three different ways, dermal via skin and eyes, orally through ingestion, and through inhalation. The toxins may have different effects depending their way of entry.

Direct poisoning by pesticides may show symptoms like vomiting, sweating, or pin-point pupils. They might also cause "lighter" symptoms like nausea, headache, weakness, dizziness, itching and so on<sup>2</sup>.

There are other especially chronic, negative health effects that are linked to the use of pesticides. Some pesticides are suspected to be or are recognized as mutagenic, carcinogenic, teratogenic, endocrine disruptors, or immunosuppressive in humans. As example there are several studies that have shown a correlation between pesticide exposure and Parkinson's disease (Kamel, 2004) and the European Union, EU, has identified over 60 additional active ingredients as possible endocrine disruptors (PAN, 2005)<sup>3</sup>.

## 2.3.2 Specific

Paraquat is the leading cause of pesticide-related poisoning in several countries (e.g., Costa Rica and South Korea) and constitutes a great risk to its user if not proper protective equipment is used, which often is the case in tropical countries<sup>4</sup>. Paraquat poisoning is characterized by multiple organ failure, most frequently kidney, liver and lung; and occasionally, the central nervous system, heart and suprarenal glands (Wessling, 1997). There are also many cases of poisoning due to organophosphate or carbamate insecticides. Both chemical groups affect humans by inhibiting acetyl cholinesterase, an enzyme essential to proper functioning of the nervous system. The effects of organophosphate and carabamate

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<sup>&</sup>lt;sup>2</sup> Univertsity of Nebraska http://ianrpubs.unl.edu/Pesticides/ec2505.htm#roe

<sup>&</sup>lt;sup>3</sup> However, four official sources (EU, German EA, U.K. EA and OSPAR) have eventually only agreed on four pesticides being endocrine disruptor; Atrazine, DDT, Lindane and Tributyltin

<sup>&</sup>lt;sup>4</sup> Press Release 8/12 2005 *Paraquat: Unacceptable health risks for users* PAN U.K, BD (Berne Declaration), PANAP (Pesticide Action Network Asian and the Pacific)

poisoning and are recalled using the acronym "SLUDGE", for salivation, lacrimation, urination, defecation, gastrointestinal upset and vomiting<sup>5</sup>.

Pyrethroids, like the insecticide cypermetrin, are commonly used insecticides but there are few cases of poisoning due to them. However, dermal contact with the substance may lead to skin irritations like stinging, burning and itching. Some herbicides like glyphosate also tend to cause the same effect<sup>6</sup>.

PAN, Pesticide Action Network, has categorization a group of toxic and particularly harmful chemicals that they call the "Dirty Dozen". This list was launched in 1985. Today, the list contains 18 different chemicals; including the 9 persistent organic pollutants, POPs, that are included in the Stockholm convention<sup>7</sup> (See Appendix I for more conventions concerning pesticides); Aldrin, Chlordane, DDT, Dieldrin, Endrine, Heptachlor, Hexaclorbenzen, Mirex (not included in the Dirty Dozen) and Toxaphene<sup>8</sup> and an additional 10 pesticides; Aldicarb, Chlordimeform, DBCPT, EDB, HCH, Lindane, Paraquat, Methyl Parathion, PCP and 2,4,5-T. (See Appendix II)<sup>9</sup>.

POPs are highly toxic, organic compounds since they are chemicals that can bioaccumulate and thus increase in concentration in the food web. The degradation products may also be stored in the fatty tissues, for example is DDT slowly transformed to DDE, which is more persistent than DDT, and also to DDD, where all of these compounds are stored in the fatty tissue. The endproduct, DDA, is however easily excreted through the urine. In different studies one has detected POPs and there different metabolites in various parts of the body, as in breast milk, adipose tissue, blood serum, plasma and so on. These might cause negative health effects including suppressed immune systems, damage to the respiratory system, reproductive problems, sex-linked disorders, shortened lactation periods for nursing mothers. and even death (abortion of foetus). Some studies show a connection between breast cancer and high levels of DDE in the breast tissue and blood serum<sup>10</sup>. There might also be some neurological effects, such as short-term memory loss and behavioural problems. Some of the POPs are also classified as carcinogens or possible carcinogens. Since POPs are semi-volatile they can subdue long distance transportation and you can find them all over the world (Allsopp et al. 2002).

Pentachloroanisole, PCA<sup>11</sup> is a metabolite of the pesticide pentachlorophenol (PCP) or from HCB. PCP is also a metabolite of HCB in living organisms. However, PCP cannot be metabolized to PCA in the body, this process is microbial. PCP was mainly used in wood preservation and to stop fungi to spread during the production of paper pulp. It was also applied as an insecticide and it has been reported to be used in bacterial soaps, and laundry products<sup>12</sup>. It has also been used on textiles to impregnate those (Palm et al. 2002). The commercial use as a pesticide has been limited and it has mainly been used in the industry production. PCP was forbidden 1978 in Sweden but it is still used in other parts of the world (Bernes, 1998).

<sup>6</sup> University of Nebraska http://ianrpubs.unl.edu/Pesticides/ec2505.htm#roe

<sup>8</sup> Homepage of the Stockholm convention http://www.pops.int/

http://www.inchem.org/documents/pims/chemical/pim405.htm#PartTitle:4.%20%20USES

<sup>&</sup>lt;sup>5</sup> www.wikipedia.org

<sup>&</sup>lt;sup>7</sup> Peru has signed this convention

<sup>&</sup>lt;sup>9</sup> PAN U.K News http://www.pan-uk.org/pestnews/Pn30/pn30p11b.htm

<sup>&</sup>lt;sup>10</sup> PAN U.K News http://www.pan-uk.org/pestnews/pn22/pn22p3.htm

<sup>&</sup>lt;sup>11</sup> Log K<sub>ow</sub> 5,45 (Palm et Al. 2002), LD<sub>50</sub> 50 mg/kg in rats (Ware, 1994)

<sup>&</sup>lt;sup>12</sup>IPCS INCHEM home

#### 2.4 International classification

Pesticides are classified according to the WHO recommendation in different groups:

**Table 2.** "The WHO recommended classification of pesticide by hazard (WHO, 2005).

Class	LD <sub>50</sub> for the rat (mg/kg body weight)						
		(	Oral	Der	rmal		
		Solids <sup>a</sup>	Liquids <sup>a</sup>	Solids <sup>a</sup>	Liquids <sup>a</sup>		
Ia	Extremely hazardous	5 or less	20 or less	10 or less	40 or less		
Ιb	Highly hazardous	5 - 50	20 - 200	10-100	40 – 400		
П	Moderately hazardous	50 - 500	200 - 2000	100-1000	400 - 4000		
III	Slightly hazardous <sup>b</sup>	Over 500	Over 2000	Over 1000	Over 4000 <sup>b</sup>		

The WHO hazard classification is used in Peru. There have, however, been recent changes in the labelling system. Previously category Ia was labelled with a red band colour and Ib with yellow. During the time of the study both of the systems were in use. All packaging have got standardized etiquettes that have to be approved by the National Agricultural Ministry, SENASA, before allowing it out to the market. They all contain the information that is required according to FAO and WHO. Illegally imported pesticides may not follow the same legislation concerning labelling and one could see other labels on the pesticides that had been confiscated by SENASA (San Martin) (Andrés Sixto, SENASA)<sup>13</sup>.

Table 3. Pesticide product label information to indicate the WHO hazard classification (FAO 1999)

WHO hazard class	Information to	Hazard statement	Band colour
	appear on label		
Ia	Extremely hazardous	Very toxic	Red
Ib	Highly hazardous	Toxic	Red
II	Moderately	Harmful	Yellow
	hazardous		
III	Slightly hazardous	Caution	Blue
Products unlikely to			Green
present a hazard in			
normal use			

## 2.5 Background and characteristics of Peru and study site

#### 2.5.1 Peru

Peru is a republic with an area of 1 280 000 km<sup>2</sup> (about three times larger than Sweden) and a population of about 28 million were 8.9 million live in Lima 15. It is a country with large social and economical differences where more then half of the population lives in poverty and about one fourth in extreme poverty (less then 2 USD/day p.p. resp. less then 1 USD/day p.p. according to the UN)<sup>16</sup>. The child mortality is the highest in Latin America and diseases like malaria, diarrhoea, and tuberculosis are common and malnutrition is another problem<sup>17</sup>. There

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<sup>&</sup>lt;sup>13</sup> During a study visit to the room where the confiscated bottles where stored we had the opportunity to see it ourselves

<sup>&</sup>lt;sup>14</sup> 2005, figures taken from INEI, <u>www.inei.gob.pe</u>
<sup>15</sup> www.wikipedia.org

<sup>&</sup>lt;sup>16</sup> Homepage of SIDA, Basfakta om Peru http://www.sida.se/sida/jsp/sida.jsp?d=498&a=1529

<sup>&</sup>lt;sup>17</sup> 2003/2004, figures taken from INEI, www.inei.gob.pe

has however, been a dramatic change of the expected lifetime, from 50 years in 1950-55 to about 73 years today. Despite of this, Peru is a "young" country where about one third of the population are 0-14.

Peru has a history of armed conflicts and corruption and it suffered from a severe economic crisis during the 1980ths. In the last years the country has been experiencing an economic upswing and during 2004 the growth of was GNP was 4.5 %. The export has increased, but so far it has had little effect on the high unemployment rate in the country <sup>18</sup>.

About one third of the labour force are farmers and 88 000 of them cultivate rice<sup>19</sup>. The area under rice cultivation fluctuates between 2000 km<sup>2</sup> to 2800 km<sup>2</sup>, and rice has become the most important source for calories and proteins in Peru. The consumption of rice is about 49 kg/capita/yr (Alva Azula et al. 2000). Over the last 30 years the production has increased and it is now higher than the demand. This has lead to a reduction of the price for rice (Palacios Agurto, 2002)

In 1999 there was accident where 24 schoolchildren were killed and 18 intoxicated by eating breakfast contaminated with pesticides in Tauccamarca, Peru. A villager had mistaken plastic bags filled with the insecticide methyl parathion for a milk substitute. In 2003 a farmer lost his whole family since they had eaten a soup contaminated with Parathion. These events led to much protest, which pressured the state to take harder actions against pesticides (RAAA, 2002).

#### 2.5.2 Study site



**Fig.3.** Map showing the localization of San Martin and Tarapoto in Peru<sup>20</sup>.

<sup>&</sup>lt;sup>18</sup> Homepage of SIDA, Basfakta om Peru http://www.sida.se/sida/jsp/sida.jsp?d=498&a=1529

<sup>&</sup>lt;sup>19</sup> 1998, www.riceweb.org

<sup>&</sup>lt;sup>20</sup> From vahoo geocities http://www.geocities.com/tarapoto peru/

San Martin is a department in the northeast part of Peru with an area of 51.253 km<sup>2</sup> and a population of 788 195<sup>21</sup>. The capital of the department is Moyobamba but the largest town is Tarapoto, which is situated in the province of San Martin (one out of the 10 in the department). The population of this province is 167 000 and about 119 000 of them live in Tarapoto. The town is situated 356 m above sea level and surrounded by rainforest, and has a hot dry tropical climate. Tarapoto is divided into different districts and during the study most of the time was spent in Morales and Cacatachi, which are situated 3 and 12 km, respectively. north of the city centre and where most of the rice fields are. The river Cumbaza runs through the town of Tarapoto and flows into the Rio Mayo, which in its turn discharges into the Rio Huallaga that eventually reaches the Amazon River.

The history of the region is quite turbulent and it used to be the main area for the cultivation of coca and thus held different drug cartels. The Maoist terrorist groups of Sendero Luminoso (The Shining Path) and MRTA (Movimiento Revolucionario Tupac Amaro) were strong in the region, Sendero in the mountains and MRTA in the jungle, before Fujimori came to power. He put an end to most of the activity with his harsh methods against the guerrilla. There is still a small active group in the department and an area is classified as "Zona Emergenciá" (Emergency zone) at this very moment (April 2006).

In the department of San Martin there are about 64 000 farmers and the region is an important agricultural area. The main crops to be cultivated are cotton, cacao, plantains, maize coffee, coca, and tobacco, but the principal crops are "Oil palm tree" (palma aceitera in spanish) (91% of Peru's production), rice and cassava. The rice production is the second most important in the country and about 7 500 of the farmers cultivate rice. In Tarapoto, 52% of the population are farmers and the rice farmers control about 80% of the economy (Sixto. SENASA). The cultivation of rice started in the beginning of the 1980s after the irrigation canal was built and the pesticides were introduced at the same time as the monoculture systems. The use of pesticides is not only limited to the rice cultivation; they are used in most of the commercial cultivations. A lot of different types and brands of are used and according to SENASA around 1000 toxic agrochemicals have been identified in the area (Leiva, SENASA).

The most common way to cultivate rice in the region is, as mentioned before, by irrigation and 93% of the rice production in Peru derives from this production system The "Selva Alta Irrigada" (irrigated highland jungle), which is the region where Tarapoto is situated, contributes with 28.4% of this figure. The average rice production is 6.0 TM/ha in San Martin (1994-1998) but there has been reported about yields of up to 9 TM/ha (Alva Azula et al. 2000). The total area of the rice cultivation in the department of San Martin is 430 km<sup>2</sup> and in the province of San Martin 38 km<sup>2</sup> (IAAP 2003/2004). The production in the department was 250 000 TM a year (1999) (Alva Azula et al. 2000).

<sup>&</sup>lt;sup>21</sup> 2005, Figures collected personally from Agronomist Christian Barta García from INEI, not yet published

#### 2.6 Pesticides in general in tropical countries

Inappropriate use and poor environmental management of toxic chemicals including pesticides causes about 355 000 cases of unintentional poisoning globally, were two thirds of them, are in developing countries<sup>22</sup>.

The agricultural production has been greatly expanded in the developing tropical countries during the past decade. Over 60% of the economically active populations in the tropical countries depend on agriculture (Wessling, 1997). Together with the expansion of agriculture, the use of pesticides has also increased drastically. It is reported that pests destroy up to one-third of the world's food crops during growth, harvesting and storage and the losses are even higher in developing countries (Carazo, 2002). Agriculture is, together with the mining and construction, one of the three most hazardous industries (ILO, 2004).

Latin America is becoming one of the fastest growing regions for agrochemical sales and many of them are known or suspected to have negative health effects (Allsopp et al. 2000). The explanations for the growth are market liberalization, growing international trade alliances, relative political stability and the increase of investments of cultivation systems based on an intense use of agrochemicals. A big problem is, that a pesticide may be sold under many different names and it is difficult to understand if it is the same chemical or not. Chemicals may also be mixed, and customs regulations are easily evaded, as it is difficult to identify the pesticides (Hagerfors, 1983).

## 2.7 Agricultural practises and pesticides at the study site

## 2.7.1 Agricultural practices, working conditions and pesticide use

In the district of Morales and Cacatachi no traces of the POPs banned in the Stockholm convention were found, nor did we observe any use of them. A study made by agronomist Bartra García for CIP, concerning the use of POPs in Tarapoto were he interviewed several stake holders who work with pesticides and have a good knowledge of what is being used, also showed that there are no POPs being used in the region, except for some use of Mirex. This pesticide is used to kill ants and it is not applied in the rice production. The pesticides used in the rice cultivation in the region are all listed in **Appendix III**. Several of the pesticides used are classified as Ia, Ib or II according to WHO. These are strongly recommended by the FAO to be used as scarcely as possible.

The farmers in the region cultivate irrigated rice, which means that during most of the vegetation period the soil is covered with water. This type of cultivation is possible, due to the canal system. The water is taken from the river C and then made, to a so-called "canal madre" ("mother" or central canal) and there after divided into many small canals that are used in the area of Morales and Cacatachi. The water is led onto one field and is then slowly distributed by gravity. The most common sorts of rice are called Capriona, Selva baja, and Selva alta. To commence the cultivation the rice seeds are first put in a small part of the field. They are then flooded by water to germinate and the plot is called an almácigo. During this time the farmers often apply herbicides to the plot. After about 30-35 days the rice plants are transplanted by hand to the rest of the field. This agriculture practice is used by over 98% of the farmers (Palacios Agurto 2002). The harvest is also mostly performed by "handlabour" since it gives a

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<sup>&</sup>lt;sup>22</sup> WHO, The health and environment linkages Initiative (HELI) http://www.who.int/heli/risks/toxics/chemicals/en/index.html

better quality of the yield and it is performed when the rice is 135-140 days. After the first harvest the farmer often let the plants stay on the field and grow to give a second harvest, before they prepare the soil for a new almasiego. This type of cultivation is called cabrilla. Most farmers have two campañas a year, which means that they harvest two times a year, but when they use cabrilla they harvest three times. During the whole time, most of the farmers use a variety of pesticides to keep the rice pest free, and fertilizers to make the soil more fruitful. In the region where the study was performed, it is possible to cultivate all year long due to the tropical climate and so the farmers do. They do not coordinate their cultivation and they all plant and harvest at different times. This is an optimal situation for the pests because they can move from one field to another since the fields in the region are all close to each other and connected by the irrigation and drainage systems. The use of cabrilla is also making it easier for the pests to stay in the region since they can spread from the old to the new plant. Rice is also very sensitive to pests. All this plays an important role for the extensive use of pesticides. The farmers mostly have small plots to support them and their whole family and they want to obtain a high yield. The labour is almost exclusive performed by men and machines are very seldom used. When used, it is mostly to work on the soil after harvest, in preparation for a new crop.

An inventory of the pesticides used in the rice cultivation that based on interviews and findings on the fields (See **Appendix III**), showed that the rice farmers in total used 61 pesticides with 31 active ingredients (14 Fungicides 10 a.i., 28 insecticides 13 a.i. and 19 herbicides 8 a.i.). The inventory holds 14 pesticides (6 are amongst the most commonly used pesticides) containing 6 active ingredients that are classified as Ia or Ib. These figures are not exact, but they are reasonable approximations.

The pesticides are mostly bought in small shops that have employees that to some extent are agronomist, but mostly this is not the case. The vendors often go out to the fields to see what kind of pests that trouble the farmers. They work under substantial pressure since they are held responsible for the yield and the competition is high since there is a whole street devoted to this commerce and the stores all sell the same product. The vendors also earn a certain percentage as commission and they tend to recommend the more toxic and expensive pesticides.



**Picture 1.** Street in Tarapoto with small Agrostores that sell pesticides but also veterinary supplies. Source: Agneta Andersson

When the farmer has bought the pesticides they are stored in different places, but mostly in his home (See **Picture 2**). To apply the pesticides a backpack sprayer is used. There are two different types of sprayers: "Motor fumigadora", which is a backpack sprayer with a motor. It makes a terrible high-pitched noise, but is not as exhausting for the farmers to use, as the second type, "Bomba mochila" (mochila means backpack in Spanish) with a hand pump. When the farmers spray with the "Motor fumigadora" they also inhale the fumes from the petrol. While preparing the pesticides for use, the farmer usually use a big bucket at the field were he/she makes a blend out of the highly concentrated pesticides. They normally mix the insecticides and fungicides and use water from the canals as a solvent (See **Picture 3**). The herbicides are applied separately. We heard about cases where the backpack sprayer broke or, were it has been leaking and thus spilling out pesticides on the back of the person spraying.

It is also worth pointing out that the farmers usually hire people to help them to spray, or to do it for them. These people are called peon (farmhand). That means that there normally are at least two people, spraying pesticides on the field at the same time. While we were watching such an event, we could clearly see that this lead to that they accidentally sprayed each other, since they walked through or sprayed in the same direction where the other person was working (See **Picture 4** and **5**).

The farmers spray every campaña but with different frequencies. Some spray only twice whereas others spray about 8 times. Normally they only spray during noon when the sun is less strong and the temperature a bit cooler.

We did not observe any women spraying, but the ones that work on the field are often involved in the spraying process by mixing the pesticide blend or helping to fill up the backpack sprayers on the field. Often the whole family is involved in the cultivation. The children help their parents on the field and many of them spray. It is then often the task of the farmer's wife to wash the clothes that the farmer wore during spraying.

One of the farmers we watched while spraying took a breakfast break during the spraying. He and his son, who worked with him, did not wash before they ate the meal and thus ate the food with pesticide contaminated hands. This is not a very common practice, of what we understood of the other interviews, but our suggestion is that it does happen quiet frequently. A schedule for spraying pesticides during a campaña for one farmer is found in **Appendix IV** 

After the spraying is finished, most farmers wash the backpack sprayers and often themselves, in the canal. The bottles are mostly left on the field (See **Picture 9**). Some of the farmers burn them as fuel wood and some burry them in the ground (See **Picture 6**). They also use them to put on sticks on the fields to mark out their borders.

The poorest farmers are the ones that live out on the field. The other farmers mainly live in Morales city and drive or walk out to their chakra everyday.



**Picture 2**. Pesticide containers and blue bomba mochila, backpack sprayers, stored in the living room of a farmer. Source: The Author



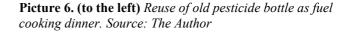
**Picture 3.** A farmer mixing different pesticides in a big bucket. The gloves she is wearing were a present from us and she normally mixes without them. The young boy on the picture works as a peon and his task this day was to run out to the fields and fill up the Motor fumigadoras when they run out of the blend. Source: The Author



**Picture 4. (to the left).** Filling up the Motor fumigadora, notice the drop of pesticides running out on the boys feet. Source: The Author



**Picture 5.** Young men applying pesticides without any protective equipment and walking through the area that has been fumigated Source: Agneta Andersson







06.16 11:32

**Picture 7.** Old pesticide bottle on the dining table. Source: The Author

**Picture 8.** Young boy playing while his father is working on the fields. Far in the background you can see two men spraying. Source: The Author



**Picture 9.** Some of the most commonly used pesticides. This is one of the many mixes that were used on the field where the bottles also are left on the field. Source: The Author

## 2.7.2 Farmers exposition of pesticides

To summarize: The farmers and peons are exposed to pesticides in the following three possible ways<sup>23</sup>:

#### • Dermaly

While mixing the concentrated pesticide with water, the farmers may get stains on their hands and arms or other parts of their body. This step potentially, presents the greater hazard because the pesticide is present at its highest concentration in this form. While the farmers or peons fill the backpack sprayer, they may also spill the mixture on parts of their body. During the spraying some of the mixture might leak out on their back if the backpack sprayer is damaged. If two persons or more are spraying, they may accidentally spray on each other. When they wash the backpack sprayers and the clothes they have been wearing during spraying they will also come in contact with the pesticide residues. Taking care of the discarded and used bottles may also present a dermal exposition.

## • Through inhalation

When the farmers mix the spraying liquid they will inhale the chemicals that evaporate. When they spray they sometimes walk back through the area they just have sprayed or they inhale the fumes from the other person spraying (in case they are two or more). Depending on where the pesticide bottles are stored they might expose the farmer and his family to the fumes in their homes.

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<sup>&</sup>lt;sup>23</sup> This is assuming they do not wear any protective equipment.

#### Ingestion

The water in the canal is not usually used for drinking water, but water from some of the small streams that are running through the area sometimes is. A guess is that in extreme cases, if there is no other liquid available, the farmers drink out of the canal. Families that live on their charkas do however sometimes take their drinking water from the canal. There are also some wells, but since the groundwater level is quiet high there might be some leaking of pesticides in to this water as well. In the end all the water from the agricultural area is led back to the River of Cumbaza from which everyone takes their drinking water. Persistent pesticides that can accumulate in the food web are also a threat to the farmers. Many of the farmers breed hens that live in the same environment as them and hence also become exposed to pesticides and might bioaccumulate them. Many farmers also cultivate fruits around the rice fields. They often rinse them in the canal before they eat them and thus may contaminate them with the pesticides residues from the canal. Again, this is especially critical to the farmers that live out on the fields and do not have easy access to tap water (Deep interview, Questionnaires and observations).

## 3. Material and Methods

## 3.1. Blood samples

## 3.1.1 Sampling.

We chose to sample blood since many hydrophobic pollutants, such as DDT and PCBs, are highly hydrophobic and are distributed to the lipids in the body i.e. adipose tissue, subcutaneous fat and blood fats (Hovander et al 2000.). Blood is advantageous since it is easier to sample than other tissue and the ethical questions are also easier to handle since it is often relatively easy to obtain volunteers and their approval for sampling. (Thomas et al. 2005).

The blood sampling of this study took place the 9<sup>th</sup> and 10<sup>th</sup> of July 2005, at the meeting place of the irrigation committee of Rosanayco. In total, blood samples were taken from 30 persons. Out of this group, 24 were rice farmers and 5 reference persons. After a Rosanyaco meeting the 12<sup>th</sup> of June, 19 of the farmers were selected randomly from a list of voluntaries. The other 5 farmers were the ones with whom we made the deep interviews, and from whose farms we also did sample water. Hence, the sampling was voluntary and random.

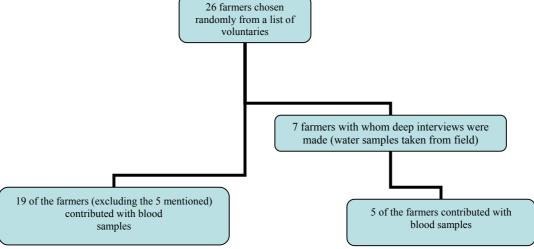


Fig.4. Sampling methods of blood samples and deep interviews

The reference persons were chosen to try to represent the same age- and sex groups as the farmers. The study was not really successful in this matter since it was harder to find volunteers amongst the city population<sup>24</sup>.

**Table 5.** Distribution of sex and age amongst sample pool and reference

	Male	Female	Age Median (Range)
Samples	21	3	46 (18-74)
Reference	2	3	26 (25-70)

## 3.1.2 Short interview/questionnaire

Before the actual blood sampling took place, a short interview was performed (See **Appendix V**) to obtain some knowledge about the test persons' pesticide use and handling. The purpose was to use this data and see if there would be some connection to the concentration of pesticides the farmers would have in their blood

#### 3.2.2 Blood analysis

The nurse student took 3 test tubes (4 ml each) blood from each person. The test tubes were made out of glass, with a content of heparin that prevents the clotting of blood and the formation of fibrin clots<sup>25</sup>. The test tubes were kept cold in a cool box after they were used. In the evening the 9<sup>th</sup> and the 10<sup>th</sup> the blood samples were taken to the university of San Martin in Tarapoto, UNSM and centrifuged for 10 min. This was done to separate the blood plasma from the red corpuscles. The plasma from each person was transferred to, two new test tubes. In one test-tube the plasma content of two test tubes with 4 ml each were transferred and in the other one, the plasma content of one 4 ml test-tube. This was made to receive plasma, to analyze and to have plasma as a control. The plasma was kept in the freezer until the workup was continued on the 11<sup>th</sup> of July 2005. This was made at the laboratory of phytophatology at UNSM. The laboratory work performed there, contained different cleaning steps to obtain the fat in the plasma. The last step of the analysis that was performed in Peru, took place in a fume cup in the microbiology laboratory of "Laboratory Referencial". This involved the evaporation of the solvents in the test tubes<sup>26</sup>. The 29 test tubes containing the evaporated fat, the 31 control tubes containing plasma and 58 test tubes containing fat dissolved in cyclohexane were transported to Sweden. They were kept as cool as possible during the whole trip and immediately put in the freezer of the institution of IMA at SLU at the arrival in Sweden. On the 3<sup>rd</sup> of October the test tubes were brought to the IMK at Stockholm University for further analysis. The 29 samples containing evaporated fat were first analyzed. They were put through different cleaning step to obtain clean solutions to analyze on the GC-ECD.

The samples were analyzed in two different fractions; neutral and H<sub>2</sub>SO<sub>4</sub> dissolved fraction. This was made to see if one could detect different pesticides in them. Samples of the neutral

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<sup>&</sup>lt;sup>24</sup> Two of the blood samples from reference persons were taken on the 4<sup>th</sup> of August and only centrifuged to separate the plasma. These samples were not analyzed yet. With these samples there would be a better representation of reference persons.

<sup>&</sup>lt;sup>25</sup>RX list-the internet Drug index <a href="http://www.rxlist.com/cgi/generic/heparin">http://www.rxlist.com/cgi/generic/heparin</a> cp.htm

<sup>&</sup>lt;sup>26</sup> After a week of evapotranspiration solid fat had formed in the bottom of the test tubes. The fat was suspected contaminated, since there were some green and black points in them. Tests were taken from some of tubes and they were cultivated to see if it possibly could be fungi contamination. They all came out negative. Maria Athanasiadou concluded that our suspected contamination was plant pigment and that it would not have any effect on the further analysis

fraction was pooled together and analyzed by mass spectrometry to determine what substances was behind the unknown peaks in the GC-ECD. The same method was performed with samples from the  $H_2SO_4$  dissolved fraction. Although the concentrations were so high within these samples that the analysis only was made with one sample at the time. Finally the further analysis only was made with the samples from the neutral fraction on the GC-ECD. (Method based on Hovander et al 2000.) For a detailed description of the Method see **Appendix VI.** 

#### 3.2 Farmers

## 3.2.1 Sampling

In depth interviews were performed with 7 farmers in the area. The farmers were picked randomly and voluntary. Through participation in meetings of the farmers the contact was established. The first meeting attended was at a barricades were the rice farmers had gathered to protest against the low prices for rice due to the import of rice from other countries. During this meeting the first contact took place and invitations to different irrigation committees were made. Some farmers also signed up immediately on the voluntary list for our interviews and blood sampling that was brought to the meeting. Furthermore, the meetings of two irrigation committees were attended were our study was presented and where even more farmers decided to participate. The information gathered was then examined and farmers thought interesting and representative was chosen (See figure 4) although the goal was also to receive at least one interview with a woman even though it would not be representative, since the ratio of female farmers and a male farmer clearly is less than 1:6. It was clear from the beginning that we would not get statistically reliable results. The wish was more, to get a greater picture of the situation and to try to understand how the farmers that we did interview cultivated their land and all the factors influencing the use of pesticides.

#### 3.2.2 Deep-interviews

The technique used during the interviews, were semi-structured and called PRA/RRA (Participatory Rural Appraisal/Rapid Rural Appraisal) methods.

**Table 6**. Purpose and description of different PRA/RRA-tools used (Fagerström et. al China (1997-2000) and Vietnam (2001-2004))

		Purpose of tool/Information
Tool	Level	_
Day clock	Farm	Labour division for men and women
Time line	Village (farm)	Infrastructure, economical reforms, land division
Sketch map	Farm and village	Relative location of the village, with roads, paths, the distribution of houses, cropland and other land and water resources and ownership
Seasonal calendar	Farm and area	The annual climate and other parameters connected to the cultivation as they are understood of the farmer and the annual cultivation practices of the farmer

The interview questions and samples of the other PRA/RRA tools used are found in **Appendix VII and VIII**.

## 3.3 Health care personnel

#### 3.3.1 Sampling

The questionnaire was filled in by a total number of 15 persons; 10 doctors and 5 nurses, at three different medical centres: the major hospital in the region, a smaller hospital in the city and also in a health care centre in the region where most of the farmers live. They were all chosen randomly<sup>27</sup>.

#### 3.3.2 Questionnaires

During a study made by RAAA in 2003 a questionnaire was made that was directed to the health care personnel concerning the knowledge about and care provided to persons poisoned by pesticides. We used the same questionnaire as RAAA, but added two questions regarding recommendations that the personnel might give the farmers to prevent intoxications or to stop them. We did this since many of the farmers drank different mixtures before and after applying the pesticides and we were interested to find out where they had heard about this kind of treatment and if this actually was recommended to them at the hospitals. For a detailed version of the Questionnaire see **Appendix IX**.

## 4. Results and discussion

#### 4.1 Pesticide content

The method being used (Hovander et al 2000.) provided two different fractions; a neutral fraction and a fraction partitioned into concentrated sulphuric acid, henceforth called the  $H_2SO_4$ -fraction. The neutral fractions from all of the samples were analysed, while the  $H_2SO_4$ -fractions from only two samples were analysed using GC-ECD, to investigate if any halogenated compounds were present. The neutral fraction of the all the samples were screened for 21 pesticides or metabolites and analysed on the GC-ECD. Some samples were also analysed on the MS for verification and identification.

## 4.1.1 $H_2SO_4$ -fractions

The GC-ECD analysis of the H<sub>2</sub>SO<sub>4</sub>-fractions from samples 1 and 2 showed at first several peaks. To further check if these peaks were from halogenated compounds the samples were also analysed using GC-MS. None of the compounds in this fraction was halogenated. Instead it detected compounds containing sulphur. It seemed to be one group containing three sulphur atoms and another containing two sulphur atoms. They are probably from the same family, but with several isomers. There are some pesticides in our inventory that could be candidates for the unknown peaks, but with the lack of information about "new" pesticides there would be nothing else but speculations.

## 4.1.2 Neutral fraction

The neutral fractions from all samples<sup>28</sup> were screened for 21 substances (See **Appendix VI**). Two substances that were found in all samples were the following: p.p-DDE and

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<sup>&</sup>lt;sup>27</sup> The questionnaires were given to a secretary or student who then distributed them to the employees
<sup>28</sup> Sample 8 and 9 were put together during the extraction and the result was multiplied with the percent their bloodplasma concluded

Pentachloroanisole<sup>29</sup>. The most abundant component was p.p-DDE. One could also detect lower concentrations of  $\alpha$ -HCH,  $\gamma$ -HCH, HCB and traces of halogenated biphenyler (PCB), and DDT in many of the samples (Chemical structures **Appendix X**). An unknown peak detected in many of the samples was not yet identified. For an example of a chromatogram **Se Appendix XI** 

**Table 7.** Limit of quantification (LOQ), plasma concentration ng/g fresh weight (f.w) and ng/g lipid (l.w) of p,p'-DDE, HCB, a-HCH, g-HCH and Pentachloroanisole are given .

Substances	LOQ	Fresh weight (f.w.), n=24 <sup>30</sup> Lipid weight			id weight (	l.w.) <sup>31</sup>	
	ng/ml <sup>32</sup>	Mean	Median	Range	Mean	Median	Range
p,p'-DDE	0.5	20	13	1,1-140	3970	2580	220-28200
HCB <sup>33</sup>	0.1	0.29	0.17	<0.1-0.84	54	29	<13-168
<b>γ-HCH<sup>34</sup></b>	0.2	0.27	0.24	< 0.2 0.54	53	50	<12 -145
PCA	0.2	7.4	5.9	0.59-27	1470	1180	42-5470

The samples were all treated as one group since there were too few samples from females to separate them into two groups based on sex. The total number of samples is quite small and therefore, the results can only be taken as an indication of the concentrations farmers in the region have in their blood. An evaluation was made to see if age, spraying intensity and different spraying practises, such as use of protective clothing, had any effect on the concentrations. This was made by grouping the samples according to the individual parameters and then comparing the median, mean and range of the groups (See **Appendix XII**). Based on the interviews made before sampling some plausible explanations for the highest concentrations among individual samples were suggested.

#### 4.1.2.1 p,p-DDE

This substance was detected in all samples including the references samples. One could clearly see a great difference between median of the reference group and the farmers<sup>35</sup>. There seems to be a correlation between the concentration of p,p–DDE and age in the samples. When excluding the extreme value from the diagram were age was plotted against concentration this became even more evident since the R<sup>2</sup> increased substantially (from 0.1256 to 0.527) (See **Appendix XIII**). One could also see this comparing the median and mean of the two age groups (See **Appendix XII**). The highest concentrations were found in the samples from the older farmers. The extreme value can to some extent be explained by a different type of exposure history. If the extreme p,p-DDE value is excluded there also seems to be a correlation between the number of years the farmer has been engaged in spraying and the concentration of p,p-DDE. The group that do not wear any protective equipment at all seem to have a higher concentration in comparison with the group that spray wearing some kind of protective clothing.

<sup>&</sup>lt;sup>29</sup> Since there was no standard for Pentachloroanisole the concentration was calculated by using a response factor from HCB from old standard curves. Pentachloroanisole was identified on the MS.

<sup>&</sup>lt;sup>30</sup> 21 men and 3 women

<sup>&</sup>lt;sup>31</sup> calculated assuming 0.5% lipid in blood

<sup>&</sup>lt;sup>32</sup> Here calculated as 1ml of plasma is equivalent to 1g plasma

<sup>&</sup>lt;sup>33</sup> n=29 (20 m and 3f)

 $<sup>^{34}</sup>$  n=29 (20 m and 3f)

<sup>&</sup>lt;sup>35</sup> Although one has to note that the second highest concentration of p,p-DDE was detected in the sample of a reference person, hence a person that never have fumigated. The explanation might be age, different food consumptions and environmental exposition

#### 4.1.2.2 Pentachloroanisole

This substance was detected in all samples including the reference. It was detected as an unknown peak in the chromatogram and identified on the GC-MS (See **Appendix XIV**) An interesting fact is that the concentrations are evenly spread throughout the samples and references and that the samples from me and my student companion do not deviate from the rest. This might be a sign of constant exposure in the surrounding (when the samples were taken, we had been three months in Peru and two months in the region). The concentration of Pentachloroanisole does not seem to correlate with age when the concentrations were plotted against it. While looking at the groupings there seems to be slightly higher median concentrations in the group that spray without any protection, have been spraying a long time and are elderly. One has to note though, that the sample with the highest concentration found is not a part of any of these three groups.

#### 4.1.2.3 HCB

Traces of this substance were detected in 96% of the samples including in all the references. There does not seem to be a direct correlation with age even though the MED+ group has a slightly higher median and mean. The same can be said of the group of farmers that have been spraying for a long time and the ones that do not use any protective equipment. As with Pentachloroanisole, the highest concentration detected is not a part of any of these groups.

## 4.1.2.4 α- and γ-HCH (Lindane)

Traces of  $\gamma$ -HCH were found in 96% samples and in all the reference samples.  $\gamma$ -HCH is not significantly higher in the sample group compared to the reference either and the concentrations seem to be evenly distributed through the sample and reference groups and amongst all the other groups. Traces of  $\alpha$ - HCH was found in 17% of the samples but they were all lower then the Limit of quantification and therefore no concentrations are found in Table

#### 4.1.3 Comments

None of the women sampled was actually spraying, but two of them helped during the mixing of pesticides and hence expose themselves to high concentrations of the substances. This might be an explanation of why the highest concentrations of HCB,  $\gamma$ -HCH and Pentachloroanisole and one of the highest  $\alpha$ -HCH concentrations was detected in one of the female samples.

One of the youngest person sampled showed quiet high concentrations, values over the median and amongst the highest, of all the substances except for  $\alpha$ -HCH. This person works as a peon and sprays about 2-3 times a week and it seems to have an effect on the concentrations in the blood. One of the other persons working as peon also has a high concentration of p,p-DDE despite his low age.

The group of farmers that do not to wear any protective clothing seem to have higher concentrations of all substances except for  $\gamma$ -HCH, in comparison to the group that claim to wear protective equipment. The protective clothing is hardly anything one would really call so in western terms and the reason for the slightly less concentrations is probably just the fact that these farmers are more aware of an actual danger and are probably more careful in their handling of pesticides than the ones that work without any protection at all.

It is a happy notion to conclude that the farmers in Tarapoto have stopped using the banned POPs in their rice cultivation. That no high concentrations were detected in the blood samples

is another proof of for this matter. The ratio DDT:DDE shows also that DDT is not used any more since mainly p,p-DDE was found in the blood. That  $\gamma$ -HCH is detected in many more blood samples than  $\alpha$ - HCH indicates that Lindane has been used. The low concentrations also show that it has not been used for a longer period of time.

There seems to be a delay in the implementation of the laws since Mirex was the last pesticide to be prohibited of the POPs in Peru and this is the single POP that one still can buy in Tarapoto, according to the study of Agronomist Gartra Barcia. However no high levels of Mirex was detected in the blood samples.

## 4.1.4. Comparison to other studies

There are some difficulties in comparing the results to other studies since one has used different analytical methods, sample media, and different bases to calculate the concentrations of pesticides. In most reports the concentration is reported in ng/g lipid. In this study the concentration has been calculated to ng/g blood plasma. To be able to do some kind of comparison with other studies, a recalculation of the blood concentrations into lipid weight (l.w.) ng/g lipid by assuming a lipid content of 0.5% in the blood was made. This only gives an indication of what the concentration in the lipid might be. The amount of lipids in the blood differs from person to person and to obtain an exact concentration it has to be calculated for each individual. This is to be considered during the following comparison.

Table. 8. Comparison in l.w, (ng/g lipid) to other studies

Comparison			concentration (ng Median (Range)	/g lipid)			
Year	Country	Sample type	p,p´-DDE	НСВ	γ-НСН	Pentachloroanisole	Reference
2005	Peru	Plasma	2578(221-28154)*	29(<13-268)*	50(<12 -145)	1185 (42-5473)*	This study (2006)
2002	Nicaragua	Serum	2800 (1240-10 600)		42(32-67)		Cuadra et al. (2003)
2002	Nicaragua	Serum	1450 (250-3420)		31(19-92)		Cuadra et al. (2003)
	Mexico	Serum	3627(242-20 856)	167(16-1160)	42		Wasliszewski et al. (2000)
2000	Ghana	Serum	380(120)^	30(10)			Ntow (2000)
	Latvia	Plasma	2200(850-4200) <sup>x</sup>	240(110-410) <sup>°</sup>			Sjödin et al. (2000)
	Sweden	Plasma	1100(330-3900) <sup>x</sup>	100(60-150) <sup>x</sup>			Sjödin et al. (2000)
	Sweden	Plasma	(110-880)	16-56			Norén et al. (1999)
	Sweden	Serum	88(10-1270)				Hagmar et al (2003)
	U.K	Serum	100(15-2600)	14 (5.4-72)	<1.7(<1.7-110)		Thomas et al. (2005)

<sup>\*</sup> calculated assuming 0.5% lipid in blood
^ Concentration are given as Mean with Standard deviation in brackets
Concentrations from men with high fish consumption, 10-90% percentile

No studies from Peru concerning the concentrations of HCB, γ-HCH and p,p-DDE and non at all concerning Pentachloroanisole, in blood of humans were found.

The data from Nicaragua are from two groups of young women that are high consumers of fish from Lake Managua. These women do not work in an agricultural area and the main source of pesticides is through food consumption mainly fish. One of the groups does have a higher median than the farmers in Tarapoto. The fish consumption is low in Tarapoto. The age of the sampled individuals in Peru is also higher than amongst the women from Nicaragua so it was to expect, due to bioaccumulation that the concentration would be higher then in that study. The median concentration of  $\gamma$ -HCH is slightly higher in Tarapoto and due to its mainly agricultural use, the exposition might have been greater there than in the area in Nicaragua.

In the study by Sjödin, Swedish and Latvian men were tested to see how the fish consumption effects the concentration of persistent pollutants in the blood. The median p,p-DDE concentrations in Latvian men with high fish consumption is almost the same as amongst the farmers in our study, while the median HCB concentration is ten times higher in the samples from the Latvian men. Today, the main source of HCB is combustion and incineration, and there may be more such industrial sources in the Baltic Region than in Tarapoto. Compared to the farmers in Tarapoto, the samples from the Swedish studies performed by Sjödin, Norén and Hagmar show lower concentrations of p,p-DDE. Hagmars study was performed on men signing in for the military and hence they were all 18 years old. A comparison to his study with the samples from the youngest individuals in Tarapoto, still shows a large difference, with the lowest concentration amongst the samples from Peru being about 4 times as high as the median of the samples from Hagmars study.

The concentration of HCB is higher amongst the Swedish men eating much fish in comparison to the farmers in Tarapoto. In Noréns study however, the Swedish samples contain lower concentrations than the one from Tarapoto.

The Mexican study was only performed on women from Veracruz, a large city at the Gulf of Mexico. These women have higher concentration of p,p-DDE and HCB than the farmers in this study. One reason for this might be that DDT and HCB was used for a longer period of time in the region than in Peru. The women live in a larger city (0.5 million) and they might also become exposed to more HCB than the people in Tarapoto due to more industries and combustion. There is no larger difference between the  $\gamma$ -HCH concentrations in the two studies; it is only slightly higher amongst the Peruvian samples.

The study in Ghana correspond the most to this study since it was performed in a farming community were half of the sample pool was female and the other half male. The concentration of p,p-DDE is about 10 times lower than in Peru. DDT is banned in Ghana and according to the results they have stopped using it in the study region (the ratio DDT:DDE indicates this). A reason for the lower concentrations might be that there were fewer quantities used in the region than in Peru. The median of HCB in this study seems to be almost the same as the mean of the study in Ghana. In the blood samples from Ghana no detectable residues from γ-HCH was found.

The goal of the study performed in the U.K was to screen the concentration of persistent pollutants in whole population and hence the sample pool was large and mixed. Compared to the general population in the U.K the concentrations of p,p-DDE amongst the farmers in

Tarapoto are much higher. This is not the case for HCB were the median is only twice as high in the samples from Tarapoto as in the samples from the U.K. The concentrations of  $\gamma$ -HCH are higher amongst the samples from Tarapoto, where one can assume that the exposition to Lindane has been more recent and intense.

## 4.2 Interviews, questionnaires and observations

## 4.2.1 Questionnaire blood sampling

During the weekend the blood samples were taken, Britta Palm filled out a questionnaire while interviewing the persons giving blood. This was done to exclude the possibility of the farmers not understanding the questions. The purpose of this interview was to obtain some knowledge about the test persons' pesticide use and handling. The goal was to use this data and see if there would be some connection to the concentration of pesticides the farmers would have in their blood (Questionnaire See **Appendix V**).

Table 9. Agricultural information and spraying habits

| 21 map and 3 women with a

General	21 men and 3 women with a median age of 46 (18-74) were
Information	interviewed. The educational levels for the questioned persons varied.
	Of the farmers, 11 had finished primaria (5a), 5 had not (2-3a), 4 had
	also completed secundaria and another 4 had even further education. It
	was, however, observed that some of the farmers clearly had
	difficulties in expressing themselves in writing and understanding
	written text.
Cultivation	The median area of the rice field that the farmer questioned owned, was
	3 ha (1.25- 8). All the farmers in the blood survey said, that they had
	two campañas and hence recieved at least two yields. They all
	cultivated irrigated rice and all of them used pesticides on their fields.
Storing of	Most of the farmers stored the pesticides in their home. Of these, 11
pesticides	had a separate room where they stored the pesticides. The living room
	was used by 3 and 1 stored the pesticides in the kitchen. Another
	common place used as storage was the garden patch or repository
	where 6 kept their pesticides. 2 of them used the pesticides at once after
	they bought them
Spraying habits	Out of the 24 persons questioned 16 sprayed pesticides. Another 4 had
	ceased to spray and 2 people had never sprayed. There were also 3
	individuals that helped to mix the pesticides but did not spray
	personally. The ones that spray or have been spraying had in average
	(median) been spraying for 11.5 (4-50) years. During each campaña
	pesticides were applied about 4-6 times. One of the farmers only had
	two application times during one campaña and a couple of others, 7-8
	times.
	13 of the farmers were also responsible for the dilution and mixing of
	the pesticides before they were used. This is quite a critical procedure
D	since the pesticides are very concentrated during this step.
Pesticides	The farmers used 6 to 18 different pesticides; the mean number was 12. According to the inventory in the region many more pesticides were
	actually used 12 of the farmer used to spray with parathion before it
	was forbidden.

Peon	There is quiet a common practice to hire a peon to help during spraying. 7 of the farmers did this and another 7 hired them to spray instead of them. Of the people questioned 3 worked as a peon. The peons tended to spray more often than the farmers owning their own land. One person working as a peon sprayed 2-3 times a week.
Protective Equipment	None of the farmers used the required protective equipment recommended on the labels, except for a facemask that 3 of the farmers claimed to use and 2 of the farmers said that they had been using <sup>36</sup> . Amongst the farmers spraying and that had been spraying, 9 used no protection. Some preventive measurements were taken by the farmers. 9 of them used a scarf to put in front of their face while they were spraying. Trousers with long legs were used by 10 of the farmers, and 6 used sweaters with longs sleeves. Gloves, footwear, and glasses were used by one farmer each. The most common combination of clothes was the use of a scarf and to wear trousers with long legs. This is to some extent what FAO recommend (FAO 1990).
Poisoning due to pesticides	18 farmers claimed to have felt poisoned by pesticides but only 4 of them had actually visited a health care centre because of the symptoms. The most common symptoms that 13 of the farmers had experienced were dizziness, itching and a burning sensation on the skin. 9 of the farmers said that they had felt nausea or headaches. In 6 of the cases the farmers had vomited and felt sick due to the pesticides. Diarrhoea had been experienced by 4 and rashes by 2 of the farmers. After spraying one farmer said that he felt numb in the body, one said that he always felt as if he was suffocating, one farmer always felt cold, two of the farmers felt very tired and another two said that they had pain in their bodies and felt weak. Two farmers stopped fumigating after they had been intoxicated. The pesticide causing the poisoning was in most cases Parathion (this pesticide is not used any more in the rice cultivation), Carbodan and Tammaron was also mentioned.
Cleaning of clothes and equipment	The clothes that are worn during spraying become contaminated by the pesticides and need to be washed or discarded after usage. One farmer burnt all his clothes after applying the pesticides. Mostly the clothes were washed in the home of the farmer by their wives. This was the case for 9 of the farmers. However, 6 farmers washed their clothes themselves in the irrigation canal and 9 farmers also washed their spraying equipment there. There were 2 farmers who washed the equipment in their homes. 3 farmers said that they avoided washing the spraying equipment in the canal since it seemed to contaminate the water. They only rinsed the equipment and poured the water onto the rice field or they did not wash the equipment at all.  The majority of the farmer showered and cleaned themselves after spraying and before they ate.

<sup>&</sup>lt;sup>36</sup> During our study in the region were we stayed 3 month we NEVER saw anyone use this and we observed several fumigation

Curing themselves	During our study we encountered many different methods and recipes
	for miracle drinks to cure oneself against poisoning by pesticides. It
	is very common to drink milk or lemonade or a combination of milk
	with dark lemonade after fumigation since the farmers believe that
	this cleans your blood from the pesticides. Salt and lemon are also a
	widely used combination

## 4.2.2 Interviews with farmers

The deep interviews were the basis for the earlier chapter *Agricultural practises and pesticides at the study site*. The main purpose and goal was to achieve a better and more complete understanding of how the farmers work and it was preferable having the deep interviews out on the field since it also gave the possibility to watch the farmer work. One could also see where and if they stored the pesticides at the field and what the farmers did with the used pesticides bottles. During two of the visits the farmers were observed while spraying. The observations made it easier to understand why some of the protective equipment was not used not even the rudimentary recommended by the FAO for the tropics (See **Appendix XV**). You simply cannot walk in the muddy fields with rubber boots without getting stuck. It is also very hot during the day and to wear thick clothes like the protective clothing would be unbearable for the people working on the field. Hence, one saw that some farmers wore a scarf around their face and they also wore trousers with long legs and sweater with longs arms but one also saw farmers spraying while wearing shorts and T-shirt and without even a rudimentary protection against inhalation of the pesticide spray.

During the deep interviews questions were also made to check the knowledge of farmers about the labels on the pesticides. All of the farmers claimed to read the labels. Hence some key questions were asked to check the knowledge about the different colour codes and in many cases the farmers clearly were not aware of what colour indicated which type of toxicity.

With help from the seasonal calendar it was also clear that the farmers had different perceptions about the seasons e.g. when it rained and when it did not, when the pests attacked the crops and when to spray. The reason might be that they all cultivate at different times, but the seasons should be the same for all the farmers.

All the farmers that were interviewed used pesticides and they were all aware of that pesticides are poisonous, to different extent. Interestingly, the farmers often referred to the pesticides as medicine.

The survey that was made with the farmers while taking the blood samples showed that although many of them felt intoxication symptoms, they normally did not go to the clinic with these problems. One of the most common symptoms were itching or burning sensation on the skin, this is typically causes by pyrethroids like cypermethrin. This is one of the most utilized pesticides in the region.

One farmer said that he had a daughter that was born with a very irritated skin in and around the mouth. He said that it looked like she had been burnt. His explanation was that this occurred because his wife always washed the clothes he had been wearing while he was applying pesticides. During this time he worked on a tobacco plantation spraying with large quantities of, e.g., Carbofuran and Mancozeb. Despite this, he did not stop to spray after his daughter was born and he and his wife tried to cure the daughter with water from cane sugar.

Most farmers said that they had gradually started to use more and more pesticides since the pests had become more resistant against the substances. Many of the farmers did not use any pesticides at all when they started to cultivate rice but today they all do. The farmers also spoke about the different animals like apes, foxes and snakes that used to live close to the field. They all said that these animals almost had disappeared today

## 4.2.3 Questionnaires health care personnel

Ten doctors and five nurses filled out the questionnaires. All the nurses and two of the doctors worked at the major hospital, five of the doctors at the smaller hospital and three of the doctors at the health care centre.

The most striking result was that only one of the nurses and three of the doctors had received any training in treating and diagnosing poisoning due to pesticides. However, nine of the doctors and two of the nurses claimed to have treated patients with poisoning caused by pesticides. The number of patients treated varied from one case to 20 annually.

Only one nurse, but six of the doctors claimed that their hospital had an established procedure to attend poisoning due to pesticides. It was, however, a bit odd, that doctors working at the same hospital not always agreed about having a procedure or not. One of the hospitals actually seemed to have a common and well know procedure since all doctors working there, mentioned a special protocol that has to be filled out, at least in case of poisoning with organophosphates. They also mentioned the same working procedures which include injections of atropine, washing of irritated skin, respiratory help if needed, help against dehydration and if the health condition is very bad, a gastric wash. One of the nurses did also mention this procedure. In the health centre, in the area where most of the farmers live, there did not seem to be a well-known standard procedure. Since it was possible to gather information about cases of poisoning due to pesticides from the ministry of health, San Martin, there seemed to be statistics and a standard protocol that had to be filled out about these cases at the different hospitals and health care centres. The problem seemed to be, that not all of the staff were aware of this matter and one can wonder if there might be cases that are not reported. Due to the lack of training and knowledge of pesticide intoxications amongst the nursing staff there might also be some cases of wrong diagnosis.

There also seemed to be some disagreement about the occurrence of pesticide poising cases that have been treated during the last 5 years. Even though they worked at the same hospital some of the health care personnel questioned claimed that there had not been any cases at all while others said there had been cases. The cases mentioned were mostly due to poisoning agents like the rodenticides Racumin and Campeon<sup>37</sup>, organophosphates, Machete (a.i Buthachlor) and Parathion.

One of the doctors claimed that the hospitals did inform farmers about first aid in case of poisoning due to pesticides, none of the other doctors or nurses agreed with him. None of the farmers mentioned anything about this matter either.

The health care personnel did in general recommend the farmers to wear protective clothes while spraying and to be careful with their equipment and to bathe themselves well after

<sup>&</sup>lt;sup>37</sup> We were told that Campeon sometimes was used to refere to people that have tried to commit suicide since this substance was the most common to use

spraying. Some of the nurses also recommend the farmers to drink milk after spraying or to drink water with large amounts of sugar or even magnesium milk ("leche de magnesia", magnesium hydroxide, it is a type of laxative or used against acidity in the stomach) or cooking oil. These are common practises amongst the farmers, but to drink milk or cooking oil after spraying can actually have negative effects on them and it is strictly recommended not to do so<sup>38</sup>.

During the period of 2001-2004 there were 483 cases of poisoning due to pesticides reported in the department of San Martin. In comparison to a CICOTOX (Centro de Información. Control Toxicológico y Apoyo a la Gestión Ambiental) study that have reported 3067 cases of poisoning due to pesticides in Metropolitan Lima during 1997-2000 (RAAA 2002). These figures correspond to 86 ppm (86 persons per million) of the population being poisoned every year in Metropolitan Lima and 153ppm of the population in the Department of San Martin

**Table 10.** Comparison of cases of poisoning between metropolitan Lima and San Martin

Part of Peru	Cases of poisoning due to Pesticides	ppm of the population poisoned every year
Metropolitan Lima	$3067^{39}$	86 <sup>40</sup>
Department of San Martin	483 <sup>41</sup>	153

The cases of poisoning due to pesticides in San Martin do not necessary have to be connected to the rice cultivation. Pesticides are used in almost all commercial cultivations, like coffee, tobacco or coca. It is also the most accessible toxic chemicals for the population and there might be cases of intoxication with pesticides that are not connected with spraying of pesticides at all. Pesticides are often used as a mean of committing suicide. Depending on where the farmer stores the pesticides it can also accidentally be consumed by children. Among the data we collected from the Ministry of health San Martin was a report of a 2-year-old child being poisoned by pesticides; this was surely an accident.

# 4.3 Summary of the other two studies

SPE (Solid phase extraction) columns were used on the 24 water samples from the irrigation canals, rice fields, small streams, ground water and the river C. The SPE columns and filters from each sample were analysed on GC-ECD and/or GC- MS and screened for 28 substances. So far, Britta Palm concluded that the following substances were found in the water samples from natural watercourses, irrigation channels and the rice field; Endosulfane-sulfate, Endosulfane-alfa, Endosulfane-beta, Butachlor and Parathion-methyl. Interestingly the highest concentration of the pesticide Butachlor was detected in a small stream, not in one of the irrigation canals, that runs through the agricultural area. The irrigation canals contained the highest concentration of Endosulfane-sulfate, Endosulfane-alfa and a water sample from a rice field the highest Endosulfane-beta concentration. Parathion-methyl was found in the same stream with the highest concentration of Butachlor and in a sample from an irrigation channel. In all water samples Metamidofos, Carbofurane and Carbosulfan was found.

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<sup>&</sup>lt;sup>38</sup> In the *Tratmiento de la intoxicación por plaguicidas-Guia para médico*, from the cooperation Bayer this is not recommended, you can also find a warning texts on some of the pesticides labels against this practice for example on metamidofos

<sup>&</sup>lt;sup>39</sup> 1997-2000

<sup>&</sup>lt;sup>40</sup> Based on a population of 8866160, figure taken from www.wikepedia.org

<sup>&</sup>lt;sup>41</sup> Figures taken from information from Ministry of health San Martin

Some of the analysis of the filters from the water samples were made on pooled samples since the amount of particles sometimes was so too low to analysis one sample at a time. Endosulfane-alfa, Endosulfane-beta, Endosulfane-sulfate, α-cypermetrin and p,p-DDT were found in the filters from the pooled water samples from the irrigation channels and water from the fields. It was a big surprise to find p,p-DDT in the filters from samples from the rice field and an irrigation channel since all of the information gathered in this study showed that there is no DDT used in this region. There might be a plausible explanation for this but as today this is not cleared

Agneta Andersson concluded in her study (Andersson, 2005), that the farmers are aware of the danger of using pesticides and that the labelling is sufficient according to FAO standards but that the farmers despite this have problems with the pesticides and use them in an inadequate way. She states that there is a problem within the information from the distributors/vendors since they are the primary source of information about pesticides to the farmers. Often the distributors/vendors also have a close relation to the farmers and they use what the distributors/vendors recommended them; highly toxic products. The farmers are also often told by them to drink milk to cure poisoning and only 50% of the distributors/vendors she interviewed claimed to give safety instructions. It is not clear if the situation would be different if the farmers would be given better information from the distributors/vendors.

#### 4. 4. Sources of error

One source of error could be that during step 17 in the extraction (See **Appendix VI**), where the samples had to evaporate to dryness; the samples were put in a fume cupboard for one entire week. The fume cupboard was not completely closed so one cannot exclude the possibility of contamination from here. During the transportation from Peru to Sweden the samples could not be kept frozen the whole time. The fact that the samples were frozen and refrozen several times may also have affected the results.

The concentrations in the samples are stated in ng/g fresh weight since fat weight in the blood was not determined. This lead to that the comparison to other studies, had to be done with approximate concentrations calculated assuming that every person had 0.5% fat in their blood. This figure actually differs from person to person.

Since the concentrations of the substances are stated in ng/g fresh weight and not in ng/g lipid the concentrations amongst the female samples might be slightly higher since women in general accumulate more fat in their body and might have a higher content of fat in the plasma. This has not been considered while analysing the results.

The sample pool was small and can only give an indication of what concentration of pesticides the farmers might have in the blood in this area.

The pesticides and their metabolites screened and quantified for are not used any more in the region and no indications were found of the opposite.

The group of reference person could have been bigger. The proportions of female and male are the opposite of the sampled farmers and the age distribution is not correlated with the group of samples. We have blood samples from tow other reference persons, a man (45) and a woman (30) from the region, but these samples are not analysed. If their values would be

included they would probably give us some more valuable information used in the reference group.

# 5. Future efforts and concluding remarks

A great possibility would be to analyze the back-up samples and the two reference samples not yet analysed, with other extraction techniques in an attempt to try to extract some of the "new" pesticides, especially the ones with high Log  $K_{ow}$  that are more likely to be found in the plasma. It would also be of interest to identify the unknown peak that was detected in many samples.

It would also be interesting to take urine samples from exposed people and analyse them to try to detect some of the more water-soluble substances.

Another thought would be to take new blood samples from me and Britta to see if there have been any changes of the concentrations in our samples. It would be especially interesting to see if there would be any change of the concentration of Pentachloroanisole since one then might me able to determine if the origin of the pollutant was from the region or not.

One could observe that the whole family was involved in the agricultural work in different ways. Hence it would be a good idea, to try to analyse the blood of the entire family and compare them to families not involved in any agricultural work. This would be done to see if the concentrations differ and if so, how the use of pesticides affects the whole family.

It would also be interesting to do a comparison with a group of farmers cultivating rice from different regions or countries. One suitable area for this is on the cost around Chiclayo in the department of Lambayeque, where largest production of rice in Peru is situated.

Another important matter would be to try to analyse some of the chronic effects from the pesticides. An option is neurological studies like it has been done in a Costa Rican study (Wesseling, 1997).

It would also be of interested to use a Test Mate-Che on samples from the farmers in the region. This device measures the activity of Acetyl Cholinesterase. The information could be used in comparison to the study that was made by RAAA during 2003 where they made this test on a number of farmers in the same region where this study was performed. They found that 1 out of 6 farmers presented chronic symptoms related to the use of organophosphates and carbamates due to a lower concentration of the enzyme in the blood serum (Gomero et Al. 2003).

The agricultural conditions that lead to an extensive use of pesticides are: The possibility of cultivating all year round, combined with the lack of coordination amongst the farmers provides the pests with an optimal situation. They can move from one field to another. When one farmer harvest another one might just transplant his plants to the great field where the pests can continue to feed. The pests are becoming more and more tolerate against the pesticides and all the farmers said that they have increased the use of pesticides during the time they have cultivated rice.

The cultivation practice of cabrilla means that the life cycles from the insects are not broken and plant disease might transfer from the old plant to the new. This also enhances the use of

pesticides. Since rice is not an ingenuous crop it is also more sensible to the pests from the region than the natural plants. Monocultures are also more susceptible to pest attacks than multicropp systems.

Some other factors I personally believe to contribute to the unsafe use of highly toxic pesticides are the poverty. The farmers have a responsibility towards their family, to be able to feed them. Since there is no social security provided by the state one depends on receiving a good yield, without it one has no income and hence no food. This often leads to decision based on short time effects and pesticides quickly kill the pest that might pose a threat to the yield. Many pesticides have may have chronic effects and they are not considered in the decision-making. Often the macho ideal also is a factor and the farmers know that pesticides might be toxic to them but the say that "a man can handle a little bit of poison".

The lack of control of implementations of laws concerning pesticides and its' handling is also a fact that influences the use of pesticides. Another fact is that the distributors preferentially recommend the most toxic pesticides since they are held responsible for the yield. The rules and conventions that concerns pesticides are not really anchored in the real life of the farmers in the tropical regions were it is very hot and hard to wear any kind of protective clothing. The companies producing the products have a responsibility towards their buyers, and enduser. They are not supposed to sell the products if they cannot assure that they will be handled properly. This is especially important with the pesticides classified as 1a and 1b according to the WHO (As mentioned before they are frequently used in the study region).

Knowledge is responsibility!

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Homepage on the Stockholm convention of POPs <a href="http://www.pops.int/">http://www.pops.int/</a>	2006-02-20
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RX list-the internet Drug index <a href="http://www.rxlist.com/cgi/generic/heparin_cp.htm">http://www.rxlist.com/cgi/generic/heparin_cp.htm</a>	2005-09-26
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# **Interviews**

24 interviews and 7 deep interviews with farmers

Sixto A, SENASA San Martin, Interview.

Questionnaires filled out by 15 individuals working at health care centres

Information from interviews made with stake holders selling pesticides, performed by Agronomist Christina Bartra Garcia in behalf of CIP

# **Other Sources of Information**

Data on cases of poisoning due to pesticides in San Martin. Distributed by the Ministry of health San Martin

# Appendix I

# **Conventions concerning pesticides:**

- The Convention on Long-range Transboundary Air Pollution (LRTAP) was adopted in 1979 and in 2005 it had been ratified by 50 countries. Peru is not one of these countries. It covers chemicals that travel long distance; 11 pesticides two industrial chemicals, and three by-products or contaminants<sup>42</sup>.
- The Basel Convention on the Control of Transboundary Movements of Hazardous Wastes and their Disposal, adopted in 1989 and entered into force in 1992. The goal is that the waste should be dealt with as close to where it is produced as possible and not be shipped over to other countries. All transboundary movement that does occur with hazardous waste has to be documented and accepted by the different state authorities. Another goal is to minimize the amount of hazardous waste. Peru has not signed this convention<sup>43</sup>.
- The Rotterdam Convention on the Prior Informed Consent Procedure for Certain Hazardous Chemicals and Pesticides in International Trade (PIC) adopted in 1998 and entered into force 2004. It is a type of warning system and concerns bans and restrictions on pesticides. When a pesticide has been banned by two countries in two regions in the world (there are 7 according to the convention) they are put in a PIC List. Countries importing and exporting pesticide have to check with this list and the goal is to enable the world to monitor and control the trade in certain hazardous chemicals. Currently this list holds 39 chemicals, 24 pesticides, 4 severely hazardous pesticide formulations<sup>44</sup> and 11 industrial chemicals. Peru has signed this convention<sup>45</sup>.
- The Stockholm Convention on Persistent Organic Pollutants, adopted in 2001 entered into force the 2005. The Stockholm convention was formed to protect humans and the environment from Persistent organic pollutants POP's. The Convention concerns 12 different chemicals with 9 of them, being pesticides. The goal of the convention is to eliminate and reduce the abundance in the environment. Peru has signed this convention 46 and all of the substances are forbidden 47.

 $<sup>^{42}</sup>$  Homepage of The Convention on Long-range Transboundary Air Pollution <a href="http://www.unece.org/env/lrtap/">http://www.unece.org/env/lrtap/</a> 2006-02-20

Homepage of the Basel convention www.basel.int 2006-02-20

<sup>44</sup> Severely hazardous pesticide formulations that present a hazard under the conditions of use in developing country Parties or Parties with economies in transition may also be included from the convention text

<sup>&</sup>lt;sup>45</sup> Homepage of the Rotterdam Convention on the Prior Informed Consent Procedure for Certain Hazardous Chemicals and Pesticides in International Trade <a href="http://www.pic.int/2006-02-20">http://www.pic.int/2006-02-20</a>

<sup>&</sup>lt;sup>46</sup> Homepage of The Stockholm Convention on Persistent Organic Pollutants, <a href="http://www.pops.int/">http://www.pops.int/</a> 2006-02-20

<sup>&</sup>lt;sup>47</sup> Aldrin, Endrine, Dieldrin, Heptachlor, Canfechlor/ Toxaphene, and DDT since 1991. Chlordane and Hexachlorbenzene in 1999 and the last two, Lindane and Mirex in 2000

# Appendix II

# Pesticides included in the list "Dirty Dozen" and POPs

The pesticides included in the Dirty dozen and the POP's of the Stockholm convention

Name of active ingredient	Type of pesticide	Usage <sup>48</sup>	DD/POP	LD <sub>50</sub> <sup>49</sup>
Aldicarb	Insecticide (acaricide and nematicide)	Applied in the soil for the cultivation of cotton, sugar beets, potatoes, peacans, oranges, soybeans, peanuts and ornamentals	DD	0.9
Aldrin	Insecticide	Used as seed dressing and against pest in the cultivation of maize, cotton and potatoes	DD/POP	39
Chlordane	Insecticide	Used in cultivations, gardens and forest	DD/POP	283
Chlordimeform	Insecticide	Mainly used in the cultivation of cotton	DD	170
DBCPT (dibromochloropropane)	Soil fumgant- nematicide		DD	170
DDT	Insecticide	Used in the past to control mosquitoes that were carriers of plasmodium (the parasite that causes malaria) and is still used in some countries to control the vectors	DD/POP	87
Dieldrin	Insecticide	Used in the fruit cultivation, against pest in the cultivation of maize, cotton and potato and to control ectoparasites in livestock/cattle	DD/POP	40
EDB (ethylene dibromide)	Insecticide	Used in the soil, in buildings as well like warehouses, mills and households	DD	146

 $<sup>^{48}</sup>$  (Ware 1994)  $^{49}$  Oral LD  $_{50}$  in mg/kg in rats. All values from (Ware, 1994)

Endrine	Rodenticide and insecticide	Used in the cultivation of maize, potato and rice and to control ectoparasites in livestock/cattle	DD/POP	3
HCH/BHC (benzene hexachloride)	Insecticide		DD	125
Heptachlor	Insecticide	Used to kill soil insects, termites, cotton insects, grasshoppers, other crop pests, and malaria-parasite carrying mosquitoes.	DD/POP	40
Hexachlorbenzene (HCB)	Fungicide	Used a seed dressing for crops such as wheat, barley, oats and rye to prevent growth of fungi. Largest source today is combustion	DD/POP	10
Lindane (γ-HCH)	Insecticide	Used in many different areas like seed treatment and some fumigant action	DD	76
Paraquat	Herbicide	Contact herbicide and desiccant with broad scope of uses	DD	150
Metyl parathion	Insecticide	Controls most cotton pest including non-resistant bollworms, tobacco budworms	DD	9
Mirex	Insecticide	Used in different cultivations to control ants and termites	POP	235
Pentachlorophenol (PCP)	Fungicide	Wood preservative	DD	50
2,4,5-T	Herbicide	Woody plant control and amine form used in rice	DD	500
Toxaphene	Insecticide	Used in the past, mainly in the cultivation of cotton, maize, cereals and vegetables	DD/POP	40

# **Appendix III**

# Inventory of the Pesticides used by the farmers in the rice cultivation in the Districts of Morales and Cacatachi, Tarapoto, San Martin, Peru

# Commercial name and active ingredient

The inventory is based on interviews; where the farmers told us what kind of pesticides they use and on observations; pesticides we found in the field. In total there are 61 pesticides and 31 active ingredients used (14 Fungicides 10.a.i, 28 insecticides 13. a.i and 19 herbicides 8 a.i). The inventory holds 14 pesticides (6 are amongst the most commonly used pesticides) containing 6 active ingredients that are classified as Ia or Ib.

Fungicide class

Classification

Log P

 $LD_{50}$ 

The most commonly used pesticides

Active ingredient /

# **Fungicides**

Commercial name

	CAS		WHO/Human health Hazard* (ADI mg/kg b.w)	(LogK <sub>OW)</sub>	mg/kg (oral)
Azufrac	Sulphur/7704-34-9	Multie-site inorganic	U/Non registered (No data)	Practically insoluble in water	>5000
Top Cop	Copper Sulfate/1333- 22-8	Multie-site inorganic	II/Not found (No data)	Insoluble in water and organic solvents	100
Antracol Invento	Propineb /12701-83-9	Multi-site alkylenebis (dithiocarbamate)	U/Non registered (0,007)	-0.26	>5000
Escudo Dithane M-45 Manzate	Mancozeb/8018-01-7	Multi-site alkylenebis (dithiocarbamate)	U/Recognized: carcinogen. Susp. endocrine toxicant, immunotoxicant, skin or sense organ toxicant (0.05)	0.26	>5000
Fordazim	Carbendazim/ 10605-21-7	Benzimidazole	U/Susp. Carcinogen, endocrine toxicant, neurotoxicant, respiratory toxicant (0,03)	1.38	6400
Fuji-one 40	Isoprothiolane/ 50512-35-1	Dithiolane	III/Non registered (No value)	3.3	1190
Hinosan	Edifenphos/17109-49-8	Phosphorothiolate	Ib/Non registered (0.003)	3.38	100-260
Silvacur	Tebuconazole + triadimenol	Triazole	III/Tebucanazole suspected. Carcinogen /Triadimenol susp. Carcinogen, endocrine toxicant, neurotoxicant (0,05)	No figure found	>2000 <sup>50</sup>

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<sup>&</sup>lt;sup>50</sup> The information for this fungicide is taken from the Homepage of Bayer <u>www.bayercropscience-ca.comn</u> since it is a mixture of two active ingridients

Folicur Orius	Tebuconazole/ 107534- 96-3	Triazole	III/Suspected. Carcinogen (0.03)	3.7	4000
Score	Difenoconazole/119446 -68-3	Triazole	III/Susp. Carcinogen (0,01)	4.4	1453

# Insekticides

Commercial name	Active ingredient / CAS	Insecticide class	Classification WHO /Human health Hazard* (ADI mg/kg b.w)	Log P (Log K <sub>OW</sub> )	LD <sub>50</sub> mg/kg (oral)
Carbodan Curater Furadan	Carbofuran/1563-66- 2, 1563-38-8	Carbamate	Ia alt. Ib depending on the concentration/Susp. Gastrointestinal or liver toxicant, Immunotoxicant, Neurotoxicant, reproductive toxicant skin or sense organ toxicant (0.002)	1.52	8
Marshal	Carbosulfan/55285- 14-8	Carbamate	Ib /Not registered (0.01)	5.4	250
Thiodan Star 3	Endosulfan/115-29-7	Cyclodiene	Ib alt. II depending on the concentration/Susp. Gastrointestinal or liver toxicant, Immunotoxicant, Neurotoxicant, reproductive toxicant, skin or sense organ toxicant, Kidney toxicant, respiratory toxicant, Cardiovascular or Blood Toxicant, Endocrine toxicant (0.006)	α-=4.74 β-=4.79	70
Caporal 540 EC	Cypermetrin + methamidofos	Combination	Ib alt. II depending on the concentration (No data)	Depending on the formulation	Depending on the formulatio
Rescate	Acetamiprid/135410- 20-7	Neonicotinoid	Not listed/Not found (0.066)	0.8	217
Cigaral Confidor <i>Lancer</i>	Imidacloprid/138261- 41-3	Neonicotinoid	II/Not found (0.06)	0.57	450
Actara	Thiamethoxam/15371 9-23-4	Neonicotinoid	III <sup>51</sup> /Susp. Carcinogen ( <i>No value</i> )	-0.13	1563
Metasac Monitor Tamaron Stermin Matador S-kemata	Methamidofos/10265- 92-6	Organophosphate	Ia alt. Ib dependening on the concentration /Susp. Neurotoxicant, skin or sense organ toxicant, (0.004)	-0.8	15.6

<sup>51</sup> This Classification was taken from *The Pesticide Manual* 

Cypercor Fastac	Alpha-cypermetrin /67375-30-8	Pyrethroid	II/Susp. Carcinogen, endocrine toxicant, neurotoxicant, Immunotoxicant, Gastrointestinal or liver toxicant, reproductive toxicant (0,02)	6.94	57
Affly Sherpa Corcell Hortrin Cypermex	Cypermetrin/ 52315- 07-8	Pyrethroid	II/Pot. Cancerogenic (0,05)	6.6	250-4150
Baytroid	Cyfluthrin/68359-37-5	Pyrethroid	II/Susp. Gastrointestinal or liver toxicant, Neurotoxicant, Kidney toxicant, reproductive toxicant (0.02)	6.0	500
Berlmark	Fenvalerate/51630-58-1	Pyrethroid	II/Susp. Gastrointestinal or liver toxicant, Immunotoxicant, Neurotoxicant, skin or sense organ toxicant, Cardiovascular or Blood Toxicant, Endocrine toxicant (0.02)	5.01	451
Hortiquim	Permethrin/ 52645- 53-1	Pyrethroid	II/Susp. Carcinogen Gastrointestinal or liver toxicant, Neurotoxicant, reproductive toxicant, Endocrine toxicant (0.05)	6.01	430-4000

# Herbicider

Commercial name	Active ingredient / CAS	Herbicide class	Classification WHO /Human health Hazard* (ADI mg/kg b.w)	Log P (Log K <sub>OW</sub> )	LD <sub>50</sub> mg/kg (oral)
Hachazo Machete Chem-Rice	Butachlor /23184-66-9	Chloroacetamide	U/susp. Carcinogen (No Data)	$4.5^{52}$	2000
Arsenal	Imazapyr/81334-34-1	Imidazolinone	U/Non registered (No data)	0.11	>5000

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<sup>&</sup>lt;sup>52</sup> This data was not found in the Pesticide Manual and hence the information was found in the SRC Physpropdatabase http://esc.syrres.com/interkow/webprop.exe?CAS=23184-66-9

Hedonal Sanamina Aminacrys	2,4-D/94-75-7	Phenoxycarboxylic acid	II/Susp.Carcinogen, Gastrointestinal or liver toxicant, Developmental toxicant, Neurotoxicant, reproductive toxicant, skin or sense organ toxicant, respiratory toxicant, Cardiovascular or Blood Toxicant, Skin or Sense organ toxicant (0.01)	2.58-2.83	639-764
Roundup Glifonox Balazo Rango Bazuka Batalla Destructor Sikosto	Glyphosate/1071-83-6	Phosphonates	U/Susp. Cardiovascular or Blood Toxicant, Gastrointestinal or liver toxicant, Neurotoxicant, reproductive toxicant, respiratory toxicant (0.3)	<-3.2	5600
Nominee	Bispyribac sodium/125401-75-4	Pyrimidinyloxybenz oic	U/Not Found (0.011)	-1.03	4111
Oryza	Cyclosulfamuron/13684 9-15-5	Sulfonylurea	U/Not found (No data)	1.58	>5000
Metsul	Metsulfuron methyl/74223-64-6	Sulfonylurea	U/Non registered (0.22)	0.018	>5000
Saturn	Bentiocarb/thiobencarb/ 28249-77-6	Thiocarbamate	II/Susp. Neurotoxicant (0.009)	3.42	1033

<sup>\*</sup>The information is taken the 22 July 2005 and 27 April 2006 from the webpage: <a href="http://www.scorecard.org/chemical-profiles/">http://www.scorecard.org/chemical-profiles/</a>

This website compiles information from different databases with information about the effects of chemicals

All values if not said otherwise are taken from *The pesticide Manual* Thirteenth Edition. Editor: C D S Tomlin. BCPC, Hampshire, 2003

The classifications and the two tables below are taken from WHO recommended Classification of pesticide by hazard and guidelines to classification 2004 published by April 12, 2005, incorporated. IPCS

Class		LD <sub>50</sub> for the rat (mg/kg body weight)				
		Oı	ral	De	rmal	
		Solids <sup>≛</sup>	Liquids <sup>a</sup>	Solids <sup>a</sup>	Liquids <sup>a</sup>	
Ia	Extremely hazardous	5 or less	20 or less	10 or less	40 or less	
Ιb	Highly hazardous	5 - 50	20 - 200	10-100	40 - 400	
II	Moderately hazardous	50 - 500	200 - 2000	100-1000	400 - 4000	
Ш	Slightly hazardous <sup>b</sup>	Over 500	Over 2000	Over 1000	Over 4000 <u>b</u>	
U	Unlikely to present acute hazard in normal use	Over 2000	Over 3000	Over 4000	Over 6000	

# Appendix IV

# A schedule for spraying pesticide during one campaña for one farmer

In total 180l/ha (pesticides+ water) is used on this field

Order	Place	Age of rice (days)	Туре	Name	Active substance	Amount used (ml or g)/ ha	Conc. of active substance
1	Field	50	Insec.	Thiodan35EC	endosulphane	500ml	350g/l
				Cigaral35EC	imidacloprid	100g	350g/l
			Fung.	Antracol700WP	propineb	1000g	700g/kg
2		75	Insec.	Marshal25EC?	carbosulphan	500ml	25?
			Fung.	Fordazim5FW	benzamidasole carbendazim (liger.)	500ml	500g/l?
3		100	Insec.	Stermin600SL (altam)	methamidophos	750	600g/l?
				Fastac	cypermethrin	250	
			Fung.	Fordazim	benzimidazole carbendazim	500ml	
				Folicur250EW	tebuconazole	250ml	250g/l
			Bio- stimulant	Stimplex??	citokinin	250	
			Fert. Foliars	Belfruto	N, P, K etc		
4		115	Insec.	Stermin600SL	methamidophos	750	
			Fung.	Antracol700WP	propineb	1000g	
				Lancer**	imidacloprid		
			Fert. Foliars	Quimifol**	N, P, K, mfl		

<sup>\*</sup>One of those pesticides of their kind is used at that fumigation

**Source**: One of the farmers in the region

Compiled and calculations are all made by Britta Palm

<sup>\*\*</sup> Used if necesarry

Appendix V  Questionnaire to farmers at the blood analysis
Provnr:
Namn:
Adress:
17.0
Kön:
Ålder:
Grado de instrucción:
Distrikt:
Antal ha ris:
Bevattning?
1.Vart förvaras bekämpningsmedlen?  ¤ I hemmet  ¤ på chakran  ¤ Annan ort:
2. Besprutar du själv?: ¤ Ja ¤Nej Om Ja, hur många år har du besprutat? Hur många ggr. per campaña besprutar du? Hur många campañas/år har du?
3.Använder du någon skyddsutrustning när du besprutar? ¤Ja ¤Nej Om, ja, va för slags sådan?
långärmat långbyxor ansiktsmask handskar stövlar öronskydd
Annat:
4. Nar och med vad besprutade du senast?
5.Om Nej, på fråga 2,Vem besprutar på ditt fält?
6.Har du själv någonsin besprutat? ¤Ja ¤Nej

På vi	lken slags odl	ing?				
¤Blaı ¤Tvä ¤Tvä ¤Är ı	ndar ihop den ttar kläderna ttar Mochilan ned ute på fäl	1 som använts 1 som används It vid bespruti	vid besprutni S		?	
8. Vilka bekämpningsmedel använder/används (på ditt fält) du?						
Antracol Lanc	Hedonal er	Nominee	Furadan	Tammaron	Thiodan	
Cypermex Mach		Fuji-One	Stermin	Folicur	Sherpa	
Chem-Rice	Andra:					
	ågonsin känt o Ja, på vilket så	O 1	förgiftad av b	ekämpningsme	del? ¤Ja   ¤Nej	
Illamående kräkningar yr diarre utslag magont						
"brännskador" ont i huvudet domningar						
Annat:						
10. Äter elle	r dricker du 1	ıågot speciellt	innan och eft	er du bespruta	t?	
Övrig infori	nation:					

Om, Ja, När då? Hur länge?

#### Appendix VI

# Chemicals, equipment and detailed method description

### Chemicals

**Standard mixture containing:** cbpest 10ng;  $\alpha$ -HCH, HCB,  $\beta$ -HCH,  $\gamma$ -HCH,  $\delta$ -HCH, Heptachlor,  $\alpha$ -chlordene, Aldrin, Heptachlorepoxide,  $\gamma$ -chlordane, Oxychlordane,  $\alpha$ -chlordane, Nonachlor, p,p –DDE, DDD, p,p-DDT, Dieldrin, Endrine, Metoxychlor, Mirex and Transnonachlor

**Internal standard** CB-189 with a concentration of the stock solution of 74.9 μg/ml **Solvents**: *In Peru*: HCl (6 M) (Peru), *iso*-propanol (pesticide quality), cyclohexane (pesticide quality), dichloromethane (pesticide quality) from Cimatec S.A (Lima, Peru) *At IMK*: Cyclohexane (pesticide quality), normalhexane (pesticide quality) and dichloromethane (DCM) (pesticide quality) from Merck (Darmastadt, Germany) sulfuric acid 98% from AnalR (BDH laboratory supplies poole England), SiO<sub>2</sub> (that has been activated and conditioned over night at 300°C)

# Equipment

**GC-apparatus**; Varian 3400 with a Varian 8200 autosampler and an electron capture detector (ECD). A CP-Sil 8 column (25 m x 0.15 mm i.d. and 0.12 μm film thickness from J&W Scientific, Folsom, CA, USA) was used for the analyses. The temperature program was as follows: 80°C (2 min), 20°C min<sup>-1</sup> to 300°C (10 min). The injector temperature was 250°C and the detector temperature was 360°C. The injector was operated in the splitless mode. Nitrogen was used as make-up gas and hydrogen as carrier gas of highest purity. The data was collected and processed by Elds data system (Chromatographic Data System AB, Stockholm)

Centrifuge, water bath (37~40°C), vibromix, Grant block heater (Grant Instruments, Camebridgeshire Great Britain) apparatus (45 °C). Nitrogen. GC-columns, Glass test tubes with heparin (5 ml)<sup>53</sup>, glass test tubes 8 ml and 12 ml, Pasteur pipettes, sterilized cannulas

## Extraction and clean up

- 1. Glass test tubes (50) were weighed and marked in Sweden.
- 2. Three blood samples (4 ml each) in glass test tubes with heparin, was taken from each person.
- 3. The blood samples were centrifuged for about 10 min.
- 4. Empty test tubes were put on the balance and reset to zero.
- 5. Plasma from two test tubes was transferred into a new empty test tube and plasma from the third test tube was transferred into a new empty test tube and was kept as a back-up.
- 6. The weight of the plasma was noted.
- 7. The test tubes containing plasma that was kept as back-up was put in the freezer.

The following extraction was made with the 29 samples taken on the 9<sup>th</sup> and 10<sup>th</sup> of July 8. To the test tubes were added HCl (6 M, 1 ml) and iso-propanol (2 ml) and mixed properly. (During the preparation of the 13 first blood samples from farmers and the blood tests from Britta and Gun the steps between 7, 8 and 9 took some more time then for the rest of the samples.)

9. The mix was divided into two test tubes

-

<sup>&</sup>lt;sup>53</sup> From Simonsen Articlenumberr, 205378

- 10. To every test tube was added additional cyclohexane (3 ml) and dichloromethane (1 ml),
- i.e. a total of 6 ml cyclohexane and 2 ml dichloromethane for every sample.
- 11. The samples were rocked carefully for about 5 min.
- 12. The samples were centrifugated for 3 min.
- 13. The organic phase was pipetted over in to new test tubes.
- 14. The remaining water phases were washed with cyclohexane (3 ml), rocked (3 min) and centrifuged in (3 min), a total of 6 ml of cyclohexane for every sample.
- 15. The organic phase was pipetted over into the same test tubes again. Some of the organic phase was left in the "old" test tubes since the new test tube could not hold such a great volume.
- 16. All of the test tubes were put in the freezer for a couple of days.
- 17. Then, only test tubes containing the organic phase were put in a warmwater bath (37~40 °C) in a fume cup to evaporate to dryness. This step took 1 week.
- 18. The samples were kept in a freezer during the rest of the stay in Peru and as cold as possible during the travel. Back in Sweden they were kept in a freezer until step 19.
- 19. The analysis continued at the department of Environmental chemistry, IMK Stockholm University. The organic phase from the two test tubes containing the "left over" from step 15 for every sample, was added to the test tube with the evaporated fat from step 17. (Sample nr. 6 and 13 only had lost one test tube containing "left overs" and sample nr. 10 had very little in one of the "left over" test tubes).
- 20. The mixture was added cyclohexane to a total volume of 4 ml. The test tubes were then mixed on a vibromix for 20 sec to dissolve the fat.
- 21. Concentrated  $H_2SO_4$  (1 ml) was added to every test tube. Thereafter the samples were carefully rocked back and forth ca. 30 times and centrifugated for 3 minutes at 3000rpm. You could clearly see two phases. One transparent neutral fraction phase and one coloured with fat, conc.  $H_2SO_4$  and different pigments.
- 22. The neutral fraction was transferred to a new test tube.
- 23. The first test tube was added cyclohexane to a total volume of 4 ml and rocked 10 times and then centrifugated for 3 min.
- 24. Two phases appeared again, one neutral and one coloured/H<sub>2</sub>SO<sub>4</sub> fraction. The neutral fraction was pipetted to the same test tube as in step 22.

Every sample was now divided into two test tubes with one neutral fraction and one coloured with the remaining of fat conc.  $H_2SO_4$  and different pigments. These two different phases were analysed in two different ways.

# Analysis of "coloured phase"/ $H_2SO_4$ fraction. This analysis was only performed with sample nr. 1 and 2. It was a test to see if it was possible to find some more interesting substances in the sample.

- 25. The test tube that only contained a coloured phase was once again added cyclohexane to a total volume of 4 ml. Two phases were formed.
- 26. KPLC-water was added, until the ratio between the water phase and the coloured phase was 2:1. The samples were shakened carefully. This caused a chemical reaction, that generated heat and therefore the samples were put in a cold-water bath. (During this step, half of the organic phase of sample 4 was spilled out and parts of the organic phase of sample 21).
- 27. After the samples had cooled down a bit, they were rocked about 15 times and centrifuged for 3 min. (Sample nr. 4 and 21 were added cyclohexane to obtain the same volume as the other samples, before they were centrifugated)
- 28. The phase solved in cyclohexane, of the sample, was put into a new test tube. The old test tubes were once again added cyclohexane to the same volume the samples had at step 26.

- 29. The samples were slowly rocked 15 times and thereafter centrifugated for 3 min. The solvent phase was once again transferred to the test-tube, containing the phase from the cleanup step before.
- 30. The samples were evaporated to complete dryness (under a gentle stream of nitrogen and a Grant block heater)
- 31. Each sample was cleaned on a separate colon made out of a washed and burnt Pasteur pipette containing  $SiO/H_2SO_4$  (2:1, 1 g) and a small piece of glass wool. Before the samples were added every colon was washed/rinsed with dichloromethane (3 ml). The samples were dissolved in dichloromethane (10 ml) and mixed with a vibromix and then put on the column. The samples were diluted to a total of 33 ml (including the 10ml used to dissolve the evaporated sample) dichloromethane and four fractions were collected 1-10 ml, 10-20 ml, 20-25 ml and 25-33 ml. The fractions were all transparent and the columns were yellow after the cleanup (The two last steps were made a day later with sample 1 were the column had become dry in-between)
- 32. The four fractions were evaporated to complete dryness and re-dissolved in hexane (1 ml).
- 33. The fractions were then analysed with the gas chromatography with electron capture detector GC/ECD and then on the mass spectrometry, MS. Before the GC/ECD analysis the samples were added internal standard CB-189 (200  $\mu$ l, 74.9  $\mu$ g/ml) and transferred into GC-vials

#### **Analysis of the Neutral Fraction**

- 25. The samples were evaporated to dryness, and a red precipitate remained in the test tubes 26. Each sample was cleaned on a separate colon made of a washed and burnt Pasteur pipette containing SiO/H2SO4 (2:1, 0.5 g) and a small piece of glass wool. Before the sample was added to the colon, it was cleaned with 3 ml of cyclohexane. From sample 8 on, hexane was used as a solvent, instead.
- 27. The samples were all added a bit of the cyclohexane/hexane to resolve them, by mixing them on the Vibromix (there were some troubles resolving the "red dots".) The samples were then put on the colon and they were allowed to "go through" the colon before they were diluted with 6 ml of cyclohexane/hexane (The cyclohexane/hexane used to resolve the sample is taken from this volume.). During this step sample 8 and 9 accidentally were put together and a fraction containing hexane (3 ml) from the washing of the colon for sample 8 and 3 ml hexane from the washing of the colon for sample 9, together with sample 8 and 9 and 2, 5 ml from every colon, added after the sample was used in the further analysis. This sample had a total volume of about 11 ml.
- 28. The samples were evaporated to a volume of approximately  $1.5\ \mathrm{ml}.$
- (During this time a test was performed with sample 1 were one part was metylazed to see if it would be preferable for the GC-analysis, or not. The latter was the case)
- 29. The samples were then added 200  $\mu$ l of internal standard CB-189 (concentration 74.9  $\mu$ g/ml) and transferred into GC-vials. In total there were 28 blood samples, 4 hexane samples and 3 standard samples that were analysed on the GC.
- 30. For the MS the samples nr. 1, 2, 18, 14, and 15 were pooled together. The samples were taken from the vials and transferred to a 8ml test tube and then evaporated to a volume of about 0,5ml and were then added about 10 drops of hexane.

(Method based on Hovander et al. 2000)

#### **Appendix VII**

# Interview form used during deep interviews with farmer

# Entrevista con campesino arrocero

Nombre:

Sexo:

Edad:

Grado de instrucción:

Familia:

Distrito:

# Sketchmap

# QUE'? CUANDO'? DÓNDE? QUÍEN? PORQUÉ? COMÓ?

- \* "Edificios" casa?, garage?, vehiculos? baño/sanidad?, agua?, electricidad?, basurero/residuos?
- Viven familias en los alrededores de los terrenos de arroz?
- Se encuentran escuelas cercanas?

# \* Campos de cultivos

- Tipo de cultivos?
- Número de ha?

# \* Fuentes de agua

- Que tipo? (pozo, rio, lago, canales)
- Que uso? pesca? riego? animales? consumo humano? lavar ropa? Canales de riego? Canal principal? Canal secundaria?
- De dónde viene el agua?
- Usa agua subterraneo?

# \* Calles/caminos

- \* Guardario de pesticidas/mochila
- Quien tiene acceso, responsibilidad del lugar?
- \* Conoce Ud. el tipo de suelo de su campo?
- Ha observado animales silvestres en o cerca de su campo de cultivo?
- Que tipo y con que frecuencia?

# Fieldsketchmap

- \* Dueño
- Quien/quienes es el dueño de los campos cultivados?
- Que tipo de arroz cultivan?

#### \*Los campos

- Como estan los campos distrubuidos?
- Que tiempo de crecimiento?
- \* Trabajo
- Quien trabaja en el campo el dueño/peónes/familiares/amgios?

#### \* Riego

- Que tipo?
- Fluidos (ingresa/sale)? Constante?
- De dónde viene?

- Cuantó se usa? (m<sup>3</sup>)
- Diferentes cantidades dependiente del estadio del arroz?
- Se paga por el agua? Cuantó? Acceso todo el año?
- Se usa el agua del riego por otras cosas tambien(lavar, tomar etc)

#### \* Siembra

- Tipo de siembra?
- Uso de maquinarias?

# \* Mantienemiento

- Fertilizantes?
  - o Que tipo?
- Adherentes?
  - o Que tipo?

### \*Costos/ganancias de producción?

- Costos?
  - o Ganancias?

#### \*Problemas de plagas

- Que'tipos de plagas y cuando?
- Como se hace para eliminarlos?

# \* Fumigación

\*Cuando?

#### \*Preparación

- \* Que tipos de plaugicidas usan?
- Nombres?

Fungicidas:

Insecticidas:

#### Herbicidas:

- Quien prepara los pesticidas y dónde?
- Como se hace?
- Se mezclan mas de un tipo de pesticidas?
- Que cantidad (cucharitas/mochila o mochilas/ha)?
- Tipo de envase y volumen?
- Lee la etiqueta del pesticidas antes de su manipulación?
- Considera las dosis recomendadas de aplicación?
- Frecuencia de uso(veces/campaña)?
- Realiza una evaluación previa del campo antes de la aplicación?
- Que observa?

- Respeta el periodo de carencia (periodo entre la ultima aplicación de un plaugicida y la cosecha en el cultivo?
- Porque no?

# \*Fumigación

- Quien fumiga y como aprendió?
- Tipo de mochila?
- Se usa equipo de protección(mascarilla?guantes?botas de jebe?camisa manga larga?algun plástico que cubra alguna parte del cuerpo cuando aplica?)
- Por que no usa?
- Ha participado de alguna capitación sobre los peligros del uso de plaugicidas y sus efectos? Quien lo organizó (Empresas de agroquímicos?, Agencia Agraria?, SENASA?, junta de usuarios? etc)
- Aplica en dirección de viento?
- Comes o bebes cuando fumiga?

# \*Despues fumigación

- Quien lava la mochila despues de la aplicación y donde?
- Que hace con los sobrantes de pesticidas?
- Que hace con la ropa de fumigación?
- Quien? lava la ropa?
- La ropa utlizada en la aplicación lo lava separada a la ropa de la casa?
- Usa la misma ropa siempre cuando fumiga?
- Se baña con agua y jabón todo el cuerpo?

#### \*Salud

- Ha tenido algúnas símtomas de intoxicación?
- Cuales? (mareos?, dolor de cabeza?, diareea?, visión borrosa?, náuseas? dolores en el cuerpo?etc)
- Cuandó?
- Que hizo despues?
- A dónde acude para su tratamiento?
- Cuantó tiempo de recuperación?
- Que tipo de plaugicida?

# \*Envases de plaguidas

- Que hace con el envase de plaugicidas?
- Reutiliza? agua/alimentos? se llena con otros pesticidas?
- Se junta con los residuos de la casa?Deja en el campo?Quema en el campo?entierra en el suelo? Arroja en canales de riego?

<sup>\*</sup>Como piensa Ud. que los pesticidas affecta el ambiente? salud?

# **Timeline**

- \* Cuando llegó a este lugar?
- \* Porque?
- Cuantos años se dedica a la agricultura?
- Cuantós años se dedica cultivando arroz?
- Miembro de algún comité?
  - o Cuál?
- Cambios de dueños y terrenos durante el tiempo?
  - o Alquilan?
- Ha comprado o vendido terrenos?
- \* Eventos importantes en su vida? Negativos? Positivos? niños?muertos? Formación del comité? 2002? Agua? Electricidad?
- \* Cambios de cultivos?
- Que?
- Porque?
- Cuando?
- Como?
- \* Introducción de fertilizantes/maquinarias/pesticidas?

#### Seasonal calender

#### **Símbolos**

- \*Siembra
- \*Cosecha
- \*Meses de sol
- \*Meses de lluvia (menos riego?)
- \*Plagas
- \*Fumigación (insecticidas, herbicidas, fungicidas?)
- \*Vacaciones? \*Peónes?

### Dayclock -2 días

# Día normal de trabajo

\*Hora de levantar?

Desayuno?

- \*Trabajo
- Que hace?
- Dónde?
- Con quién?
- \* Almuerzo?
- Dónde?
- Que?
- Con quien?
- \*Cuantás horas de trabajo?

<sup>\*</sup>Otras actividades despues el trabajo?

- \*Cena?
- Dónde?
- Que?
- Con quién?
- \*Hora de acostarse?

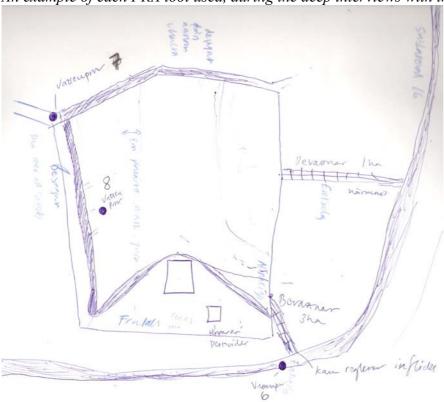
# Día de fumigación

- \*Hora de levantar?
- Desayuno?
- \*Fumigación
- Con quién?
- Cuantó dura?
- Dónde?
- \* Almuerzo?
- Dónde?
- Que?
- Con quien?
- Se lavan las manos antes de comer?
- Dónde?
- \*Cuantás horas de fumigación durante el día?
- \* Despues de la aplicación de plaugicidas que actividad realiza?
- \*Cena?
- Dónde?
- Que?
- Con quién?
- \*Hora de acostarse?
- Se sienten más cansado comparado con un día normal de trabajo?

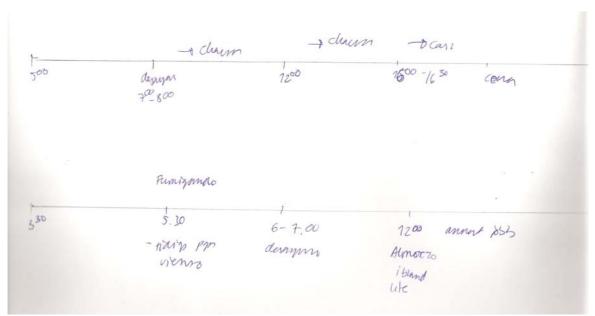
# **Appendix VIII**

#### PRA/RRA Tools

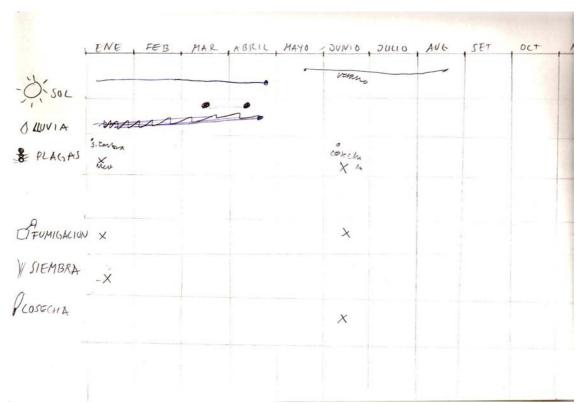
An example of each PRA tool used, during the deep interviews with the farmers.



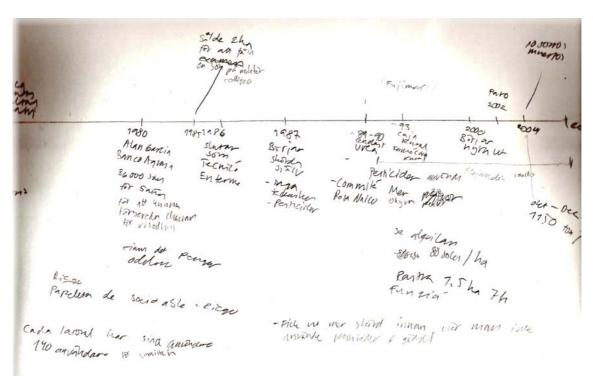
**Fig. 1.** A Field Sketch. It was drawn together with the farmer and it shows the field and the irrigation channels. Some notes about the sample sites and the use of the channels, pesticide storage and other important sites, was made on the map



**Fig. 2**. *A Day clock*. The line at the top shows how the tasks of the farmer are distributed during a day when he is not spraying. The line at the bottom shows a day when he is spraying.



**Fig 3.** A Seasonal calendar. The farmer was asked to make a cross during the month that different agricultural and seasonal events took place.



**Fig 4.** A Timeline: The line drawn represented the life of the farmer. The most important events concerning family and agriculture were marked chronologically on the line.

# **Appendix IX**

# Questionnaire to health care personnel

- 1. Nombre del establecimiento, dirección, distrito y teléfono
- 2. Grado de intrucción del encuestado (Indicar profesión y cargo)?
- 3. Cuantós profesionales de slaud laboran en este establecimiento? (listar indicando profesión y tiempo dedicad?)
- 4. Cuántos establecimientos de salud (Postas médicas, enfermerias) existen en el didtrito y cuáles son?
- 5. Ha recebido capacitaciones sobre el diagnóstico y tratamiento de intoxicaciones por plaugicidas?
  Si su repuesta fue Si, especifique en que curso, entrenamiento o charla participó (en que año)?
- 6. Tiene experiencia en el tratamiento de intoxicaciones producidas por plaugicidas? Si su repuesta fue Si, cuantos casos ha tratado? Como ha evolucionado el paciente?
- 7. El donde labora tiene un procedimiento establecido para atender intoxicaciones producidas por plaguicidas?
  Si su repuesta fue Si, especifique en qué consiste? Indicar medicamentos empleados?
- 8. Su establecimiento de salud brinda algún tipo de capacitación a los agricultores sobre primeros auxilios en caso de intoxicación por plaugicidas? Si su repuesta fue Si, con qué frequiencia? (indicar lugar, fecha y número de participantes)
- 9. En los últimos años se ha presentado casos de intoxicación por plaugicidas en su establecimientos de salud? Si su repuesta fue, por qué tipo de plaugicidas? (revisar y recoger resultados de estadísticas
- 10. Que recomienda usted a los fumigadores antes y despues la fumigación?
- 11. Conoce usted algún tratamiento casero para prevenir o curar un caso de intoxicación?

The Questions 1-9 are taken from Appendix 3 from the report. *Plaugicidas Químicos en el Peru- Propuesta participative para el Fortalecimiento de Politicas y Marco Normativo sobre plaugucidas químicos en el Perú- Investigación sobre Política y Legilación Ambiental.* by RAAA, that was published 2002. The last two questions were added by the author and Britta Palm.

# Appendix X

# Chemical structures of compounds detected in the blood samples

1,1-dichloro-2.2-bis(p-chlorophenyl)ethane p,p'DDE

Hexachlorobezene (HCB)

Pentachloroanisole (PCA)

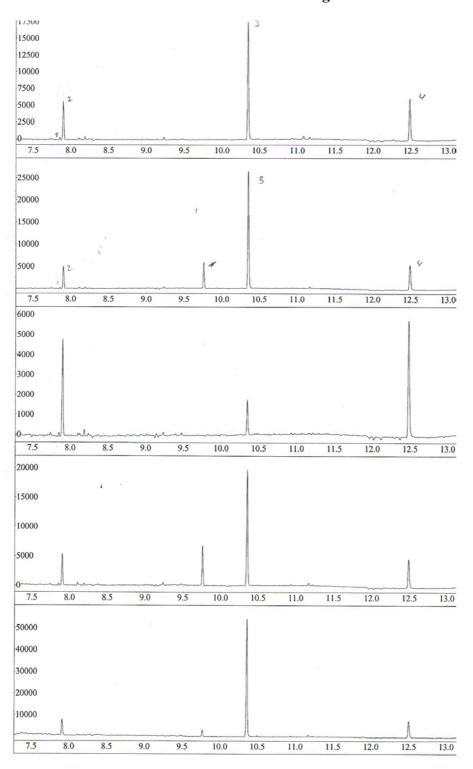
O-Hex achlor o cyclohex ane (O-HCH)

yHex achlor ocyclohex ane (yHCH)

Provided by Maria Athanasiadou

# Appendix XI

# **Chromatograms**



Chromatograms from some of the blood samples. The results are from the analysis of the neutral fraction (See **Appendix VI** for method description). The substances that were found and quantified are marked with a number in the two chromatograms on the top. Peak nr.1HCB, nr.2 Pentachloroanisole, nr.3 p,p-DDE, nr.4 the internal standard cb-189 (\* a unknown substance.)

# Appendix XII

# Concentration in blood samples of different sample groups

# **All Samples**

	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.17	0.24	13	5.9
Mean	0.29	0.27	20	7.4
Range	<0.1-0.84	<0.2-0.54	1.1-141	0.59-27.4

## **MED AGE+ (-8)**

n=11(10m.1f)

	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.20	0.26	16	7
Mean	0.32	0.28	32	8.3
Range	<0.13-0.74	<0.2-0.54	3.29-141	2.2-16

## **MED AGE - (-9)**

n=10 (9m. 1f)

	HCB (ng/g) b.p	gamma HCH (ng/g) b.p	p.p-DDE (ng/g) b.p	pentachloroanisole (ng/g) b.p
Median	0.13	0.25	7.8	5.5
Mean	0.25	0.27	12	7.6
Range	<0.1-0.84	<0.2-0.72	1.1-35	2.4-27

# SPRAYINGTIME MED+

n=9 (all m)

	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.20	0.23	13	7
Mean	0.34	0.24	17	8.6
Range	0.17-0.74	<0.2-0.52	3.3-47	2.2-16

### YEARS SPRAYING MED-

n=9 (all m)

	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.14	0.27	18	5.8
Mean	0.22	0.27	27	6.3
Range	<0.1-0.61	<0.2-0.54	1.2-141	2.4-13

### NO PROTECTIVE EQUIPMENT

n=7 (all m)

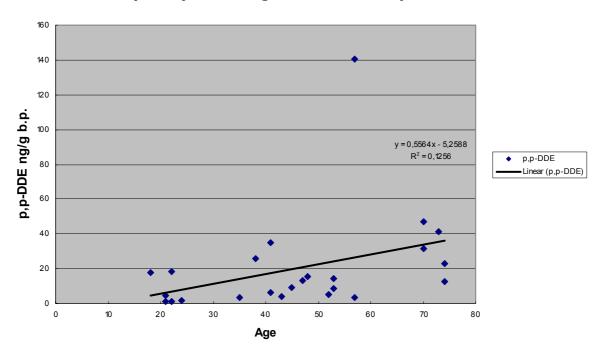
(*****)				
	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.30	0.24	14	9.2
Mean	0.35	0.26	36	9.1
Range	0.13-0.72	<0.2-0.54	5.2-141	2.4-17

# n=11 (all m) PROTECTIVE EQUIPMENT

	НСВ	gamma HCH	p.p-DDE	pentachloroanisole
	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p	(ng/g) b.p
Median	0.14	0.27	6.1	5.8
Mean	0.23	0.25	14	6.4
Range	<0.1-0.84	<0.2-0.72	1.1-47	2.2-28

### **Diagrams**

# Concentrations of p,p-DDE and Pentachloroanisole in the blood samples, plotted against different parameters



Fig, 1. Concentration of p,p-DDE in ng/g b.p. plotted against age of the farmers

#### p,p-DDE and age, without extreme value

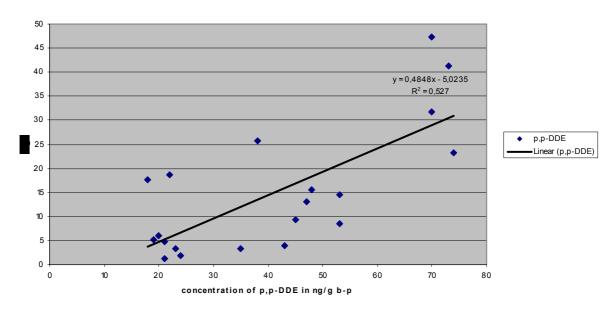


Fig. 2 Concentration of p,p-DDE in ng/g b.p. plotted against the age of farmers after removal of the extreme value

### Pentachloroanisole conc. Correlated to age

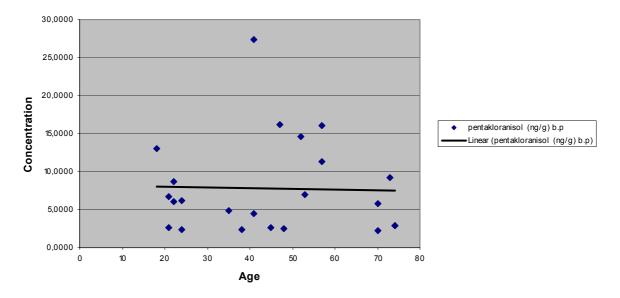


Fig 3. Concentration of Pentachloroanisole in ng/g b.p plotted against age in all the samples.

### Spraying intensity and p,p-DDE

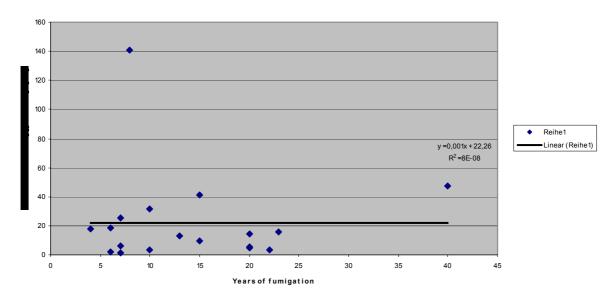
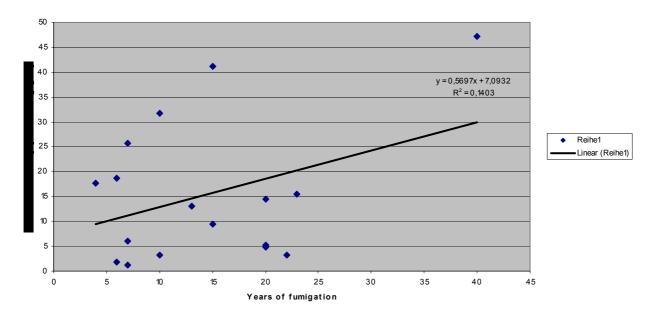


Fig 4. Concentration of p,p-DDE in ng/g b.p plotted against the amount of years the farmers have been Spraying

### Spraying intensity and p,p-DDE, without extreme value



**Fig. 5.** Concentration of p,p-DDE in ng/g b.p plotted against the amount of years the farmers have been spraying, with excluded extreme value

### Spraying intensity and pentachloroanisole

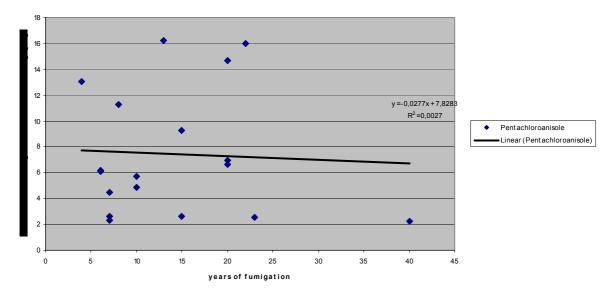
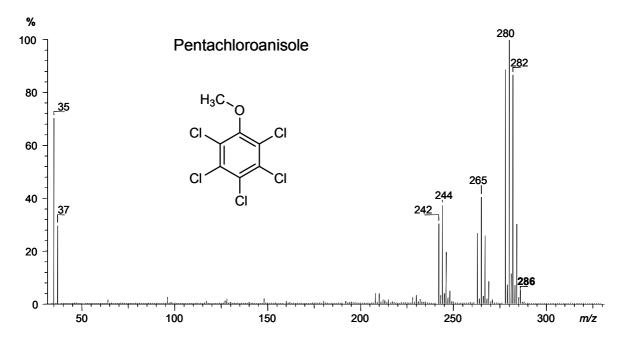


Fig 6. Concentration of Pentachloroanisole in ng/g b.p plotted against the amount of years the farmers have been spraying

# Appendix XIV

# GC-MS (ECNI) chromatogram of pentachloroanisole from the neutral fraction.



Provided by Maria Athanasiadou

### Appendix XV

### Excerpt from FAO's guidelines for working in a tropical country

In the FAO guidelines (FAO 1990) the minimum requirement for all types of pesticide operations is lightweight clothing covering most of the body. The clothing recommend in the text is defined as clothing which can be normally obtained and worn by the operator in his place of work. This may not be the additional items of protective equipment that he may be required to wear due to label recommendations FAO conclude that they should wear a long-sleeve upper garment, clothing covering the lower body including the legs, footwear (boots or shoes) and, if spraying high crops, a hat. While preparing the pesticides by pouring, mixing or loading, FAO recommend the use of protective gloves. It is also recommended to wear a simple face shield to protect eyes and face during mixing and loading. It is often impractical to wear boots or shoes when applying pesticides in water filled rice fields, but in this situation contamination from pesticides is low due to the high dilution by the water. Regular washing of the exposed feet and legs with clean water is recommended to further reduce possible contamination. FAO also recommend that the material used in the clothing should be as thick as possible, especially if cotton is worn. This is to avoid a higher exposure that actually can occur if the clothes get soaked in the pesticide.

From. Guidelines for personal protection when working with pesticides in tropical climates, FAO, Rome, 1990.

