LEVELS OF ORGANOCHLORINE PESTICIDE RESIDUES IN BREASTMILK AND COW MILK IN CENTRAL DIVISION OF MACHAKOS DISTRICT.

BY

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DECLARATION.

I, PATRICIA NTHENYA KIMANTHI, declare that this thesis is my original work and has not been presented for a degree in any other University.

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<u>DEDICATION</u>

To my parents, Angela & Fredrick Kimanthi and my children, Fredrick and Angela.

TABLE OF CONTENTS.

Chapter 3 MATERIALS AND METHODS

LIST OF TABLES.

LIST OF FIGURES AND MAPS.

List of Appendices.

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ABBREVIATIONS.

DDD(TDE) - 1,1' - (2,2 - dichloroethylidene) - bis (4 - chlorobenzene) (WHO, 1979).

DDE - 1,1' - (2,2 - dichloroethenylidene) - bis (4 - chlorobenzene) (WHO, 1979).

DDT - 1,1' - (2,2,2 - trichloroethylidene) - bis (4 - chlorobenzene) (WHO, 1979).

HCB - hexachlorobenzene.

PCB - polychlorinated biphenyls.

HEOD - dieldrin 3,4,5,6,9,9-hexachloro - 1a alpha, 2a alpha, 3 beta, 6 beta, 6a alpha, 7 beta, 7a alpha - octahydro - 2,7: 3,c - dimethanonaphth (2,3-b) - oxirene (WHO, 1989b).

PIPD - photodieldrin - 1,1,2,3a,7a - hexachloro-6,7-epoxy- 2,4,7-metheno-decahydro-3H- cyclopenta (a) pentalene (WHO, 1989b).

HCH - hexachlorocyclohexane.

α - alpha.

β - beta.

τ - gamma.

δ - delta.

E - epsilon.

ml - millilitres.

µl - microlitre.

Glc - gas liquid chromatography.

OC - organochlorine.

OCs or OC'S - organochlorine compounds.

ABSTRACT.

The levels of organochlorine pesticide residues were determined by gas chromatography in 147 human milk and 21 pooled cow milk samples, collected from two different agro-ecological zones of central division of Machakos, Kenya. The human milk samples were from mothers who had lived in the study area for over 5 years, were healthy and at the time breastfeeding first or second child. The milk samples were collected at 1 week to 4 months postpartum.

A total of 23 pesticide residues were detected in breastmilk, while 17 pesticide residues were detected in cow milk. The main organochlorine contaminants in human milk and cow milk were the DDT group, heptachlor/heptachlor epoxide and the HCH group at frequencies of 97%, 66%, 23% in breastmilk and 100%, 71%, 46% in cow milk, respectively.

There was a higher prevalence of the pesticide residues in the high potential production zone as compared to the low potential production zone. Significantly ($p < 0.05$) higher levels of β-HCH and τ-HCH in breastmilk and p,p'- DDD in cow milk were found in the low than in the high potential production zone.

In breastmilk, significantly higher levels of sum DDT and α-endosulfan were found in cases where pesticides were used in households as compared to cases where no use was reported. Levels of oxychlordane and α- endosulfan were lower in areas where pesticides were reportedly used on crops in the farms as compared to areas where no use was reported. Finally, lower levels of α-endosulfan were found in cases where cash crops were grown compared to areas where subsistence crops were grown while incidences were higher in the high potential production zone.

The mean levels of endrin and chlordane in human milk and chlordane in cow milk were above the prescribed codex alimentarius maximum residue limit (MRL) for milk and milk products.

The calculated daily intake by suckling infants in the study area of heptachlor epoxide, endrin, oxychlordane, endosulfan, chlordane and lindane from human milk exceeded the WHO/FAO acceptable daily intakes (ADI) by 70, 40, 9, 6.3, 4 and 1.6 times respectively. For cow milk on the other hand, calculated daily intakes by infants of heptachlor epoxide, oxychlordane, chlordane and endosulfan exceeded the ADI values by 30, 6, 5 and 1.5 respectively.

In summary, organochlorines had been used in the study area to the extend that consumption of human and cow milk by infants exposed them to risky levels of especially heptachlor epoxide, endrin, oxychlordane, endosulfan, chlordane, and lindane.

Monitoring programmes should be initiated to ensure that all avenues of exposure are documented and ways of reducing the environmental contamination by pesticide residues are developed and implemented.

CHAPTER I

INTRODUCTION.

1.0 GENERAL INTRODUCTION

A pesticide may be defined as any substance or mixture of substances intended for preventing, destroying or controlling any pest, including vectors of human or animal diseases, plant pathogens, unwanted species of plants or animals causing harm during or otherwise interfering with the production, processing, storage, transport or marketing of food, agricultural commodities and wood products or animal feedstuff or which may be administered to animals for control of insects arachnids or other pest in or on their bodies. The two important groups of pesticides which affect human health are the insecticides and herbicides (Jayaratnam, 1989).

Early attempts to control pests chemically used naturally occurring toxic substances such as mercury, sulphur; or plant extracts such as nicotine, pyrethrum or deris (rotenone insecticides). Synthesis of DDT (1,1' -(2,2,2-trichloroethylidene)-bis(4-chlorobenzene) by Zeidler in 1874, and later the discovery of its insecticidal properties by Müller in 1939, led to the development of organic (organochlorines) synthetic pesticides (WHO, 1979). These organic synthetic pesticides became very popular due to their broad spectrum of biological activity, stability and relatively low cost.

Man's endeavour to increase food production to match the rising human population stimulated widespread use of organochlorine pesticides after the second world war. This enthusiasm was changed by the discovery that these compounds are lipophilic and had the ability to persist in nature, hence a great threat to the environment.

In view of the potential health and environmental hazards, many countries banned the use of organochlorine pesticides such as DDT and limited the use of others (Jensen, 1983). However, organochlorine pesticides are still in use in many developing countries. Although OC pesticides have been used in both agricultural and public health programmes, studies to evaluate the impact of these compounds on human health and environment are scarce.

The chemical stability of organochlorine compounds coupled with their widespread usage in agriculture and in the home has led to the rising concentrations of these compounds in the air, water, soil and food. Due to their lipophilic nature, these chemicals accumulate in the body in the adipose tissue and in biological fluids such as blood and milk (WHO, 1979; Jensen, 1983).

Since the first report of the presence of DDT in human milk in 1951 (Laug et al., 1951) numerous investigations on organochlorine pesticides in human milk have been carried out, but only a few in developing countries, although in these countries, use of organochlorine pesticides has been most extensive. From these few studies, the levels of DDT reported are two orders of magnitude higher than those in industrialized countries of North America, East and West Europe (Jensen, 1983; Kanja, 1988).

The first study in Kenya, reported mean levels of sum DDT ranging from 1.69 mg/kg in milk fat from nomads (Loitokitok) to 18.73 mg/kg in milk fat from inhabitants of Rusinga island (Kanja, 1988). The latter level is among the highest found in any country after 1974, but is only about 20% of the sum DDT residue levels found in Guatemalan human milk between 1970-1974, which are also the highest levels of DDT in human milk so far reported (De Campos and Olszyna-Marzys, 1979). The relatively high levels of sum DDT reported for human milk in Kenya indicate widespread continued use of DDT as an insecticide in agriculture and vector control.

Human adipose fat and milk fat usually accumulate more than ten times higher levels of persistent organochlorine pesticides and polychlorinated biphenyls (PCBs) than milk fat from cows (WHO, 1979). This is due to the fact that humans are placed at the top of most food chains (Gochfield, 1972). The lower level of contamination of cow milk is due to continuing mobilization of fat and the contaminants therein, followed by excretion in milk (WHO, 1979). Apart from DDT, other organochlorine compounds have also been found in milk. These include dieldrin, aldrin, alpha and beta-Hexachlorocyclohexanes (HCH), lindane (τ-HCH), Hexachlorobenzene (HCB) and Polychlorinated biphenyls (PCBS) (Jensen, 1983).

There is very limited information on organochlorine compounds in milk from developing countries. In Kenya a study has been done by Kanja (1988)) on breastmilk, and one by Maitho (1978) on cow milk. The choice of milk for this study was based on the fact that, milk is the most important route of excretion of persistent organochlorine pesticides in human beings and other mammals (Yakushiji et. al; 1978 and 1979). Also, due to its high fat content, milk is a suitable indicator for monitoring exposure and levels of persistent organochlorine compounds.

1.1 STATEMENT OF THE PROBLEM.

Pesticide residues in human milk were first reported in 1951 in a study with normal healthy black American women which showed that their milk contained considerable amounts of the organochlorine insecticides DDT (Laug et.al, 1951). Since then, DDT and related organochlorine pesticides have been reported in human milk throughout the world (Jensen, 1983), with the levels being generally higher in developing countries as compared to the industrialized countries.

Although most developed countries have banned or restricted the use of organochlorine pesticides, the chemicals are still widely used in developing countries including Kenya (Weir and Schapiro, 1981). These compounds accumulate in the adipose tissue and milk and have known toxic effects in humans (Wassermann et. al, 1972). Studies in Kenya have demonstrated appreciable levels of these compounds in human milk, adipose tissue, eggs, fish, meat and other foods from various parts of the country (Wassermann, 1972; Kahunyo, 1983; Mugambi, 1986; Kanja, 1988).

The idea behind most human milk investigations has been to illustrate the infant's burden of these chemicals from breastfeeding. However, such studies have also been used as general monitoring tools for assessment of environmental pollution by organochlorines (Kanja, 1988).

Determination of the levels of organochlorine pesticide residues in milk from different areas with different agricultural practices (as in this study) would serve as a tool for generating data for a country's food and environmental pollution load (Slorach and Vaz, 1983).

1.2 Justification of the study

Organochlorine compounds are lipophilic in nature and once absorbed, accumulate in adipose tissue, milk and common foodstuffs. The levels in human tissues provides a tool for assessing concentrations in the human body, which reflects the extent of exposure. The intake of organochlorine compounds by the breastfed infant can also be estimated.

Foods especially those of animal origin contribute a lot to the build-up of organochlorine compounds in the body. The levels of organochlorine pesticide residues in milk fat are generally better correlated to levels in blood or serum lipids than to those in adipose tissue (Slorach and Vaz, 1983). Milk can be used to show the source of organochlorine pesticides in the body and also demonstrate the environmental pollution by these compounds.

The consequences of these pesticide residues to the health of the infant, whose main food source for a long period may be milk are for the time being unknown. Therefore, it is important to monitor the levels in milk to limit the possibility of exposure to high rates.

1.2.1 The rationale for assessing human exposure to persistent organochlorine pesticides through their levels in Breast and Cow milk.

Breastmilk and cow milk, due to their high fat content, provide a good tool for assessing the human exposure and environmental concentrations of organochlorine compounds. Continuous monitoring of these compounds in milk would provide data which would detect trends in levels of exposure.

Milk constitutes the major diet of infants, therefore, apart from monitoring levels of organochlorine compounds to measure human and environmental exposure, the intake of organochlorines from milk by infants can also be estimated.

1.3 Study Objectives.

The main objective of the present investigation was to establish the residual levels of organochlorine pesticides in samples of Breastmilk and Cow's milk from Central Division of Machakos District, Kenya.

The study was designed to achieve this through the following specific objectives.

(1) To determine the types of pesticides used in central division of Machakos district.

(2) To determine the levels of organochlorine pesticide residues in breastmilk.

(3) To determine the levels of organochlorine pesticide residues in cow milk.

1.4. Expected Benefits.

Information from this study would contribute towards a data bank on pesticides used and the residues in human and cow milk from Machakos District, Kenya. This data can be used to:-

(1) Show the extent of contamination of human and cow's milk with organochlorine pesticides and serve as baseline data for control of the levels of these compounds in milk from Machakos district.

(2) Influence the policy on pesticide use in the district and the country, and also the implementation of good agricultural and veterinary pesticide usage.

(3) Serve as added baseline data to set up other studies for biological monitoring of environmental contamination by organochlorine pesticides.

CHAPTER 2

LITERATURE REVIEW.

2.1 ORGANOCHLORINE PESTICIDES AND THEIR USE.

2.1.1 The DDT Group.

The term DDT is generally understood throughout the world and refers to p,p'- DDT (1,1'-(2,2,2-trichloroethylidene)-bis(4-chlorobenzene). The compound's structure permits several different isometric forms, such as o,p'-DDT. A typical example of technical DDT (commonly used as insecticide) had the following constituents: p,p'-DDT, 77.1%, o,p'-DDT, 14.9%, p,p'-TDE, 0.3%, o,p'-TDE, 0.1% p,p'-DDE, 4%, o,p'-DDE, 0.1%, and unidentified products, 3.5% (WHO, 1979; WHO, 1989a).

Most of the commercially available DDT insecticidal analogues have strikingly different properties. Especially remarkable is the slow metabolism and marked accumulation of DDT and its metabolite DDE. Notable, also is the rapid metabolism and negligible accumulation of methoxychlor (WHO, 1979; WHO, 1989a).

Technical DDT has been formulated in almost every conceivable form, including solutions in xylene or petroleum distillate, emulsifiable concentrates, water-wettable powders, granules, aerosols, smoke candles, charges for vaporizers and lotions. Aerosols and other household formulations are often combined with synergized pyrethroids (WHO, 1979).

A number of developed and developing countries have restricted or banned the use of DDT except when it is needed for the protection of health. However, DDT is still used extensively for both agriculture and vector control in some tropical countries. It is argued that, if DDT were not used, vast populations would again be condemned to the ravages of endemic or epidemic malaria. Substitution of Malathion or Propoxur for DDT would increase the cost of malaria control by approximately 3.4 or 8.5 fold, respectively (WHO, 1979). These increases could not be supported by some countries without decreasing the coverage of their control programmes.

2.1.2 Cyclodiene Group

This group of organochlorine pesticides consists of aldrin, dieldrin, endrin, heptachlor, heptachlor epoxide, chlordane, oxychlordane and trans-nonachlor.

Aldrin, dieldrin and endrin are persistent organochlorine insecticides with a higher acute toxicity than DDT. Dieldrin is an oxygenated metabolite of aldrin and is more persistent. These pesticides have been manufactured commercially since 1950, and have been used throughout the world up to the early 1970's. The compounds are used as insecticides in agriculture for the control of many soil pests and in the treatment of seeds. Insects controlled by these compounds include termites, grasshoppers, wood borers, beetles, and textile pests. Dieldrin has also been used in public health for the control of tsetse flies and other vectors of debilitating tropical diseases. Both aldrin and dieldrin act as contact and stomach poison for insects (Jensen, 1983).

Since the early 1970's, organochlorine compounds have been severally restricted or banned in a number of countries especially their use in agriculture due to their persistent character in the environment (WHO, 1989b). However, the use for termite control continues in other countries and they are still in use in most of the third world countries. When the intake of aldrin/dieldrin ceases or decreases, the body burden decreases. The biological half-life in man is approximately 9-12 months. Heptachlor, heptachlor epoxide, chlordane, oxychlordane and trans-nonachlor are closely related organochlorinated insecticides. Heptachlor epoxide and oxychlordane are very persistent epoxy metabolites of heptachlor and chlordane, respectively. Heptachlor epoxide and transnonachlor both occur as impurities in technical grade chlordane.

Heptachlor has been used for more than 30 years as a stomach and contact insecticide, mainly in the control of termites and soil insects. Heptachlor epoxide, the most persistent metabolite, is rapidly formed and can be found in the body, mainly in adipose tissue. Excretion takes place via urine, faeces and milk (WHO, 1984b). In its country of origin, the USA, its use is now restricted to underground termite control. In several other countries, approved uses have been gradually withdrawn. The same applies to chlordane which has been used for more than 35 years as a broad spectrum contact insecticide, mainly on non agricultural crops and on animals (WHO, 1984a).

2.1.3 Hexachlorocyclohexanes (HCH)

The commercial insecticide HCH is a mixture of different isomers each denoted by a Greek letter Viz:- α-HCH (53-70%), β-HCH(3-14%), τ-HCH(11-18%). The τ-HCH isomer, also known as lindane, is the most acute toxic isomer and is an important insecticide, often used as a substitute for DDT. It is more volatile and significantly more soluble in water than DDT (Jensen, 1983).

2.1.4 Hexachlorobenzene (HCB)

Hexachlorobenzene is one of the most persistent organochlorines known. In some countries HCB has been used as a fungicide to stain seeds. It is used as a raw material in the industrial production of pentachlorophenol. Pentachlorophenol in itself is the main metabolite of HCB in mammals. HCB is formed as a by product in many industrial processes involving chlorine and organic chemicals, as it may be formed during disinfection (chlorination) of waste water (Jensen, 1983).

2.1.5 Endosulfan.

Technical endosulfan consists of alpha and beta isomers in the ratio of approximately 70:30. It is used as a broad spectrum contact and stomach insecticide mainly on fruits, vegetables, tea and non-food crops like tobacco and cotton. It is also used for tsetse control, a wood preservative and for the control of home garden pests. In some countries it is used in horticulture to control insects and mites (WHO, 1984c). The main source of exposure of the general population is food, but residues have generally been found to be well below the FAO/WHO maximum residue limits. Smoking may be an additional source of exposure, due to the use of endosulfan on tobacco (WHO, 1984c).

2.2. "Use of Organochlorine Pesticides in Kenya."

Kenya imports pesticides from all over the world, so long as the types and quality have been tested for suitability under local conditions and are acceptable for registration in the country. A number of companies and individuals are engaged in the pesticide trade. These range from large multinationals to cooperative unions and societies, as well as individuals. In the present legislation there are no restrictions as to who may engage in the importation of pesticides.

Pesticide importation in Kenya amounted to about KSh. 600-700 Million or approximately 10,000 tons in 1989 (Table 2.1). The value of importation over four years is shown in Table 2.1.

Table 2.1 Value of Imports of pesticides into Kenya (Millions Kshs.) 1986-89

Year	Insecticides & Acaricides	Herbicides	Fungicides	Others	Total
1986	134.9	121.3	281.3	42.6	580.1
1987	182.3	173.4	357.3	28.1	741.1
1988	158.9	145.2	329.9	28.5	662.5
1989	208.1	154.2	328.2	30.7	721.2

Source: Pests Control Products Board.

Most farmers in Kenya buy their pesticides from either the distributors, stockists, cooperative societies and/or unions, or local outlets like kiosks. The cooperative societies or unions are more favourable to small scale farmers (especially when one is a member), since credit facilities can be availed to members on condition that their produce acts as security and is sufficient to cover the loan required.

When buying pesticides, many farmers are confused by the many pesticide products available in the market, with different promotional advice and literature presented by various manufacturers. Since all manufacturers claim their product to be the best, the farmers choice in many cases is based on the cost, what other farmers are using, information from agricultural extension agents, size and nature of packaging of the products for ease of transportation, or whether the containers can be re-used, and availability in local outlets like kiosks.

2.2.1. Legislation of pesticide use in Kenya.

There is a distinct machinery in Kenya which monitors the legislation of pesticides to be used in the country. Several bodies are involved in the machinery and deliberate on the safe use of pesticides, pesticides to be used in the country and whether a chemical banned from its country of origin should continue to be used in Kenya. Some of the bodies regulating the use of pesticides in Kenya are:-

1. Kenya Environmental Secretariat which coordinates all matters pertaining to the protection of our environment and provides linkage to international bodies like FAO, WHO, UNEP.
2. Pesticides Chemicals Association of Kenya (PCAK) which

incorporates most of the agrochemical manufacturers in Kenya. It ensures that members abide by the ethical standards of pesticide manufacture such as safety, packaging, labelling and use of products.

3. Pest Control Product Board of Kenya (established 1984) regulates the use of pesticides in the country at all stages. This includes the importation, transportation, labelling, formulation, storage, usage and disposal. It also investigates the role of pesticides and their impact on the environment and collaborates with international organizations (e.g. FAO/WHO, UNEP etc). It also acts as a source of technical information on pesticides use, management, hazards and advises the government on status of approved pesticides.
4. Kenya Bureau of Standards(KBS) develops standards (in collaboration with the relevant industry and other national bodies) and monitors the quality of all pesticides manufactured and/or imported into the country for sale. The KBS has developed a code of practice for safe handling, storage and use of pesticides. It has also developed standards on household insecticides and is currently working on a guide to the maximum limits of pesticide residues in food. The KBS is a member of the above mentioned bodies regulating the use of pesticides in Kenya.

As a policy, all pesticides are first tested by the Pesticides Control Board to determine their rates, active ingredients and subsequent side effects and the results communicated to the Ministry of Agriculture before they are released for use to the farmers. From time to time the board draws up a list of pesticides which have been banned or restricted in the country (Table 2.2).

Table 2.2 Pesticides banned or restricted in Kenya.

Chemical Name	Use	Action
1. Dibromochloropropane	Soil fumigant	Banned
2. Ethylene Bromide	Soil Fumigant	Banned
3. 2,4,5, - T	Herbicide	Banned
4. Chlorodimeform	Insecticide	Banned
5. Mixture of isomers of Hexachlorocyclohexane (HCH)	Insecticide	Banned
6. Lindane (Pure gamma-BHC (HCH)	Insecticide	Restricted for use on seed dressing only
7. Chlordane	Insecticide	Banned
8. Heptachlor	Insecticide	Banned
9. Endrin	Insecticide	Banned
10. Aldrin	Insecticide	Restricted for termite control in building industry
11. Dieldrin	Insecticide	"
12. Toxaphene	Insecticide	Banned
13. DDT	Insecticide	Restricted use to public health only for Mosquito breeding grounds banned, for agricul tural use
14. Caftafol	Fungicide	Banned

Source: Pest Control Products Board, Kenya, 1987

2.2.2 Prevalence of Organochlorine Residues in Kenya.

Organochlorine pesticides have been used widely in agriculture and Public health in Kenya for over four decades. The impact for this widespread usage on the environment and its effects on organisms therein has not been thoroughly investigated (Kanja, 1988). The few studies done indicate that detectable amounts of organochlorine pesticide residues can be found in various body tissues including milk and body fat. They have also, been detected in many food commodities and the environment.

Kanja (1988) collected food samples in parallel to human milk samples from six different areas in Kenya and investigated for organochlorine pesticides residues. The main organochlorine compounds found in all milk samples were p, p' - DDT and p, p' - DDE. Fifty percent (50%) of all the food samples had at least one organochlorine pesticide residue. Great regional differences were found and mean levels of sum DDT and DDT/DDE ratio ranged from 1.1 to 18.7 mg/kg milkfat and from 0.7 to 5.7 respectively. Mugambi (1986) studied the organochlorine pesticide contamination of chicken eggs and chicken feed samples collected from Embu and Meru districts. All the eggs contained p,p' - DDE. Kahunyo (1983) detected 12 different organochlorine pesticide residues in a similar study of chicken eggs and chicken fat samples collected from 11 districts in Kenya. The DDT group showed both the highest incidence and the highest levels. Maitho (1978) found low levels of the DDT group, lindane, aldrin and dieldrin in cow milk and body fat. Lincer et al. (1981) found that African Cormorants (Phalacrocorax africanus) collected in 1970, contained approximately eight to fifteen times as much DDE as the single white-necked cormorant (Phalacrocorax carbo) from the same lake. The DDE level of a white pelican liver in 1970, was only half that found in the same species in 1981, which indicate a possible increase of organochlorine contamination of the lake system.

Koeman et al (1972) studied the contamination of Rift Valley lakes and found extremely low levels of DDT, DDE and dieldrin in the tissues of birds and fish collected in lake Nakuru. Other very low levels of DDE were detected in species in lakes Naivasha, Baringo and Elementaita. Greichus et al. (1978) also studied the contamination of Lake Nakuru by organochlorine compounds and found low values of DDE, DDT and dieldrin, though the values were slightly higher in 1971 (Koeman et al, 1972). Allsop (1978) investigated the effect of dieldrin, sprayed by aerial application for tsetse fly control on game animals. He reported that all tissues collected from the animals before and after spraying had low levels of HEOD (the active ingredient of dieldrin) and its photo-isomer (PIPD).

Wasserman et al. (1972) investigated the storage of organochlorine insecticides in the adipose tissue of people from Kenya and found DDT to be the main contaminant, while β-HCH, dieldrin and heptachlor epoxide were also detected. There was a positive age association with DDT - derived material stored in the adipose tissue of the age groups investigated.

The levels of organochlorine compounds reported in food and human samples by developed countries are generally low (Jensen, 1983). However, there are not many extensive studies of this kind done on food and human samples in developing countries where these compounds are still in use. It is important that more studies should be encouraged in Kenya.

The high levels of DDT group of residues in human milk from Kenya could be correlated to the diet of the mothers (Kanja, 1988). As discussed earlier on, a number of studies in Kenya on some dietary components like eggs (Kahunyo, 1983; Mugambi, 1986) and cow milk (Maitho, 1978) have shown relatively high levels of DDT. These results probably reflect on contribution of the diet to the relatively high levels of DDT detected in human milk in Kenya (Kanja 1988).

2.2.3 Possible Toxicity of Pesticides to Humans.

Available epidemilogical data are insufficient to conclusively show the potential carcinogenicity, mutagenicity and teratogenicity of the various organochlorine pesticides for man (WHO, 1984a; WHO, 1984b; WHO, 1984c; WHO, 1979; WHO, 1989b; WHO, 1992a and WHO, 1992b). However, there are suggestions that OCs play some role in enzyme induction and affect the nutritional status of exposed workers.

The toxic action of a number of compounds is clearly the result of their inhibition of one or more enzymes. So far, only a few enzymes or enzyme systems are known to be induced by chemicals. The outstanding example is the induction of microsomal mixed function enzymes of the liver. Microsomal enzymes are associated with oxidation and most of the changes they produce render oil-soluble compounds more water soluble, hence more easily excreted (WHO, 1979).

DDT and other organochlorine compounds have been shown in vitro and in vivo studies to inhibit some enzymes of intermediary metabolism and other miscellaneous enzymes. So far evidence is lacking to show this inhibition is sufficient to have influence on the function of an intact organism (WHO, 1979; WHO, 1984a; WHO, 1984b; WHO, 1984c; WHO, 1989b; WHO, 1992a and WHO, 1992b). When insects are exposed to organochlorine pesticides, microsomal oxidase enzymes may be induced as an adoptive response. The induction of the microsomal enzymes is valuable to the insect (or organisms), in that it is protected against the insecticide which it encounters in sub-lethal concentrations. This response is different from the one which leads to resistant strains. The latter is an inheritance of the ability to avoid the harmful effects of the insecticide, possibly by degrading it and passing this capacity on to their offspring (Kanja, 1988).

Animal studies indicate that nutritional status influences the toxicity of DDT (WHO, 1979). The preferential storage of DDT and other organochlorines in the fat tissues can mitigate the effect of acute poisoning. If rats that have stored large amounts of DDT are starved, they may suffer toxic effects due to mobilization of the fat and the bound DDT.

In man, nutritional status will have similar effect to that found in experimental animals. However, the possibility that starvation in man could precipitate toxic manifestations is regarded as unlikely as the stored levels do not approach those found in laboratory animals and the lower metabolic rate of man would in any case, result in slower mobilization (WHO, 1979). In fact, severe weight loss sometimes does not cause any increase in storage of DDT in connection with certain wasting diseases. However, people with full-time occupational exposure to DDT average ten times more stored

DDT than the highest values reported in connection with disease, but do not exhibit predisposition to the diseases in question (WHO, 1979).

Hussein and El-Tohamy (1988) showed that younger Egyptian pesticide sprayers/workers with a mean age of 14 years had poorer overall nutritional status with significantly lower mean haemoglobin, plasma phospholipids, and retinal binding protein concentrations compared with the mean values obtained for sprayers with a mean age of 33 years.

2.3 "Possible Hazards from use Organochlorine pesticides".

Pesticides are used in an effort to increase crop production and reduce waste/loss of food during storage. They are also used to control vector-borne diseases with an objective of attaining self-sufficiency in food. Some of the compounds used in pest control, are the organo-synthetic pesticides which date back to 1939, when the insecticidal properties of DDT were discovered by Müller (WHO, 1979). These compounds are now generally referred to as organochlorines and include DDT, aldrin, dieldrin, and hexachlorocyclohexanes (HCH). The organochlorines present a great threat to the environment because they are lipophilic and have the ability to persist in the environment. The pesticides are a health hazard to man because they can accumulate mainly in fatty tissues of man (Wassermann et. al., 1972).

There are many dangers posed by the use of pesticides. The prolonged use of organochlorine pesticides has led to the creation of resistant species, elimination of major species and sprouting of minor species of pests due to elimination of natural predators.

In Kenya, mass spraying over wide areas has been done in the fight against mosquitoes, tsetse flies and other agricultural pests of importance in the country. These chemicals drift and build up in the atmosphere and may be hazardous to beneficial insects such as bees, wild animals which feed on the crops, and also other smaller organisms. Other residues are washed by rain into rivers and lakes where they may affect fish and other aquatic life. Furthermore, when these residues are picked up by the lower food chain groups (plants and animals) they progressively accumulate in the food chain and eventually end up in the human body (Adamovic et. al., 1978; and Noren (1983a), 1983). As a result of these findings, use of many organochlorine compounds has been banned in developed countries and most countries have placed restrictions on their major uses (Skaare, 1981; Kanja, 1988). However, organochlorine pesticides are still in use in developing countries including Kenya (Weir and Schapiro, 1981).

The problem of pesticide misuse is further aggravated by illiteracy, lack of proper knowledge leading to indiscriminate use and the fact that pesticides which have been banned in other countries are still available to Kenyan farmers. Improper use, coupled with the lack of protective clothing during spraying, leads to an increased exposure and high ingestion rate. Furthermore, there is a general shortage of personnel and funds to test pesticides locally to ensure proper control. Many a time, disease, signs, symptoms and poisoning caused by pesticides are not diagnosed, and if they are, the proper therapy or prophylaxis may not be instituted (Kanja, 1988).

Reporting machinery of pesticide poisoning is absent, though there have been newspaper reports of poisoning and deaths. Some deaths and cases of improper use and or application of pesticides that have been reported (Kanja, 1988; Weir and Schapiro, 1981) are:

- A woman put a packet of powder pesticide on her food shelf. She mistakenly mixed up the content with maize meal while preparing "ugali" for her family. She and her children died.
- A family ate food previously stored in pesticide drums. They all fell ill.
- Spraying of pesticides while workers are working in the field.
- Mixing pesticides with unprotected hands.

- Some people think that pesticides are medicine and try to use them to treat skin diseases.
- Unscrupulous fishermen use the chemicals to kill fish, which are sold to unsuspecting consumers.
- In the field, plantation workers have been known to strip down to their shorts, leaving their arms, legs and chests bare while spraying pesticides.
- A worker was overhead boasting to his friend that he could identify all pesticides in the store by taste.

Acute toxicity occurs when organochlorine pesticides are ingested directly. This occurs in suicidal cases, or when the pesticides are accidentally mistaken for similar looking food products. A large number of such cases reported are often in the local newspapers, but unfortunately no official figures are available. According to the Waiyaki Report (1988) there were at least two cases of pesticide poisoning daily at Kenyatta National Hospital in Nairobi, Kenya.

Chronic toxicity on the other hand, is as a result of bioaccumulation of these compounds in the body from repeated ingestion of sublethal doses. This is of particular high risk to foetuses, infants and young children due to the vulnerability and immaturity of their developing physiological system. DDT has been correlated with abortions in humans and dairy cattle, although no significant relationship has been established (Kanja, 1988). The type and effects these chemicals have on the foetus depend on the concentration and duration of exposure, and they may be modified by other factors, such as dietary deficiency, age, viral infection (WHO, 1979; WHO, 1984a and WHO, 1984b).

2.3.1 "Organochlorine pesticide residues in milk".

There are great differences in human milk contamination with organochlorine pesticides between countries and regions. Levels of persistent pesticides, e.g. DDT, are highest in many Third World countries where these chemicals are still in use, while levels of PCBs and heavy metals are highest in the heavily populated areas of many industrialized countries. Within a country, levels are lower in mothers living in rural areas than in mothers living in urban areas. On the other hand, levels in agricultural rural areas may be high in cases where these persistent pesticides are in regular use (Jensen, 1983).

Wassermann et al. (1972) observed that in S. Africa, Thailand, Brazil and Nigeria after the age of 45 years, there is a decrease in the storage of organochlorines in human adipose tissue, while in Israel and Kenya, there is continued increase after the age of 45 years. Therefore, it has been suggested that the age group of 25-44 years can be considered a characteristic age for defining the organochlorine storage level in a community (Wassermann et al., 1972 & 1974).

In Kenya, the presence of organochlorine compounds in human milk (Kanja, 1988) and cow milk (Maitho, 1978) has been demonstrated. To the authors' knowledge, no further study has been done using human milk or cow milk to find out if organochlorine residue levels in the Kenyan population has increased due to the continued use of pesticides.

Excretion of organochlorine substances in milk is important because of the transfer of these contaminants to the breastfeeding children. The same are transferred from cows' milk and other foods to humans. Milk contains 3-5% fat and is a major excretory route of organochlorine compounds. Women exposed to organochlorines excrete more of these compounds than those without any known exposure (Knowles, 1974). There was for example slightly greater secretion of DDT and much greater secretion of various isomers of HCH by urban mothers (compared to rural mothers) in Japan which was attributed to their greater intake of cow milk (WHO, 1979).

It is important when comparing results from different studies to note that several factors may influence the level of organochlorines in human milk and cow milk. The organochlorines are present in the fat of milk and the fat content can vary widely. For example, it is higher at the end of each feed ("hind milk") than that at the beginning ("foremilk"), and higher during the middle of day than early in the morning. Thus, if the organochlorine levels are expressed on a whole milk basis, wide variations can be found between different feeding occasions and different times post parturition. However, the organochlorine levels expressed on a fat weight basis show much less variation and thus comparisons should be made on this basis (Slorach and Vaz, 1983). Other factors which can affect the levels of organochlorines in breastmilk and cow milk include:

1. Food intake - A large intake of calories results in higher sum DDT content in human milk (Jensen, 1983). Fish consumption may be a major source of this sum DDT intake (Jensen, 1983).
2. Smoking - Smokers (especially cigarette smokers) have a higher sum DDT level in the milk than do non smokers, probably due to the use of DDT in tobacco fields (Jensen, 1983, Slorach & Vaz, 1983).
3. Season - Several studies have shown that there are great seasonal variations in the sum DDT levels in human milk. This might be due to the different levels of DDT usage between seasons and countries (Jensen, 1983).
4. Number of Children - Most investigations have shown

that the content of sum DDT in human milk is highest during the nursing of the first child (Slorach & Vaz, 1983). Mothers giving birth to twins excrete relatively more sum DDT in the milk due to a higher milk production and increased depletion of fat deposits (Adamovic et al., 1978).

5. Maternal body weight - Slim persons generally have a higher concentration of sum DDT in the adipose fat tissues (Polishuk et al., 1977; Jensen, 1983) and a slimming treatment increases the DDT level in the blood, both in the mother and in the infant. The higher concentrations in slim persons might be explained by their smaller fat deposits and therefore, lower dilution of residues (Jensen, 1983).

6. Length of Lactation - During the first three months of lactation, the sum DDT concentration in milk fat decreased around 30% and sum DDT in whole milk decreased 45% (Jensen, 1983). In Japan p.p' - DDT content of human milk fat showed a gradual decrease from four to one ppm during ten months (Yakushiji et al., 1978b). Milk fat concentrations of sum DDT are also higher in 'hind milk' than in 'fore milk' (Slorach & Vaz, 1983).

7. Maternal age - No clear consensus has been reached regarding the factor of maternal age. Some studies

indicate that younger women excrete more sum DDT in their milk, though they have lower serum DDT concentrations; while other studies indicate lower sum DDT levels in milk from young women (Jensen, 1983; Slorach and Vaz, 1983). These divergences might be explained by the existence of confounding factors, such as number of previous childbirths.

8. <u>Maternal Residence</u> - In general, mothers from urban areas have higher sum DDT levels in the milk than mothers from rural areas, as long as DDT has not been commercially used in the area. In some areas with DDT spraying (agricultural or malaria prone areas) extremely high levels of DDT and DDE in the milk have been demonstrated, especially in central American cotton growing areas where DDT is aerially sprayed (De Campos and Olszyna-Marzys, 1979). DDT levels in human milk from developing countries are usually very high. This is due to the continued use of DDT for agricultural and vector control purposes.

9. <u>Home use of Pesticides</u> - A high use of home and garden organochlorine pesticides has in some cases been related to increased levels of sum DDT in breast milk (Jensen, 1983).

10. <u>Different Cultures</u> - In the United States and Brazil, black population groups had higher sum DDT levels in their milk than did white people (Matuo et

al., 1980; Jensen, 1983). These differences were attributable to social class, which had been found relevant in evaluating DDT body burden (Jensen, 1983). In other countries higher sum DDT levels were found in immigrants as compared to that of the indigenous people (Jensen, 1983).

11. Occupation - The very high levels of DDT found in human milk from agricultural workers and pesticides formulators (handlers or workers) has been partly due to occupational exposure (WHO, 1979).

12. Trends in levels and effect of regulations - Many investigations have shown that enforcement of DDT regulations had, after some years caused significant and steady decrease of sum DDT levels in human milk. DDT and TDE decreased faster than did the more persistent DDE (Jensen, 1983).

2.3.1.1 Levels of Organochlorine Pesticide residues in Human and Cow' Milk.

Most human milk monitoring studies have concentrated on analyzing mainly the organochlorines DDT, DDE, PCBs, HCH, HCB, dieldrin and heptachlor epoxide (Jensen, 1983). The population studied has been a typically representative group of mainly 50 to 100 health individuals from each area excluding immigrants. Persons with known occupational exposure, or mothers with sick children were excluded. Often

100 to 200 ml of milk was collected from each donor, but sometimes only a tenth of this amount was available, making the analytical work more difficult and the results less certain. The daily variations in fat content in an individual's milk made the timing of milk sampling very important, a consideration which was not always been taken into account (Jensen, 1983; Slorach and Vaz, 1983).

The investigations have mostly used samples from single donors to determine individual variations in levels and combined with a questionnaire, to seek possible sources of exposure or other risk factors (Jensen, 1983). Most contaminants found in human milk are fat soluble compounds which have been detected mainly in the fatty phase of milk. DDT and its main metabolite DDE have been detected in almost every human milk sample analyzed, (WHO, 1979; Kanja, 1988), while PCB have been mainly detected in human milk from industrialized countries (Slorach and Vaz, 1983).

Since 1951, when Laug and co-workers reported that the breast milk from normal and healthy black American women contained considerable amounts of organochlorine insecticide DDT, many studies have been done on organochlorine pesticide residues in breast milk throughout the world.

It has been known for a long time that human milk may contain a higher concentration of DDT than cow milk in the same country (WHO, 1979). The concentrations of DDT and DDE in human milk reported from different countries are listed in Table 2.3. Especially high values have been reported from Kenya, where the highest mean sum DDT residue level was 18.73 mg/kg (Kanja, 1988). These were among the highest mean levels found in any country after 1974, though they are only about 20% of the sum DDT residue levels found in Guatemalan human milk in 1970-71. The Guatemalan levels are the highest residue levels of DDT in human milk ever reported (De Campos and Olzyna-Marzys, 1979). The main cause of the very high accumulation of DDT in Guatemalan human milk was indoor spraying of DDT for Malaria eradication, while in Kenya it was due to the continuing use of DDT as an insecticide in agriculture and vector control.

As already mentioned earlier, DDT was the first environmental chemical detected in human milk (Laug et al., 1951). Since then DDE and other DDT isomers have been detected in most human milk and cow milk investigations. A few investigators have also determined p.p' - DDD and the o.p' - isomers. Animal experiments indicate that o,p'-DDT is excreted in the milk to a lesser extent than p.p' - DDT (Jensen, 1983).

Table 2.3 MEAN/MEDIAN LEVELS OF the DDT - GROUP RESIDUES (mg/kg fat) IN HUMAN MILK FROM DIFFERENT COUNTRIES.

COUNTRY	YEAR	NO.OF SAMPLE%+VE	% FAT	p,p'-DDT	p,p'-DDE	SUM DDT	DDT/DDE RATI-O	REFERENCE
Kenya	1983-1985	264	4.1	3.73	2.95	6.99	1.26	Kanja, 1988
Nigeria	-	44	2.79	2.37	1.33	3.83	0.56	Atuma et al. 1987
Rwanda	-	75	3.13	1.68	2.36	4.16	0.71	Warnez et al., 1983
India	-	-	-	1.2	4.8	6.55	0.25	Slorach & Vaz, 1983
China	-	-	-	1.8	4.4	16.7	0.41	Slorach & Vaz, 1983
Iran	1974-1976	131	-	1.0	1.1	2.9	0.91	Jensen, 1983
Israel	1975	-	1.5	0.97	1.81	5.77	0.53	Jensen, 1983
Turkey	-	163	3.2	0.65	4.61	5.84	0.14	Kanja, 1988
Spain	1979	45	-	3.5	6.3	9.8	0.56	Jensen, 1983
Italy	1975	30	2.6	096	1.38	2.69	0.69	Jensen, 1983
Poland	1979	40	-	1.2	8.7	9.98	0.14	Jensen, 1983
Denmark	-	57	-	011	1.04	-	0.11	Kanja, 1988
Belgium	-	20	2.64	0.35	1.59	1.94	0.22	Kanja, 1988
Hungary	1975 - 1976	21	4.2	0.52	2.97	3.55	0.18	Jensen, 1988
Sweden	1978 1979	23 41	- -	0.24 0.21	1.3 1.3	1.6 1.5	0.18 0.16	Hofvander et al., 1981 & Kanja, 1988
U.K.	1979-1980	102	2.7	0.11	1.6	-	0.07	Kanja, 1988
U.S.A.[a] [b]	1973-1975	34 6	- -	4.25 0.4	14.7 1.9	19.2.4	0.29 0.21	Jensen, 1983
Federal Republic of Germany	1979	374 (100%	-	-	-	1.76/1.26	-	Jensen, 1983

Key.
a - pesticide area.
b - non pesticide area.
%+ve - percent positive.

In most recent human milk studies made in industrialized countries, the levels of DDE in the milk have far exceeded the levels of DDT (Jensen, 1983). These findings led to the banning of any kind of direct exposures in those countries and hence, the most important remaining exposures are now through food contamination (Jensen, 1983).

2.3.1.2 Other Organochlorine pesticides in human milk.

Other Organochlorines that have been detected in human milk are dieldrin, aldrin, heptachlor, heptachlor epoxide hexachlorobenzene (HCB) and hexachlorocyclohexanes (HCH) (Jensen, 1983). The levels of aldrin and dieldrin detected from different studies are given in Table 2.4. Heptachlor, Heptachlor epoxide, chlordane, oxychlordane and trans-nonachlor have not been extensively used as compared to DDT, but their residues have been detected in human milk (Tables 2.5 and 2.6), adipose tissues and food samples (Jensen, 1983).

TABLE 2.4 MEAN LEVELS OF DIELDRIN AND ALDRIN (mg/kg) IN HUMAN MILKFAT FROM DIFFERENT COUNTRIES.

COUNTRY	NO. OF SAMPLES	DIELDRIN	ALDRIN	REFERENCE
IRAQ(BAGHDAD)	50	0.03_a	0.017_a	Al-Omar et al. 1985
IRAN	131	0.33		H.Tonkabony et al.;1976
DENMARK,1982 (COPENHAGEN)	4_b	0.04		Jensen,1983
SPAIN,1979				
RURAL	21	0.031		
URBAN	24	0.01	0.004	Jensen,1983
SWEDEN 1978-9	23	0.016		Hofvander et al.;1981
GUATEMALA 1971				
RURAL AREA	27	2_a		Jensen,1983
ISRAEL,1975	29_c	0.58		Polishuk et al.; 1977
AUSTRALIA				
RURAL AREA	20	0.81		Jensen,1983
GERMANY (1975 & 1979	30/374	0.06	0.04	Jensen,1983
INDIA,1979(LUCKNOW	25		29.8_a	Siddiqui et al.; 1981
ITALY, 1975 (MILAN)	30		0.04	Jensen,1983
KENYA	264	0.37	0.05	Kanja, 1988

Key.
a - whole milk. b - pooled samples. c - colostrum.

TABLE 2.5 MEAN LEVELS OF CHLORDANE AND OXYCHLORDANE IN HUMAN MILKFAT mg/kg) FROM DIFFERENT COUNTRIES.

COUNTRY	NO. OF SAMPLES %POSITIVE	OXYCHLORDANE & CHLORDANE	REFERENCES
SPAIN, 1979	45	0.026c	WHO, 1984a
MEXICO	620	0.4_m	Jensen, 1983
HAWAII, 1979-80	50 (100%)	0.059	Jensen, 1983
MISSISSIPPI, 1973-5			
Non-pesticide area	6 (68%)	0.05	WHO, 1984a
Pesticide area	34 (100%)	0.13	WHO, 1984a
KENYA			
MERU, 1985	44 (2%)	0.004	Kanja, 1988

Key.
c - chlordane
m - median

Several metabolites of chlordane have been identified and species differences have been found. Oxychlordane is the most relevant animal metabolite, being more persistent and toxic than the parent compound (WHO, 1984a).

TABLE 2.6 MEAN LEVEL OF HEPTACHLOR AND HEPTACHLOR EPOXIDE (mg/kg) FROM HUMAN MILKFAT FROM DIFFERENT COUNTRIES.

COUNTRY	NO OF SAMPLE	HEPTACHLOR/HEPTACHLOR EPOXIDE	REFERENCE
SPAIN 1979	45	2.51_E/0.035	JENSEN, 1983
RURAL AREA	21	2.56_C/0.017	JENSEN, 1983
GERMANY 1979	374	0.014/1.008	JENSEN, 1983
GUATEMALA 1971 RURAL AREA	46	0.004_A	de Campos et al.; 1979
ISRAEL (1975)	29_C	0.72	WHO, 1984b
SWITZERLAND	50	0.07	JENSEN, 1983
JAPAN 1979	33	0.005A	JENSEN, 1983
CANADA 1967-8	147	0.13	JENSEN, 1983
HAWAII 1979-80	50	0.35	WHO, 1984b
U.S.A 1975	1436	0.09	JENSEN,1983
AUSTRIA 1977-8 VIENNA	20/182	0.010/0.013	WHO, 1984b
MEXICO 1975	0.01_M		JENSEN, 1983
1976	0.01_M		JENSEN, 1983
KENYA 1979	33	0.5_M	FAO/WHO, 1981
KENYA MERU 1985	44(5%)	0.041_E	KANJA, 1988
KARATINA 1983	50(16%)	0.015_E	KANJA, 1988
TURKANA	30(3%)	0.007	KANJA, 1988

Key.
A - whole milk
m - median.
E - heptachlor
C - colostrum

Table 2.7 gives the levels of HCH isomers in milkfat from different countries. The ratio between different HCH isomer residues changes from the beginning of food chains until deposition in human milk fat, resulting in relatively higher levels of the more persistent β-isomer in the milk (Jensen, 1983) The α-, τ- and δ- isomers might also isomerize into the β-isomer in living organisms (Jensen, 1983).

TABLE 2.7 MEAN HCH - ISOMERS (mg/kg) IN HUMAN MILKFAT FROM DIFFERENT COUNTRIES.

COUNTRY	NO. OF SAMPLE	α-HCH	β-HCH	γ-HCH	δ-HCH	SUM	REFERENCE
SWEDEN UPPSALA 1978-9	23	0.006	0.084	0.003		3.2^{m}	Hofvander et al., 1981
FEDERAL REPUBLIC OF GERMANY 1979-84	2709	0.024	0.37				WHO, 1992a
CZECHO-SLOVAKIA		0.15	0.5	0.35	0.1		JENSEN, 1983
RWANDA RURAL AREA		0.04		0.22			WARNEZ et al.; 1983
URBAN AREA				0.07			"
NIGERIA BENIN	35	0.04	0.47	0.05			ATUMA et.al. ;1986
BENDEL	44	0.015	0.61	0.087			ATUMA et.al. ;1987
SPAIN RURAL	21		0.067	0.011		0.07	JENSEN, 1983
URBAN	24		0.073	0.037		0.11	"
CANADA	210	0.007	0.214				WHO, 1992a
NETHER LANDS	36	0.01	0.28_{m}				WHO, 1992a
BELGIUM RURAL	34	0.05	0.57	0.06			JENSEN, 1983
URBAN	20	0.02	0.25	0.08			"
KENYA	264		0.09	0.04			KANJA, 1988

Key.
m - median

Therefore, except in areas where lindane has been directly applied, β-HCH will normally account for 90% of the HCH residues.

The persistent molecule hexacholorobenzene is fat soluble and it bioaccumulates in food chains. It has been detected in most surveys of human tissues and in milk (Jensen, 1983).

On the other hand, several studies on dairy milk have shown that organochlorine pesticide residues are excreted in cow milk. Maitho (1978) detected five different types of pesticides (organochlorine) residues in 48% of the samples. He reported that the DDT group was the most frequent organochlorine residue in animal products (cow's) in Kenya. Table 2.8 gives a sample of some of these studies and the levels of the various organochlorine pesticide residues in cow milk.

2.3.2 Sources of organochlorine pesticide residues in Milk

Food (diet) represents the major source of intake of organochlorine pesticide residues in the general population and animals. These compounds reach man through the food chain and accumulate in fatty tissues and other organs. Studies in several countries have shown that the levels of organochlorine pesticide residues in milk are related to dietary habits (Noren (1983c), 1983; Kanja, 1988).

TABLE 2.8 MEAN LEVELS OF ORGANOCHLORINE PESTICIDE RESIDUES (mg/kg) IN COW'S MILK FROM DIFFERENT COUNTRIES.

Country	p,p'-DDT	p,p'-DDE	Dieldrin & Aldrin	α-HCH	β-HCH	Lindan-e	Hept. epox.	REFERENCE
GREAT BRITAIN, 1966-7	0.002	0.002	0.002			0.001		Maitho, 1978
U.S.A 1973-4D	0.0003	0.002	0.002			0.008	0.0004	"
CANADA 1972-3D	0.001	0.001	0.001			0.001		"
KENYA	0.0067	0.009	0.039			0.004		"
"		0.02-0.27*				0.1-0.28T*		Keating & Oduor, 1982
UNITED KINGDOM 1984-87						0.01-0.03		WHO, 1992b
SWITZER-LAND				0.004	< 0.01b	0.01		WHO, 1992a, WHO, 1992b
FRANCE					0.03b	<0.1		WHO, 1992a, WHO, 1992b

Key.
a- mg/kg
b- Milk and milk products
c- mg/litre
n - diary products
T - Total HCH i.e alpha and beta HCH
* - Butter

Following restrictions on the application of DDT and other organochlorines to livestock, to their barns, and to the forage crops on which they fed, there was a gradual decrease in residues in animal products used as human foods. The same kind of restrictions on crops eaten directly by man resulted in reduced residues in vegetable foods (WHO, 1979). Human milk has been known to contain a higher concentration of DDT than cow milk in the same country (WHO, 1979; Kanja, 1988; Maitho, 1978; Olszyna-Marzys et al., 1973). This has been attributed to the diet, as man is placed at the top of most foodchains.

Food of animal origin has been found to contain high levels of organochlorine pesticide residues due to biomagnification of compounds in the food chain. Hofvander et al. (1981) reported that the major non-occupational source of organochlorine pesticide residues in Sweden was the diet, especially foodstuffs of animal origin such as fish from inland waters and the Baltic sea. Noren (1983c) also reported that levels of organochlorines and PCBs in human milk was related to the dietary habits of the mothers. High levels were reported in mothers who regularly ate fatty fish as compared to lacto-vegetarians. Vegetarian women who consume food low in the food chain, were reported to have had lower levels of the organochlorine pesticide residues - p.p'- DDT, dieldrin, β - HCH, pp - DDE, heptachlor epoxide and oxychlordane (Jensen, 1983).

Other studies have demonstrated that non-vegetarian mothers excreted relatively higher amounts of organochlorines pesticide residues through body tissues (Noren, 1983c; Kanja, 1988). As reviewed by Jensen (1983) in some studies, a significant positive correlation has been observed between DDT content in human milk fat and cigarette smoking.

2.4 CURRENT METHODS FOR THE ANALYSIS OF ORGANOCHLORINE PESTICIDES IN HUMAN AND COW MILK.

Various methods have been used in pesticide residue analysis in different laboratories. The choice of the method may be based on applicability to a wide range of pesticides and commodities, speed, cost, availability of reagents and equipment (Kanja, 1988). Analysis of different pesticide residues in milk samples require analytical methods of high sensitivity and accuracy. The analysis of pesticide residues is often complicated by the fact that the spray history is not known. Table 2.9 shows some of the published methods of analysis for organochlorine compounds in milk. The method developed by Brevik for fat extraction from milk (and widely researched by others) has shown good recoveries of a wide range of organochlorine compounds. Acid (concentrated sulphuric acid) and base (ethanol and potassium hydroxide) clean-up are used to reduce the levels of any undesirable coextractives from the organochlorine compounds, in order to limit or lower their detection limit and reduce the contamination of the GLC column and detector.

These two clean-up procedures have been reported to produce very clean extracts with milk samples. GLC with electron capture detector has been reported to be most accurate method for qualitative and quantitative estimation of organochlorine pesticide residues.

Table 2.9 Review of methods for the analysis of organochlorine pesticide residues in milk.

Sample	Solvent Extraction	Clean-up	Identi-fication	Reference
Milk	Extract diethyl ether and hexane. Partition acetonitrile extract hexane		GLC	WHO, 1984b
Human milk	Acetonitrile/ hexane	Florisil	GLC/ ECD	Winter et al.; 1976
Human milk	formic acid/ hexane	H_2SO_4	GLC/ ECD	Siddiqui et al.; 1985
Milk	Acetonitrile	Florisil /H_2SO_4 activated charcoal	GLC/ ECD	Kaphalia et al.; 1985
Human milk	Dimethyl formamide	alumina	GLC/ ECD	Warnez et al. 1983
Human milk	20% CH_2CL_2/hexane hexane/aceto-nitrile	Florisil	GLC/ ECD TLC Alumina ($AgNO_3$)	Stacey et al.; 1985.
Human milk	ethanol/diethyl ether/Petrol ether	florisil	GLC/ ECD	Adamovic et al.; 1978
Milk	Potassium oxalate +ethanol,ethyl/ light petroleum water/dimethyl formamide, hexane	Alumina	TLC/ GLC	Noren et al.; 1968
Milk	Acetonitrile/ hexane	florisil	GLC/ ECD	Kaphalia et et al.; 1985
Human milk	Acetone/hexane	florisil	GLC	Kaphalia et et al.; 1985
Human milk	Acetone/hexane	Conc. H_2SO_4 10% Potassium hydroxide in methanol	GLC/ ECD	Kanja, 1988
Cow milk			GLC/ ECD	Maitho, 1978

Key.
GLC - gas liquid chromatography.
ECD - electron capture detector.
TLC - thin layer chromatography.

2.5 SUMMARY.

In conclusion, this literature review shows that several studies have detected the presence of residues of most organochlorine compounds in tissues of organisms of all trophic levels. Extensive information is available on the occurrence of these compounds and their metabolites in human fat, blood and milk from developed countries. However, there have been limited information especially on milk from Kenya. Continued monitoring of the environmental levels of these compounds is justified in order to learn the rate at which concentrations decline following progressive reduction in their use. Therefore, more extensive monitoring is needed for countries (like Kenya) where base data are either not available or insufficient and where use of these insecticides has been found necessary (e.g for malaria control programmes). The rational use of pesticides can be achieved through an integrated pest management system, which utilizes various techniques to control pests and only uses pesticides when necessary.

As suggested by Rudd, the future role of the entomologist is likely to be that of an "ecological counsellor" prescribing and regulating the use of insecticides much as the physician prescribes medicine (Brooks, 1974). In this way dangers to human health and the environment will be minimized.

CHAPTER 3

MATERIALS AND METHODS.

3.1 "Description of the study area".

3.1.1 Geographical location and climatic conditions.

Machakos district, is one of the six districts within Eastern Province. It has an area of approximately 14,250 sq Km ranging from 125 Km, wide in the north and less than 20 Km wide in the south. It extends some 275 Km North west to south east. Administratively it is divided into ten divisions (see Table 3.1 and Map 1). One division was chosen for the study using the following criteria:

a) Ease of transportation and general accessibility to the study population.

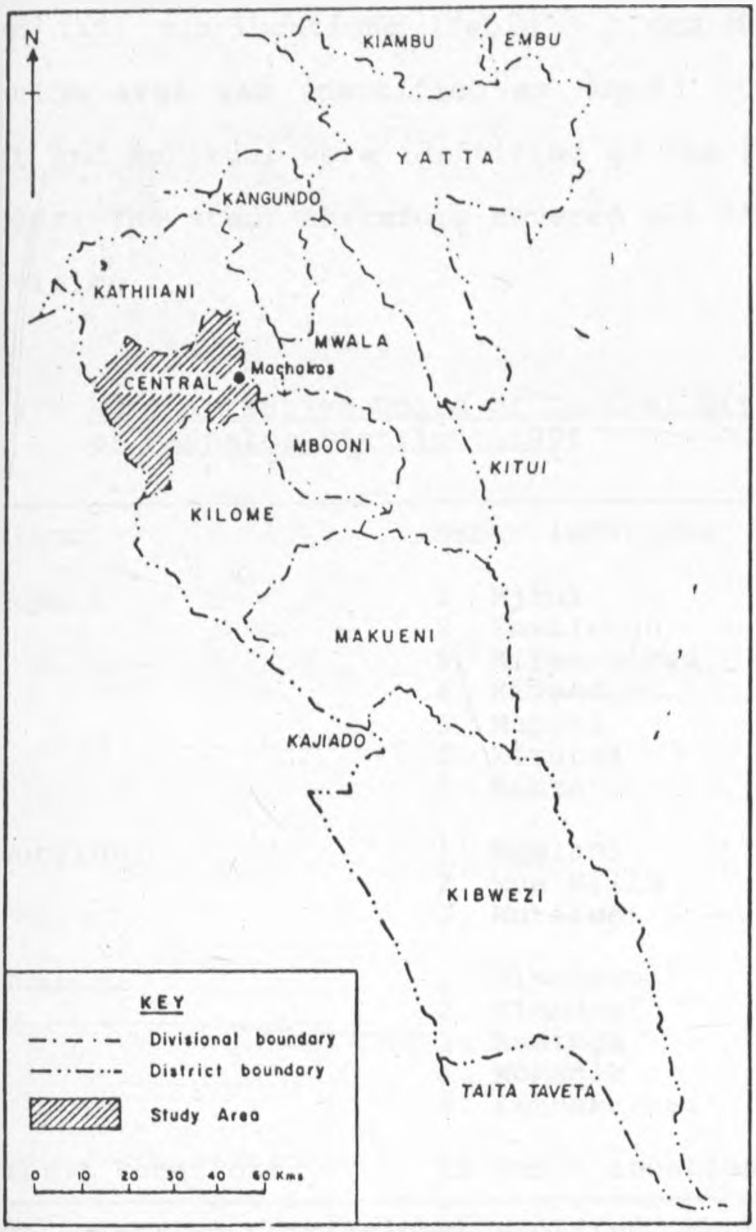
b) Availability of a low and high potential production zone within the same division.

(c) Population density of the division.

Table 3.1. Administrative Units of Machakos District - Distribution of Location and Sub-locations.

Name of Division	Number of Locations	Number of sub-locations
1. Central	3	15
2. Kathiani	3	17
3. Mwala	7	46
4. Mbooni	4	27
5. Yatta	5	18
6. Kilome	6	34
7. Makueni	5	29
8. Kangundo	4	24
9. Kibwezi	4	23
Total	41	233

Source: District Commissioner's Office, Machakos.



MAP I: LOCATION OF CENTRAL DIVISION IN MACHAKOS DISTRICT

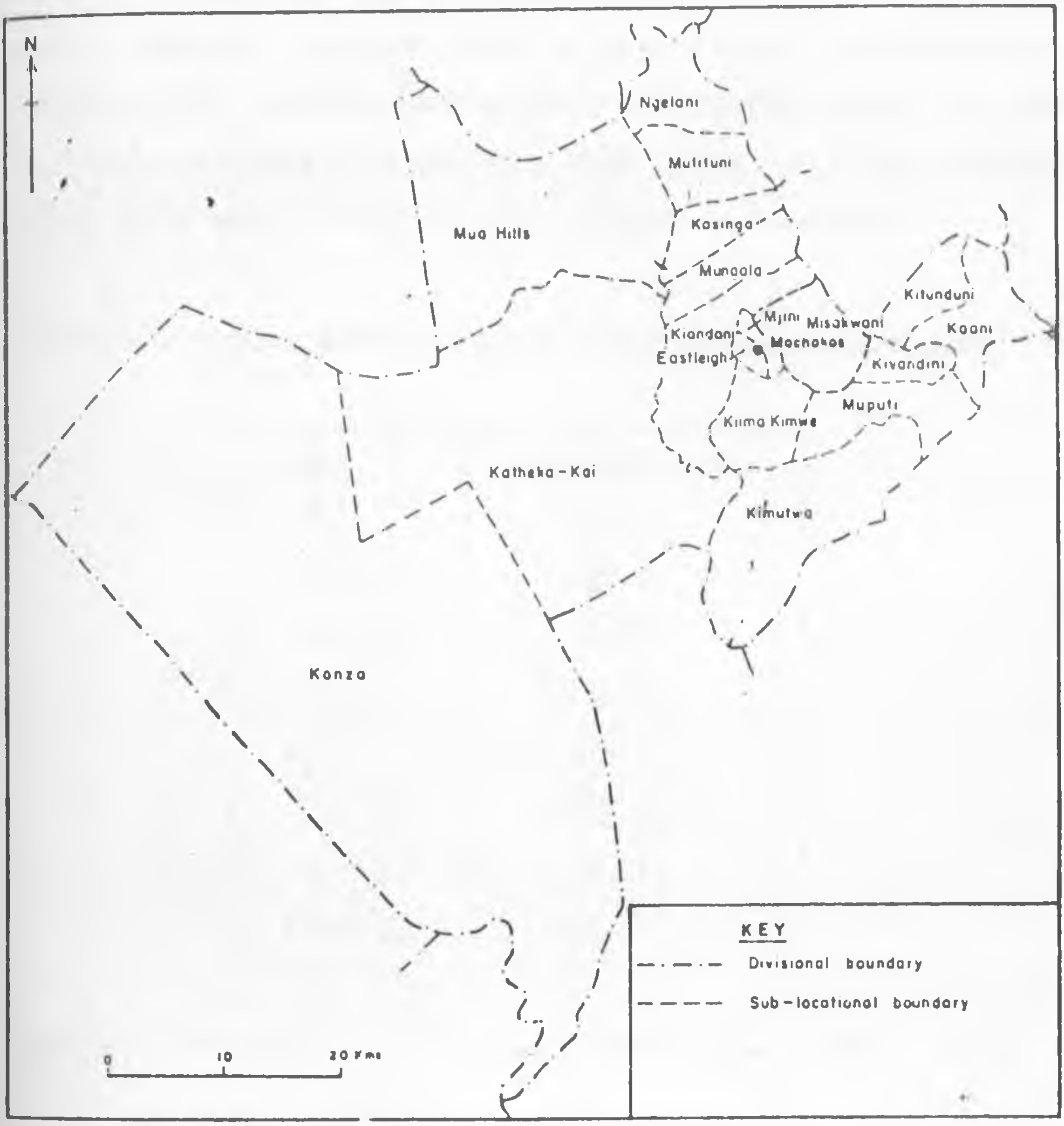
Using the above features Machakos Central division was chosen as the study area. This Division has three (3) locations and fifteen (15) sub-locations (Table 3.2 and Map 2). The low production area was identified as Muputi location whereas Mumbuni and Mutituni were identified as the high production locations. The study therefore covered all the locations of the division.

Table 3.2 Administrative Units of Central Division of Machakos District, 1991.

Location	Sub - locations
a. Muputi	1. Mjini 2. Eastleigh 3. Kiima-kimwe 4. Kivandini 5. Muputi 6. Kimutwa 7. Konza
b. Mutituni	1. Ngelani 2. Mua Hills 3. Mutituni
c. Mumbuni	1. Misakwani 2. Kiandani 3. Kasinga 4. Mungala 5. Katheka-kai
Total: 3 locations	15 Sub - locations

Source: District Commissioner's Office, Machakos.

Machakos district falls within the zone of arid/semi arid lands, more characterized as an area of extreme variability of rainfall. Typically good seasons are interspersed with extremely dry periods and variation in the onset of rainy



MAP 2 : CENTRAL DIVISION - ADMINISTRATIVE SUB-LOCATIONS seasons adds to the difficulty of ensuring adequate crop production. The rainfall varies roughly with the altitude. Annual rainfall averages slightly over 1000 mm. The rainfall has a bimodal pattern with a significant difference in distribution between seasons during different years, as can be noted in Table 3.3 Machakos Town. The two rainy seasons occur from March to April and November to December.

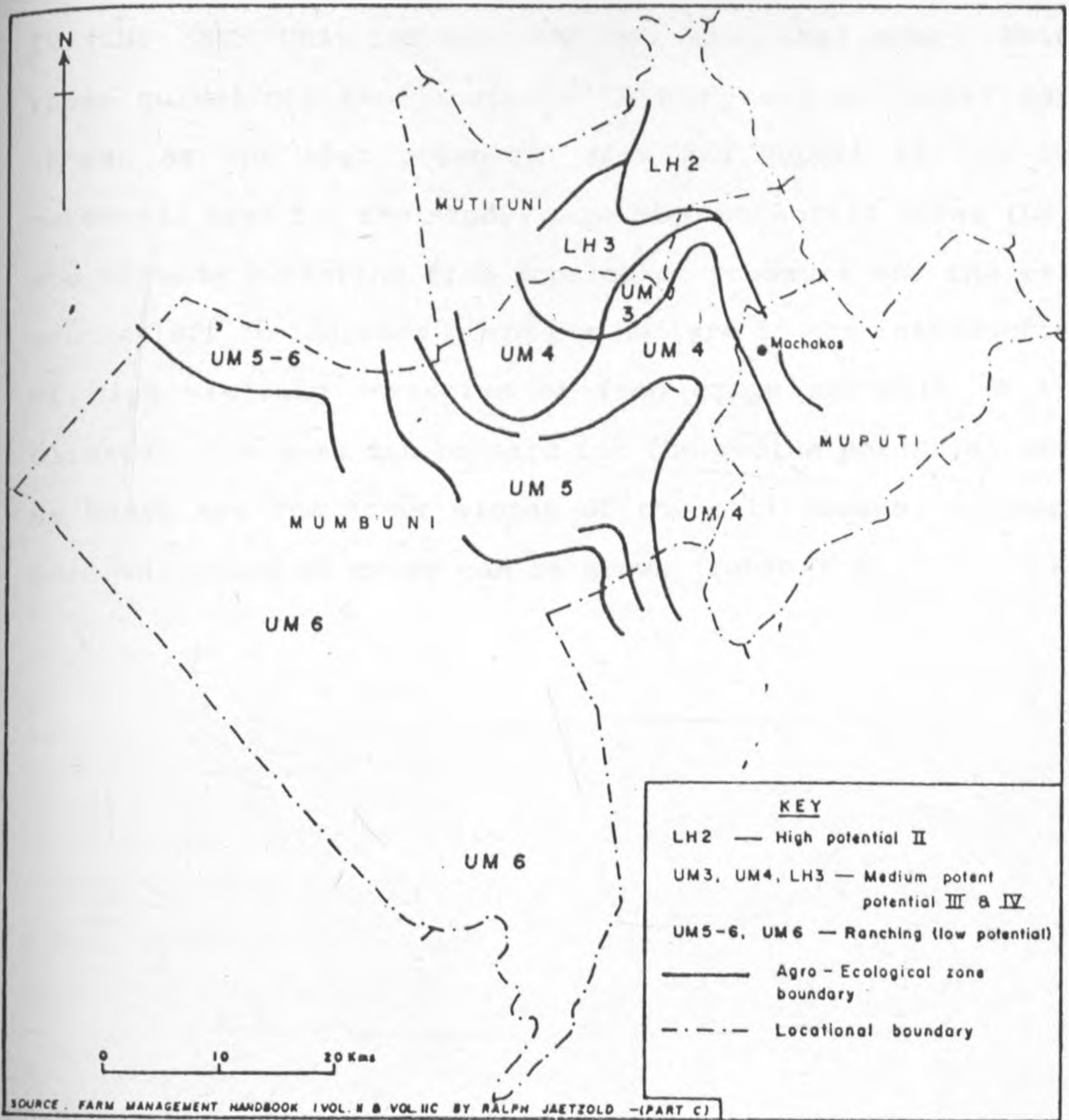
Table 3.3 Annual Rainfall Totals for Machakos Town (mm): 1973-80.

Year	Rainfall (mm)
1973	772.7
1974	897.6
1975	722.3
1976	670.8
1977	955.7
1978	1112.9
1979	122.1
1980	1021.1

Source: Machakos District Development plan (1989 - 1993)

3.1.2 Agriculture and Livestock Potential

A large portion of Machakos Central Division has high agricultural potential. Land use is distributed along agro-climatic zones. As shown in Table 3.4 and map 3 Mumbuni and Mutituni locations fall within the high potential II (LH2)



SOURCE . FARM MANAGEMENT HANDBOOK (VOL. II & VOL. IIC BY RALPH JAETZOLD -(PART C)

MAP 3: CENTRAL DIVISION - AGRO-ECOLOGICAL ZONES AND ADMINISTRATIVE LOCATIONS zone while Muputi within the medium potent potential III and IV (LH3, UM2, UM3, LH3-LH4, 4M3-UM4, LM3, LM4) zone. Using these guidelines two locations (Mumbuni and Mutituni) were chosen as the high potential area and Muputi as the low potential area for the study. The high potential areas (LH2) are already suffering from population pressure and the only avenue left to increase yield per hectare is the introduction of high yielding varieties of food crops suitable to the climate. The same can be said for the medium potential zone as these are the lower slopes of the hill masses, although more varieties of crops can be grown (Table 3.4).

Table 3.4 Agro-ecological zones and land use patterns.

Agro-climatic zone	Percentage of district agric-area	Description of area	Mainland land use activity
High potential II LH2	5.4%	Hill masses of Iveti, Mua, Mbooni, Kilungu, Mbitini and Kangundo	Coffee, maize beans, citrus forest
Medium potent potential III and IV, LH3, UM2, UM3, LH3-LH4, UM3-UM4, LM3, LM4	38.2%	Lower slopes of hills and adjacent locations of Okia, Nzaui, Mukaa, Kisau, lower Mbooni Mwala, Kiteta, Matungulu, Mitaboni Masii, Muthetheni, Muputi, Ithanga and Kalama	coffee, maize cotton, beans pigeon, peas, sun-flower, sorghum, citrus fruits, live-stock rearing
Low potential LM5, UM5, UM5-UM6, LM6	56.4%	Whole of Yatta, division, Wamunyu, Kibauni, Makueni, Kikumbuyu, Ngwata, Makindu and Mtito-Andei	Maize, pepper sorghum, beans, cotton pigeon peas, kapella, sunflower livestock rearing

Source: 1989-93 Machakos District Development Plan

Some livestock rearing is carried out in Machakos Central division mainly for commercial purposes. Zero grazing is mainly practised, especially in the high potential areas and to a great extent in Muputi location. Table 3.5 shows the estimated populations of cows and the milk production figures for the district between 1988 - 1990. A quarter of the population of cows in the district were from Machakos central division.

Table 3.5 Milk Production in Machakos District, 1988-90.

Year	Estimated Popn. cows in central division	Estimated Popn. of cows in district	Milk Produ. vol. in district (Kgs)	Income from Milk Sales
1988	9,753 cows	41,181	6,006,120	21,133,565.80
1989	10,243	43,017	5,706,579	23,697,874.65
1990	10,550	42,005	5,541,496	23,054,290.00

Source: District records of Ministry of Livestock Development - Animal production department.

3.1.3. Population and the people of Machakos Central Division.

The people of Central division belong to a bantu tribe, the Akamba. Their traditional diet since a long way back, is a mixture of the staple foods, maize and beans or peas, and is consumed practically everyday in large quantities. This traditional dish is sometimes supplemented with vegetables, some fat and milk. Most inhabitants are engaged in smallholder farming, which constitutes their main source of income.

The population of Machakos district was reported in the 1991 economic survey as 1,373,000 with an intercensal (1979-1989) growth rate of 3.09%. The district's projected population by administrative units is shown in Table 3.6.

Table 3.6 Population Projections of Machakos District by Division: 1979-90.

Division	Residential area (sq. Km)	1979	1988	1989	1990
Central[a]	727	82,272	121,136	125,715	130,575
Kathiani	1,069	74,415	109,567	113,709	118,105
Kilome	1,323	152,428	224,432	232,917	241,920
Mbooni	535	92,666	136,439	141,598	147,071
Makueni	2,005	125,974	185,481	192,494	199,935
Kibwezi	3,400	98,980	145,736	151,246	157,092
Yatta	2,459	137,258	202,096	209,736	217,843
Kangundo	598	133,012	195,844	203,248	211,105
Mwala[b]	1,332	125,517	184,808	191,796	199,210
Total	13,448	1,022,522	1,505,539	1,562,459	1,622,856

Source: Machakos District Development Plan 1989-93

a - These divisions did not exist at the time of 1979 census.
b - Residential area: - where man can settle excluding water bodies i.e rivers, dams, lakes etc.

3.1.4. Health facilities.

Machakos Central division is well served by the Machakos district hospital which is at a close proximity. Other health facilities include Kaani, Kimutwa, Mumbuni and Ithaani dispensaries. There is one mission dispensary (Ngelani dispensary) and three private hospitals/clinics (i.e. Machakos nursing home, Bethany and S.K. Maingi (herbal hospital).

3.2 "Methodology of the study".

This was a cross-sectional study carried out to establish the levels of organochlorine pesticide residues in breastmilk and cows milk from central division of Machakos district. It consisted of two phases, i.e., the preparatory phase (pilot study) and the definitive (actual) study phase. The actual study phase consisted of two parts - the qualitative wherein a questionaire was administered and the quantitative in which milk samples were collected and analysed in the laboratory.

3.2.1. The qualitative phase of the study.

3.2.1.1. Pilot study.

During the pilot study which was carried out in Kalama, Athi River and Machakos General hospital, the sampling techniques for breastmilk and cow milk were tested. The questionaire was also tested for clarity. The pretested questionnaire and sampling techniques were modified where necessary before use in the main study. Similarly, laboratory analysis methods were tested and modified and equipment for analysis assembled. At the same time, field and laboratory assistants were trained as described below.

3.2.1.2 Field Assistants.

All field assistants engaged in the study were either residents of the area, or agricultural/or Livestock staff working in the area, or health workers who were conversant with English and Kikamba languages. The assistants were trained on the questionnaire administration and sample collection techniques before the pilot study and also before the main study by the principal investigator.

Before commencing on the main survey, the same field assistants plus a few others were retrained for one week at the Machakos General Hospital for breastmilk collection and adminstration of questionnaire. The assistants to collect cows milk were trained in Muputi sub-location for two days.

All questionnaires and samples collected were checked at the end of each working day for completeness and proper labelling and finally the samples stored frozen at - 18° C to await laboratory analysis.

3.2.1.3. <u>Laboratory Assistants</u>.

The laboratory assistants were trained on the laboratory analysis of samples by the principal investigator for one week before commencing the actual analysis of samples collected. Two of them were picked and were closely supervised during the fat extraction and clean-up of samples for GLC analysis by the principal investigator. The principal investigator worked with a consultant chromatographer during the whole period of GLC analysis of all samples collected.

3.2.2 <u>Selection of Mothers for sampling of breastmilk</u>.

To be able to compare data collected from this study to other studies from different countries, the criteria given by Slorach and Vaz (1983) was also used to select the mothers.

- Mothers must have been residents of the study area for five years or more.
- Age of mothers should have been between 18-30 years.
- Parity - Mothers should be nursing first or second child.
- Both mother and child should have been healthy. Women with visible skin diseases on breast were excluded.
- Mothers should have been breastfeeding only one child.
- Mothers' milk sample could be collected between 1 week - 4 months post partum.

3.2.3 Sampling Procedure

3.2.3.1 Sample size determination

Organochlorine pesticide residues were found in all the breastmilk samples analyzed in Kenya (Kanja, 1988). Therefore sample size was determined using the following formula.

$$n = \frac{Z^2 pq}{d^2}$$

where

Z = The desired confidence level (1.96)

P = Proportion of milk with any level of organochlorine (0.9)

q = Proportion of milk with no organochlorine (0.1)

d = Degree of accuracy (0.05)

n = Sample size

$$n = \frac{(1.96)^2 (0.9)(0.1)}{(0.05)^2}$$

$$= 138.3 \simeq 140 \text{ breastmilk samples.}$$

Considering the expenses and the time involved in organochlorine residue analysis, a total of 150 samples of breastmilk were collected from both the low and high potential production zones, but eighty samples of cows milk were collected and pooled together to make 21 analysis samples.

3.2.3.2 <u>Sampling Method for breastmilk.</u>

Figure 1 presents the sampling methodology. All the eight health centres in the Division were used as sampling points for breastfeeding mothers. Using the criteria in 3.2.2, 150 mothers were chosen on a daily basis from all the health centres. Breastmilk was collected from the mothers daily basis until the required sample size was attained for each of the two zones. Initially, the questionnaire was administered to each of the mothers from whom breastmilk was to be collected.

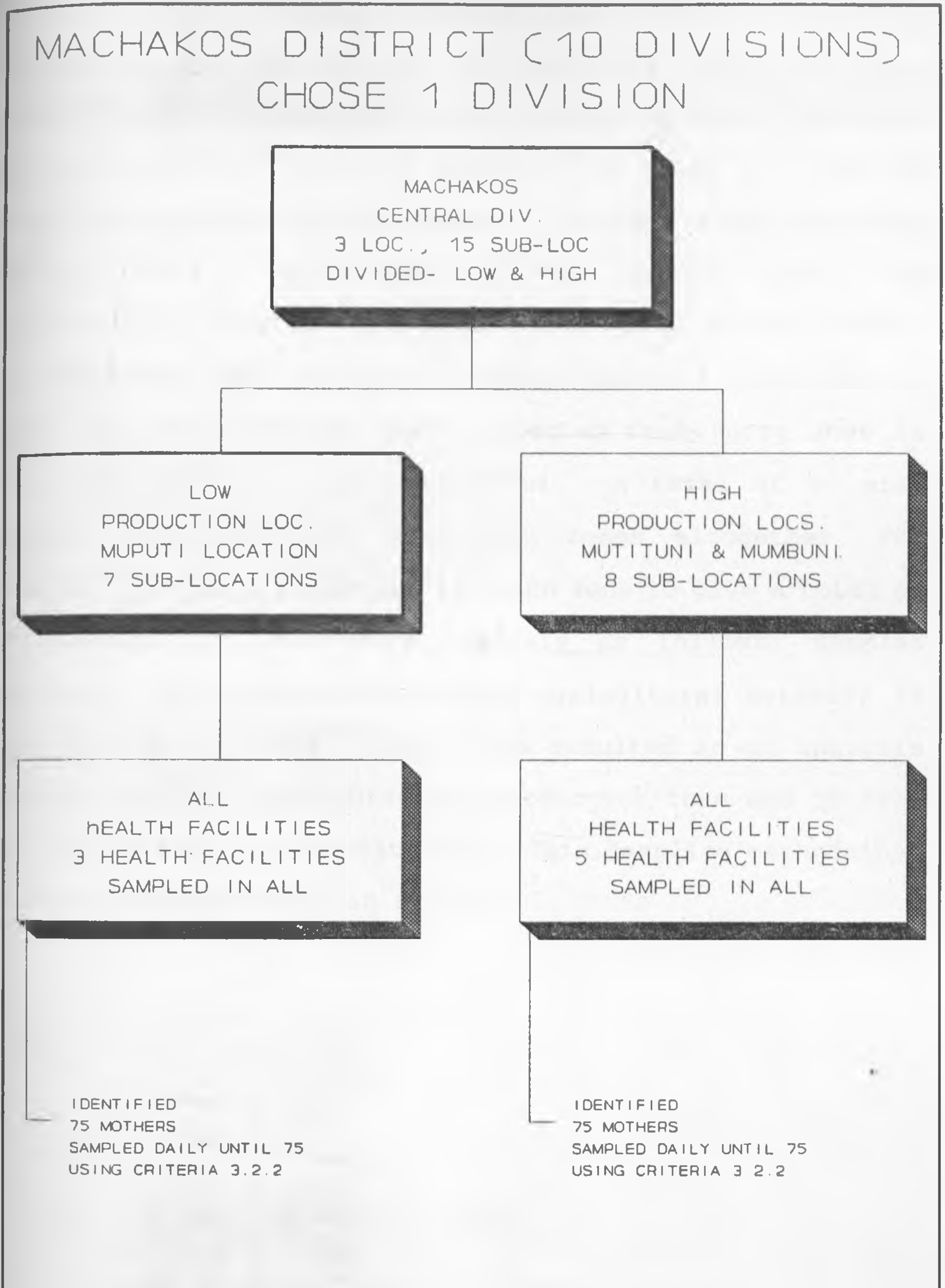


Fig. 1 SAMPLING OF BREASTMILK.

3.2.3.2 Sampling of cow milk.

Initially, the agricultural or livestock staff filled a questionnaire on pesticide usage in each location. Cow milk was collected from the same areas of the study (i.e low and high potential production zones). Farmers with lactating cow's were identified from each zone by agricultural/livestock extension staff. Cows to participate in the study were randomly selected from all locations in each zone. Milk samples were collected from forty cows in each zone (low or high production). A total of 80 milk samples were collected from both zones altogether. The samples were pooled together for each zone to give a total of 21 samples for laboratory analysis as follows: Samples collected from areas with similar agricultural activity in each sublocation were pooled. This resulted in 11 analysis samples from the high potential production zone and 10 from the low potential production zone. This sampling methodology is shown schematically in Figure 2.

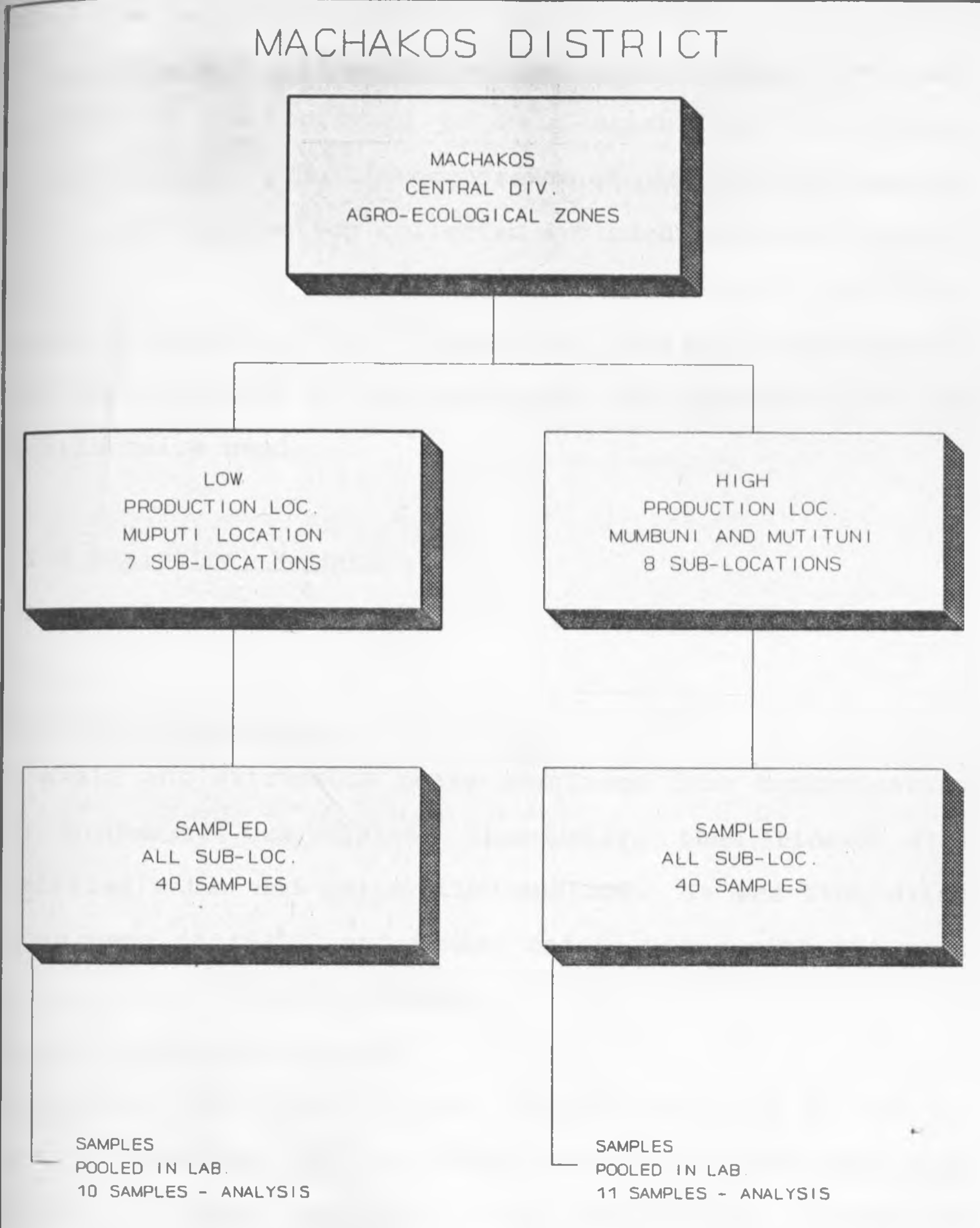


Fig. 2 SAMPLING OF COW'S MILK.

3.2.4 The Qualitative study.

With the help of a pretested questionnaire, relevant information was obtained to help establish the probable etiologic factors for the occurrence of pesticide residues in milk. The information collected included date and place of collection of milk samples, mother's age, parity, days post-partum, occupation, diet, staple food, and pesticide usage in the homestead/farm of the respondent (see appendix 2 for the questionnaire used).

3.2.5 Analytical Methods.

3.2.5.1 "Preparation of glassware and reagents for GLC".

Cleaning of glassware

To avoid any extraneous peaks resulting from contamination all glassware was cleaned thoroughly, then rinsed with distilled water and redistilled acetone. It was then dried in an oven at 150^0 C and rinsed before using with the same solvent to be used in analysis.

Redistillation of solvents

Cyclohexane and acetone were redistilled using a Tom-jet rotary evaporator with a vacuum constanter and heat pump solvent recovery equipment. The redistilled cyclohexane (10ml) was concentrated twenty times to 0.5 ml. 1µl of this was injected into the gas chromatograph and if no peaks were observed, then the cyclohexane distillate was suitable for pesticide analysis. Ten (10 ml) of acetone were added to 5 ml of redistilled cyclohexane in a separatory funnel and shaken. To separate the two fractions 2 ml of 2% sodium chloride were added and shaken. The cyclohexane layer was taken out and concentrated to 0.5 ml and checked in the GLC as above.

Cleaning of Sulphuric Acid

500 ml Sulphuric acid was added to 100 ml redistilled cyclohexane in a separatory funnel and shaken. The cyclohexane layer was discarded and the cleaning repeated three times. 10 ml of cyclohexane was taken from the third rinse and concentrated to 1 ml and tested by GLC.

3.2.5.2 Collection and preparation of milk samples for analysis.

Milk samples were obtained by manual expression into prewashed glass containers fitted with teflon caps. The samples were frozen immediately or stored at about 4°C until they were transfered to freezers in the study laboratory. Care was taken to avoid contamination during and after sample collection. In the study laboratory, the samples were frozen at - 18°C or below to await analysis.

For analysis, the milk samples were thawed and homogenised using an Ultrasonic disintegrator for about 2 minutes. Ten grammes were taken for extraction and clean-up using the method described by Brevik (1978), with slight modifications, as follows:

(a) To the sample, 35 ml cyclohexane: acetone (2:1.5) mixture were added and mixed using an ultrasonic disintegrator for 2 minutes. The mixture was centrifuged at 3000 rpm for 5 minutes. The upper organic phase was transferred to an empty pre-weighed glass tube with teflon cap. The residue was further extracted with 15 ml of cyclohexane: acetone (2:1) mixture. The organic phases were combined and evaporated on a sand bath (50^0 C) under a stream of dry nitrogen until almost dry. The lipid residue was cooled and weighed to estimate the fat content.

(b) Clean-up.

The lipid residue was redisolved in cyclohexane to an approximate concentration of 0.05 g fat/ml cyclohexane and 1 ml taken for acid or base clean-up.

Acid clean-up- 1 millitre of the redissolved lipid residue was shaken with 2 ml concentrated sulphuric acid and allowed to stand in the dark for 1 hour. It was then centrifuged at 3000 rpm for 5 minutes and the organic phase tranferred into a separate tube ready for GLC analysis.

Base clean-up - Two potassium hydroxide pellets, 0.1 ml distilled water and 1 ml ethanol (99.6%) were mixed and allowed to dissolve. 1 millitre of the lipid solution was then was added and the mixture was placed in a water bath at 50^0 C for 30 minutes then cooled at 4^0 C for 10 minutes to stabilize cyclohexane. Five millitres of a mixture of sodium chloride and 85% orthophosphoric acid (11.6g + 6.83 ml in 1000 ml distilled water) were added and mixed well. It was cooled and allowed to separate in a fridge then centrifuged at 3000 rpm for 10 minutes. The organic phase was transferred to a separate test tube, anhydrous sodium sulphate added and left to settle. The organic supernatent was transferred to a glass tube ready for GLC analysis.

3.2.5.3 GLC Analysis.

Preparation of standard solutions

The standard solutions used were Promochem Pesticide mixtures supplied by Promochem - Germany. Promochem standard mix XIII in Toluene was used extensively in the identification of peaks. It was originally prepared as a BGA Monitory programme 1988 standard solution and it contains thirty one (31) organochlorine compounds each at a concentration of 10 ng/µl. Promochem standard mix V in cyclohexane containing eight (8) organochlorine compounds was used to spike cow milk. Working standard solutions were diluted with redistilled cyclohexane.

Gas chromatographic equipment.

The acid-cleaned and some base-cleaned extracts were analysed for pesticide residues using a gas chromatograph (varian GC 3400 with varian data system DS 601 and spectra physics integrator). The equipment was equiped with ^{63}Ni electron capture detector (sensitivity range 10) and megabore fused silica capillary column of length of 60 metres, 0.32 mm internal diameter and 1.5 micron film thickness (DB-5 - composed of 5% methyl polysiloxane). The carrier gas was high purity nitrogen (whitespot). The injector flow was set at 50 (24 psi) and make up ECD set at nitrogen flow rate of 50 ml/minute. The minimum detectability was greater than 0.1 picogram lindane with a nitrogen carrier using the temperature programme:

80° C (1 min) <u>25°C/min</u> 180° C (1 min) <u>4° C/min</u> 280° C (5 min) <u>10° C/min</u> 300°C (20 min).

<u>Preparation of gas chromatograph</u>

The gas chromatograph was left on for a whole day (day and night) to condition the column for pesticide analysis. A blank was first injected into the column each day to check on the column performance. The performance of the column was also monitored at regular intervals during the day to detect any changes in peak shape and retention times.

Chromatographic analytical Procedure.

1 µl of the cleaned sample or standard was injected into the capillary megabore column. From the chromatograms obtained, the retention times of the sample components were compared to the retention times of the standard components by matching the peaks on the two chromatograms.

The integrated peak areas of the sample were matched with the peak areas of the standard and the concentration of the compounds of interest calculated using the following relationship:

x units of standard mixture a represent b nanogram(ng).

y units of compound c represent

$$c=\frac{yb}{x}ng$$

Where x and y are peak areas of the same substance in the standard and sample respectively, and b is the concentration of the substance in the standard mixture. The above level of the compound was then used to calculate the concentration of the component as milligrams (mg) of pesticide per kilogram (kg) of sample.

The identity of the oc peaks were confirmed by comparing the peaks from similar samples which had been either acid or base- cleaned to the standard peaks.

Validation of analytical procedure.

The accuracy and precision of the analytical methods to assess the relationship between the actual amounts of OC's present and the results obtained was done by the analysing cow's milk samples spiked with different concentrations of the standard mix V. The analysis was repeated until good reproducible recovery data was obtained. The spiked samples were analysed at regular repetitive intervals as an internal check on the performance of the laboratory and analytical methods used. The spiking levels were 0.005, 0.002 and 0.001 of standard mix V. This gave the performance level of the laboratory as well as the reproduceability of the analytical procedure.

3.3 CALCULATION OF DIETARY INTAKE OF PESTICIDE RESIDUES BY INFANTS.

Table 3.7 was used in the calculation of dietary intake of the various organochlorine pesticides. During calculation it was assumed that at the age of 4-6 months the infant is at the peak of milk consumption. The average weight of this age group was assumed to be 5 kg and therefore the milk consumption of 655 g/ day was assumed to be the average intake since over 70% of the energy needs of this age group comes from milk.

Table 3.7 AVERAGE CONSUMPTION OF COW MILK AND PERCENT ENERGY DRAWN FROM MILK BY INFANTS.[A]

AGE (MONTHS)	AVERAGE CONSUMPTION IN FOUR EUROPEAN COUNTRIES g/day	% ENERGY FROM MILK
1 - 3	627	
4 - 6	655	70 - 73
7 - 9	596	30 - 44
10 - 12	740	30 - 44

[A] - WHO, 1985 GUIDELINES FOR THE STUDY OF DIETARY INTAKES OF CHEMICAL CONTAMINANTS.

The dietary intake of the pesticide residues from human and cow milk was obtained by multiplying the residue level in the milk from the study area, by the amount of the milk consumed by infants. The estimated maximum daily intake (EMDI) in this study was calculated using the formula (WHO, 1989c):

$$EMDI=\frac{AB}{C}$$

Where A = Milk consumption of child (kg per child per day)

B = Residue level in milk (mg pesticide per kg of milk)

C = Average body weight (5 kg)

No adjustments were made for processing and cooking losses since these processes are not performed on breastmilk.

3.4 <u>"Data analysis"</u>.

All the data was entered and cleaned in Data base III and Lotus 1 2 3 / Quattro Pro (at the Applied Nutrition computer laboratory). Data was analyzed in EPI INFO and SPSS computer packages. The Harvard graphics 3.0 package was used for graphs.

CHAPTER 4.

RESULTS.

The results of this study are presented in two parts: Initially results from the descriptive study, then results of the laboratory analyses of milk samples.

4.1 RESULTS OF DESCRIPTIVE STUDY.

4.1.1 Characteristics of the mothers from whom breastmilk was sampled.

All the mothers used in the study were from the same ethnic group the Akamba. They were aged between 17 - 30 years, with 55% being in the 17 - 21 age group and 45% in the 22 - 30 age group. Most of these mothers were housewives (83%) who had lived in the study area for over 5 years. Ninety six percent of the mothers had lived in the study area for over 17 years. Fifty eight percent of mothers were breastfeeding their first child while 42% were breastfeeding their second child. Majority were married (61%) while the others to (38%) were either single or divorced.

All the 150 respondants practiced some form of farming, growing cereals and grains. In addition, 99% were growing vegetables, 99% fruits, 97% root crops and 62% cash crops. No mother reported using tobacco. The main staple food for most of the mothers was maize and beans (99%) with vegetables. Around 80% of the mothers reported eating both animal and vegetable foods, while 19% reported that they consumed vegetarian foods with milk and eggs. Up to 99% of the mothers consumed cow milk with only 1% consuming goat milk.

The results indicate that over 50% of the mothers sampled were using pesticides either in their households, on livestock or on crop/fruit trees in their farms as shown in Table 4.1.

TABLE 4.1 PROPORTION OF MOTHERS USING THE DIFFERENT TYPES OF PESTICIDE TREATMENTS IN MACHAKOS CENTRAL DIVISION, MACHAKOS DISTRICT.

TYPE OF PESTICIDE TREATMENT	PROPORTION (n = 150)
Household	51
Food in storage	27
Crop/fruit trees in field	61
Livestock	64

4.1.2 "Use of pesticides in Machakos Central Division".

The different types and varieties of pesticides available for purchase in the area are given in Tables 4.2 and 4.3. Most of the farmers in Machakos Central division buy pesticides mainly from Kenya Grain Growers Cooperative Union

(K.G.G.C.U.) stores. Other stockists include Machakos District Cooperative Union, Cooperative Societies and Shops and/or Kiosks. The bulk of pesticides in the district are used on the farm on horticultural crops, coffee and for grain storage. The acaricides presently in use in Machakos district

Table 4.2 Volumes of Pesticide sold at K.G.G.C.U. stores (1990/91) in Machakos District.

Chemical name	Units sold	Uses	Unit Price
Fernadan D-Lindane (10 gm Units)	2906	Soil Pest. Seed dressing	7.05
Agrocide 3 - 400 gm (gamma HCH)	268	Soil Pest	7.65
Sumicidin - 1 litre	44	Insecticide	266.80
Dawa ya Mboga (BHC) (400 gm - powder)	2108	Vegetable pests	22.85
Sumithion 50% (5 litre)	2	Insecticide	930.00
Sumithion 50% (200 c.c.)	216	Insecticide	64.00
Thiodan 35% (endosulfan) (5 litres)	6	Vegetable pests, citrus, cabbage etc.	1,090.50
Thiodan 35% - 20 ml	1177		59.50
Actellic 1% - 25Kg Dust	138	Grain weevils	340.00
- 2 Kg	8014		43.00
1% - 400gm	720		17.95
- 3 kg	172		92.25
25% - 1 litre	17		279.50
1% Dust- 100gm	1485		4.60
Kocide 101 - 10 Kg	7	Fungicide for coffee berry disease	1000.00
Kocide 101- 1 Kg	103		110.00
Malathion 50% (5 litres)	9	Insecticides on stored grain and farm	600.00
Malathion 50% (1 litre)	244		154.20
Malathion 50% (200 cc)	692		63.40
Malathion - 200 gm (2%)	81	Grain in storage pests	12.50
Malathion dust - 400gm	1729		21.60
Malathion 2% - 25 Kg	10		285.00
Malathion - 2 Kg	850		47.00
Diazinon 60% (1 litre)	64	Plants like oranges mangoes and domestic pests like lice and bedbugs	

Source: Machakos town K.G.G.C.U. stores and Ministry of Agriculture Report 1990.
NB: 1) The last stocks of DDT, Aldrin, dieldrin, Heptachlor were sold in 1988.
2) There are still large stocks of Lindane.

include Sevin 85 (Carbaryl), Delnav (dioxathion), steladone, supona 50% (chlorfenvinphos), Triatix, Bacdip (quiniofos) supadip, coopatox, tick gease and Amprol 20% (which is used for Occidosis in chickens). Most farmers take their cattle to dips, where they are assisted to use the correct strength of chemicals by veterinary officers. Table 4.2 shows the volumes of pesticides sold in K.G.G.C.U. stores and other outlets in Machakos central division in the years 1990/91.

The most common public health and household pests in Machakos Central Division were flies, mosquitos, cockroaches, fleas, lice, ants, bedbugs, rats and mice. The insecticides available included mosquito coils, baygon bait, diazinon, doom, vermin powder, strike, Killtox and Johnson It (trade names) etc. It was not possible to know the active component of these insecticides, since not all manufacturers indicate on their labels.

Table 4.3 Range of Pesticides in use Central division of Machakos district in 1991.

CHEMICAL NAMES		
INSECTICIDES	FUNGICIDES	STORED GRAINS
Brigade 25%E.C.	Anthracol 70%	Malathion 50% E.C
Dipterex	Bayleton 25%wp	" 2% dust
Lebacid 50% EC	Dyrene 75% wp	Actellic 1% dust.
Metasystox 250 EC	Cupravit 50%wp	
Folimat 500 EC	Dithane M45	
Gusathion	Kocide 101	
Baygon dust	Baycor 300	
Karate 1.75%	Daconil 75% wp	
Diazinon	HERBICIDES	SEED DRESSING
Agrocide 3	Lasso EC	Kurtano
Ambush cy	Round up	Farnasan D (lindane)
Kelthane EC	Lasso atrazine	
Sumicidin	Gramoxone	
Sumithion	Kamata	
Thiodan	Tordon 101	
Cybolt 100 E		
Dawa ya mboga (BHC)		

Source: K.G.G.C.U. Stores, M.D.C.U. Stores, H.C.D.A and local stockists.

Key
Kenya grain growers cooperative union - K.G.G.C.U.
Machakos district cooperative union - M.D.C.U.
Emulsifiable concentrate - E.C.
Wettable powder - wp

4.2 "Results of Laboratory analyses".

Although a total of 150 samples of breastmilk were collected following the sampling procedure in 3.2.2, only 147 were analysed because three samples were lost during the laboratory analysis. As already stated, 21 pooled samples of cow milk from the 80 collected were also analyzed. The average fat content of breastmilk was found to be 4.4%, with a range of 4.3% (in the high potential production zone) to 5.1 % (in the low potential production zone). Pesticide residue levels are given on a fat weight basis.

4.2.1 "Organochlorine pesticides in breastmilk".

The main pesticide residues in breastmilk as given in Table 4.4, were p,p'-DDE (65*[1]), heptachlor epoxide - 63* o,p'-DDT (46*) and p,p'-DDT (36*). Residues of DDT (sum DDT) were found in 97* of the samples analysed while heptachlor (total Heptachlor was found in 66* of the samples. The mean levels (mg/kg) of other organochlorine pesticides detected were for o,p'- DDD - 0.031 (0.6*), aldrin - 0.037 (6*), dieldrin - 0.035 (1*), isodrin - 0.038 (0.6*), oxychlordane 0.065 (5*), chlordane - 0.033 (1*), endrin - 0.06 (2*), methoxychlor - 0.30 (0.6*) and HCB - 0.32 (0.6*).

[1]* - % positive samples.

TABLE 4.4 MEAN LEVELS OF PESTICIDE RESIDUES IN HUMAN MILK FROM CENTRAL DIVISION OF MACHAKOS DISTRICT.

PESTICIDE RESIDUE	NUMBER OF POSITIVE SAMPLES	LEVEL OF RESIDUES IN IN MILK FAT (mg/kg) mean (range)
p,p'-DDT	53	0.156(0.017-1.813)
o,p'- DDT	67	0.161(0.003-0.918)
p,p'-DDE	95	0.057(0.009-0.315)
o,p'-DDE	21	0.026(0.005-0.069)
p,p'-DDD	10	0.095(0.020-0.453)
sum DDT	142	0.184(0.092-1.819)
HEPTACHLOR	27	0.028(0. 07-0.131)
HEPTACHOR EPOXIDE	93	0.047(0.0 3-0.342)
TOTAL HEPATCHLOR	97	0.053(0.0 5-0.371)
α-HCH	4	0.031(0.003-0.052)
β-HCH	15	0.189(0.018-0.878)
τ-HCH	8	0.044(0.008-0.158)
σ-HCH	10	0.013(0.006-0.03)
E-HCH	12	0.032(0.011-0.124)
TOTAL HCH	33	0.117(0.003-0.879)
α-ENDOSULFAN	13	0.07(0.009-0.153)
β-ENDOSULFAN	3	0.39(0.038-0.586)

The incidence and mean levels of the various organochlorine pesticide residues were higher in the milk from the high potential production zone than the low potential production zone (Table 4.5 and Fig. 3). A significant difference ($p < 0.05$) was noted for β-HCH and τ-HCH with the two pesticide residues occuring in higher amounts in the milk from the low potential production zones than the samples from the high production areas.

TABLE 4.5 MEAN LEVELS OF ORGANOCHLORINE PESTICIDE RESIDUES DETECTED IN HUMAN MILK BY REGIONS OF MACHAKOS CENTRAL DIVISION.

ORGANOCHLORINE PESTICIDE RESIDUE	LOW PRODUCTION REGION (MUPUTI) %+VE (N=74)	LEVEL IN MILKFAT mg/kg	HIGH PRODUCTION REGION (MUMBUNI & MUTITUNI) %+VE (N=73)	LEVEL IN MILKFAT mg/kg	LOW VS HIGH PRODUCTION REGION
p,p'-DDT	32	0.14	40	0.17	NS
p,p'-DDE	60	0.05	70	0.06	NS
o,p' DDT	50	0.16	40	0.16	NS
o,p'-DDE	15	0.02	14	0.03	NS
p,p'-DDD & o,p'-DDD	7	0.14	8	0.08	NS
ALDRIN	4	0.05	8	0.03	NS
	1	0.018	1	0.053	NS
HEPTACHLOR	12	0.03	25	0.03	NS
HEPTACHLOR EPOXIDE	61	0.04	66	0.06	NS
β-HCH	7	0.45	14	0.06	S
α-HCH	1	0.31	0	NOT DETECTED	
δ-HCH	1	0.012	12	0.01	NS
E-HCH	1	0.056	15	0.03	NS
τ-HCH	1	0.159	10	0.03	S
OXYCHLORDANE	4	0.08	7	0.05	NS
CHLORDANE	0	NOT DETECTED	3	0.03	
ENDOSULFAN	9	0.08	8	0.06	NS

Key.
NS - No significant difference (p>0.05). n - Number positive.
S - Significant difference (p<0.05). +Ve - percent positive

The incidence and mean levels of the various organochlorine pesticide residues was also higher in mother's milk from areas reportedly using pesticides as compared to those areas with no reported use of pesticides (Table 4.6). The levels of sum DDT were significantly higher ($p < 0.05$) in the milk from households using pesticides than from those not using, while the levels of α- endosulfan were higher in areas not

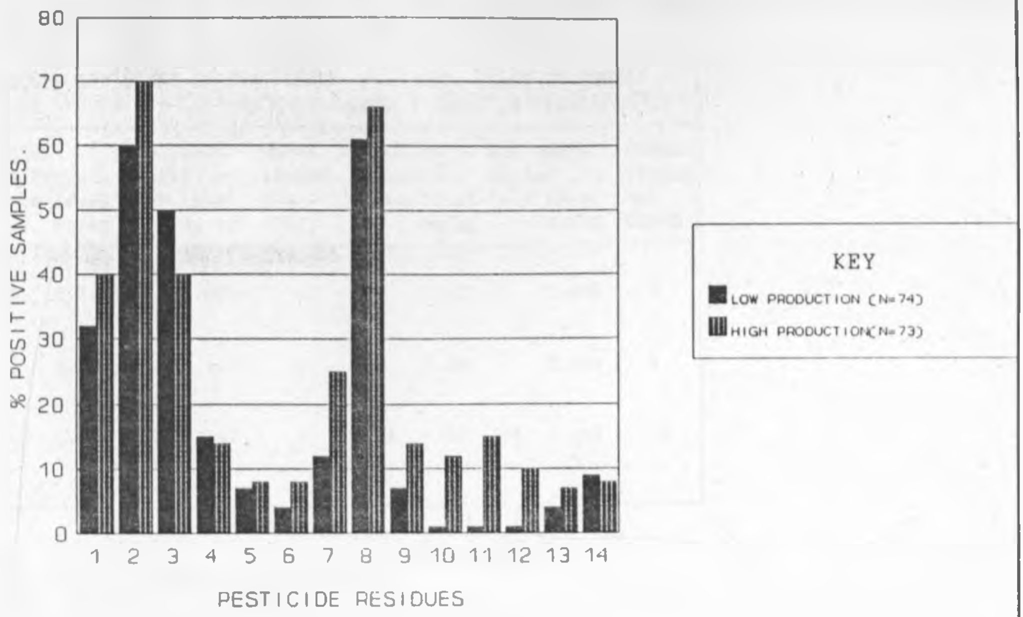


Fig 3 INCIDENCE OF OC RESIDUES IN HUMAN MILK BY AGRICULTURAL ZONES MACHAKOS CENTRAL DIVISION, 1991.

PESTICIDE RESIDUES KEY.

1. p,p'- DDT
2. p,p'- DDE
3. o,p'- DDT
4. o,p'- DDE
5. p,p'- DDD & o,p'- DDD
6. Aldrin
7. Heptachlor
8. Heptachlor epoxide
9. β - HCH
10. δ - HCH
11. E - HCH
12. τ - HCH
13. Oxychlordane
14. Endosulfan

using pesticides. The levels of oxychlordane and α-endosulfan were significantly higher in milk from areas where pesticides were used on crops/fruit trees in the farms than from areas not using.

TABLE 4.6 LEVELS OF SOME ORGANOCHLORINE PESTICIDE RESIDUES DETECTED IN HUMAN MILKFAT BY PESTICIDE USAGE IN HOUSEHOLDS AND ON CROP/FRUIT TREES IN MACHAKOS CENTRAL DIVISION.

PESTICIDE RESIDUE	HOUSEHOLDS USAGE OF PESTICIDES					PESTICIDE USAGE ON CROPS/ FRUIT TREES IN FIELD(FARMS)				
	USERS (N=76)		NON USERS (N=71)		USERS VERSUS NON USERS	USERS (N=91)		NON-USERS (N=56)		USERS VERSUS NON-USERS
	n+ve	level mg/kg	n+ve	level mg/kg		n+ve	level mg/kg	n+ve	level mg/kg	
OXYCHLOR-DANE	5	0.101	1	0.014	NS	5	0.032	1	0.363	S
α - ENDO-SULFAN	6	0.054	3	0.118	S	6	0.068	3	0.088	S
SUM DDT	40	0.41	36	0.132	S	56	0.325	16	0.167	NS

Key.
n+ve - number positive. n - Total number.
NS - No significant difference (p>0.05).
S - Significant difference (p<0.05)

Older mothers (22 - 30 year olds) showed higher incidences of sum DDT, total heptachlor, α- endosulfan and total aldrin than young mothers, while for total HCH and oxychlordane, younger mothers showed higher incidences. The mean levels of the pesticide residues were also higher in older mothers (Table 4.7).

The same trend was also observed among mothers growing cash crops as compared to those not growing cash crops (Table 4.8).

TABLE 4.7 ORGANOCHLORINE PESTICIDE RESIDUES IN HUMAN MILK BY AGE GROUP.

PESTICIDE RESIDUE	17-21 YEAR OLD WOMEN (n = 81)		22-30 YEAR OLD WOMEN (n = 66)	
	NUMBER +ve	MEAN LEVEL OF RESIDUE IN MILKFAT (mg/kg	NUMBER +ve	MEAN LEVEL OF RESIDUE IN MILKFAT (mg/kg)
OXYCHL-ORDANE	4	0.037(0.06-0.114)	2	0.187(0.011-0.363)
α-ENDO-SULFAN	4	0.071(0.009-0.147)	6	0.103(0.043-0.227)
SUM DDT	37	0.244(0.009-1.819)	57	0.3(0.01-7.706)
TOTAL HEPTACHLO	27	0.056(0.006-0.286)	44	0.083(0.005-1.412)
TOTAL ALDRIN	2	0.073(0.022-0.125)	4	0.067(0.018-0.201)
TOTAL HCH	9	0.187(0.013-0.728)	5	0.144(0.003-0.879)

Key.
+ve - positive. n - total number.

TABLE 4.8 LEVELS OF SOME ORGANOCHLORINE PESTICIDE RESIDUES DETECTED IN BREASTMILK OF MOTHERS IN AREAS GROWING CASHCROPS.

PESTICIDE RESIDUE	MOTHERS NOT GROWING CASHCROPS. (N=54)		MOTHERS GROWING CASH CROPS. (N=91)		NON-GROWERS VERSUS GROWERS OF CASH CROPS
	NUMBER +VE	MEAN LEVEL IN MILKFAT (mg/kg)	NUMBER +VE	MEAN LEVEL IN MILKFAT (mg/kg)	
OXYCHL-ORDANE	2	0.238	4	0.11	NS
α-ENDO-SULFAN	4	0.149	6	0.051	S
SUM DDT	39	0.227	55	0.314	NS

Key.
S ($p<0.05$) - Significant difference. NS ($p>0.05$) - No significant difference.

Mothers breastfeeding their first child produced milk with higher mean levels of the various pesticide residues as compared to those breastfeeding their second child (Table 4.9).

TABLE 4.9 ORGANOCHLORINE PESTICIDE RESIDUES DETECTED IN HUMAN MILK BY PARITY OF THE MOTHERS.

PESTICIDE RESIDUE	MOTHERS BREASTFEEDING FIRST CHILD - LEVEL OF RESIDUE IN MILKFAT (mg/kg).(N=87) MEAN FAT% = 4.5%	MOTHERS BREASTFEEDING SECOND CHILD - LEVEL OF RESIDUE IN MILKFAT (mg/kg).(N=60) MEAN FAT %= 4.2	FIRST VERSUS SECOND CHILD
OXYCHLO-RDANE	0.098	0.064	NS
CHLORDANE	0.032	0.035	NS
α-ENDO-SULFAN	0.092	0.7	NS
β-ENDO-SULFAN	0.387	NOT DETECTED	
SUM DDT	0.322	0.193	NS
TOTAL HEPTACHLOR	0.078	0.063	NS
TOTAL ALDRIN/ DIELDRIN	0.065	0.073	NS
TOTAL HCH	0.155	0.179	NS

Key.
S ($p < 0.05$) - Significant difference.
NS ($p > 0.05$) - No significant difference.

In general, the mean levels of the pesticide residues in breastmilk from this study were low compared to other studies (see Tables 2.3 - 2.7) and codex alimetarius commission recommendations (FAO & WHO, 1986). However, the mean levels of endrin and oxychlordane/chlordane in human milkfat in Machakos were higher than the codex maximum residue limits. Fig. 4 shows the levels of the various residues as compared to Codex maximum residue limits.

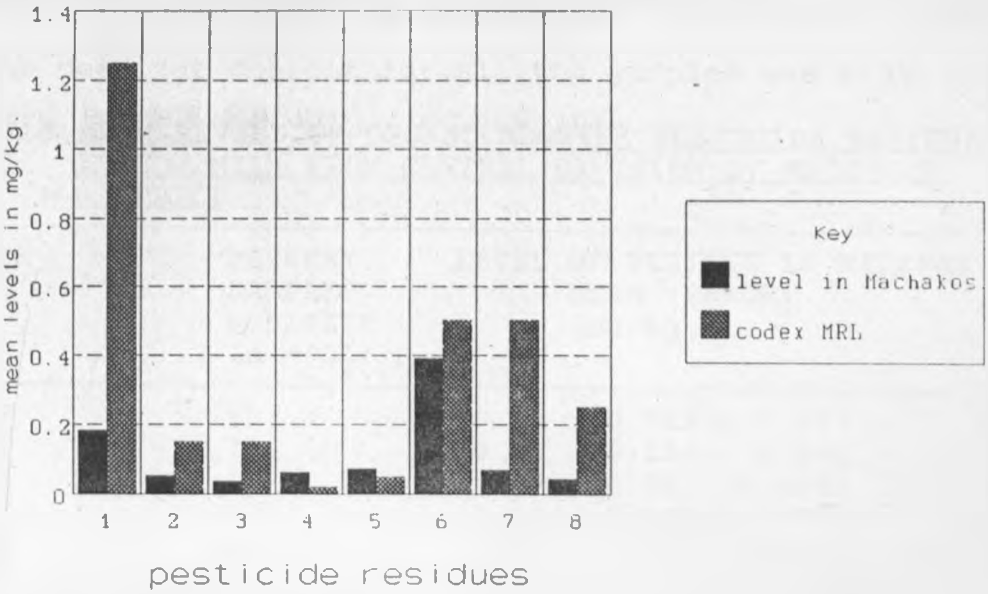


Fig 4 OC PESTICIDE RESIDUES IN HUMAN MILK COMPARED TO CODEX LIMITS
LEVELS IN mg/kg

<u>Pesticide residue key</u>.
1 - total DDT
2 - total heptachlor
3 - total dieldrin/aldrin
4 - endrin
5 - oxychlordane/chlordane
6 - beta endosulfan
7 - alpha endosulfan
8 - lindane

4.2.2 <u>Organochlorine pesticides in cow milk.</u>

The main pesticide residues reported were o,p'-DDT (71%*[2]), (SUM DDT (100%*), heptachlor epoxide (71%*), τ-HCH (24%*), p,p'-DDD (28%*), o.p'-DDE (19%*) and oxychlordane (24%*). Great individual variations in the mean levels of the various residues was indicated (Table 4.10). There was a higher excretion of o,p'-DDT than the other DDT metabolites as was the case with τ-HCH and α-HCH compared to other HCH isomers.

[2]* - % positive samples.

Notable was the absence of the persistent β-HCH isomer in cow milk. The mean fat content for all the samples was 4.8%.

TABLE 4.10 MEAN LEVELS OF ORGANOCHLORINE PESTICIDE RESIDUES IN COW MILK FROM CENTRAL DIVISION OF MACHAKOS DISTRICT.

PESTICIDE RESIDUE	PERCENT SAMPLES POSITIVE (n = 21)	LEVEL OF RESIDUE IN MILKFAT MEAN (RANGE) mg/kg
p,p'-DDT	10	0.03 (0.029 - 0.03)
o,p'-DDT	71	0.41 (0.159 - 2.28)
p,p'-DDE	10	0.09 (0.06 - 0.134)
o,p'-DDE	19	0.04 (0.006 - 0.09)
p,p'-DDD	29	0.05 (0.03- 0.097)
o,p'-DDD	5	0.13
SUM DDT	100	0.162 (0.006 - 0.228)
ALDRIN	14	0.07 (0.008 - 0.183)
DIELDRIN	10	0.05 (0.016 - 0.087)
HEPTACHLOR	19	0.02 (0.008 - 0.047)
HEPTACHLOR EPOXIDE	67	0.02 (0.005 - 0.033)
OXYCHLORDANE	24	0.02 (0.007 - 0.032)
CHLORDANE	14	0.04 (0.019 - 0.080)
α-HCH	14	0.03 (0.013 - 0.052)
τ-HCH	24	0.03 (0.011 - 0.062)
E-HCH	5	0.09
TOTAL HCH	42	0.04 (0.01 - 0.086)
HCB	10	0.013 (0.007 - 0.019)
α-ENDOSULFAN	10	0.091 (0.084 - 0.099)

There were no significant difference ($p > 0.05$) in the incidence and mean levels of the various organochlorine pesticide residues between the two regions (Table 4.11 and Fig. 5). However, a significant difference ($p < 0.05$) was found between the mean levels of p,p'-DDD from low potential production zone as compared to the high potential production zone, with the higher mean value being found in samples from the former zone.

TABLE 4.11 MEAN LEVELS OF ORGANOCHLORINE PESTICIDE RESIDUES IN COW MILK BY REGIONS OF MACHAKOS CENTRAL DIVISION, MACHAKOS DISTRICT.

PESTICIDE RESIDUE	LOW PRODUCTION REGION (MUPUTI)		HIGH PRODUCTION REGION (MUMBUNI AND MUTITUNI)		LOW VS HIGH AREA
	%+VE (N=10)	LEVEL OF RESIDUE IN MILKFAT-(mg/kg) mean (range)	%+VE (N=11)	LEVEL OF RESIDUE IN MILKFAT- (mg/kg) mean (range)	
p,p'-DDD	10	0.097	45	0.037(0.031-0.048)	S
SUM DDT	71	0.206(0.006-0.228	71	0.119(0.01-0.299)	NS
TOTAL HEPTACHLOR	80	0.012(0.005-0.029)	91	0.02(0.005-0.046)	NS
SUM HCH	80	0.038(0.01-0.086)	10	0.023	NS
TOTAL ALDRIN	40	0.073(0.008-0.183)	9	0.019	NS
OXYCHLOR-DANE	30	0.016(0.007-0.031)	18	0.018(0.013-0.022)	NS
ENDOSULFAN	10	0.099	9	0.083	

Key.
S (p<0.05) significant difference.
NS (p>0.050 - no significant difference.

The mean levels of all the pesticide residues were generally lower than reported in other studies (Table 2.8). The levels were also lower than the maximum levels recommended by Codex Alimentarius Commission (FAO & WHO, 1986). However, the mean level for total chlordane at 0.06 mg/kg was slighly higher than the maximum allowed by the Codex Alimentarius Commission (0.05 mg/kg) (Fig. 6).

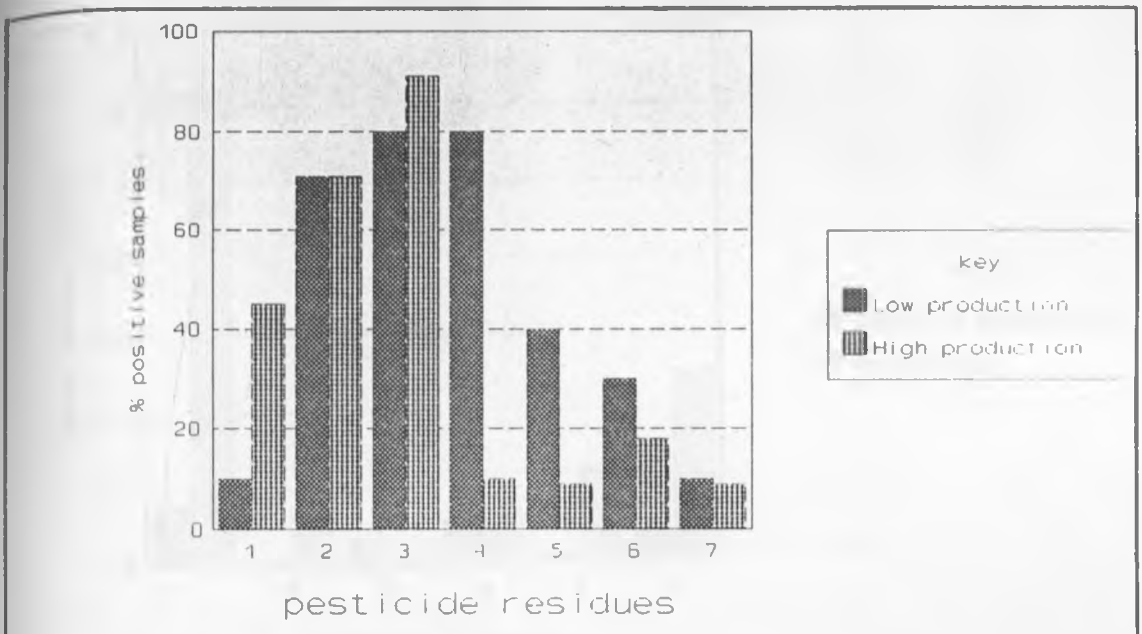


Fig. 5 INCIDENCE OF OC RESIDUES IN COW'S MILK BY AGRI ZONES
MACHAKOS CENTRAL DIVISION, 1991

<u>Pesticide residue key.</u>

1 - p,p'-DDD
2 - total DDT
3 - total heptachlor
4 - total HCH
5 - total aldrin
6 - oxychlordane
7 - endosulfan

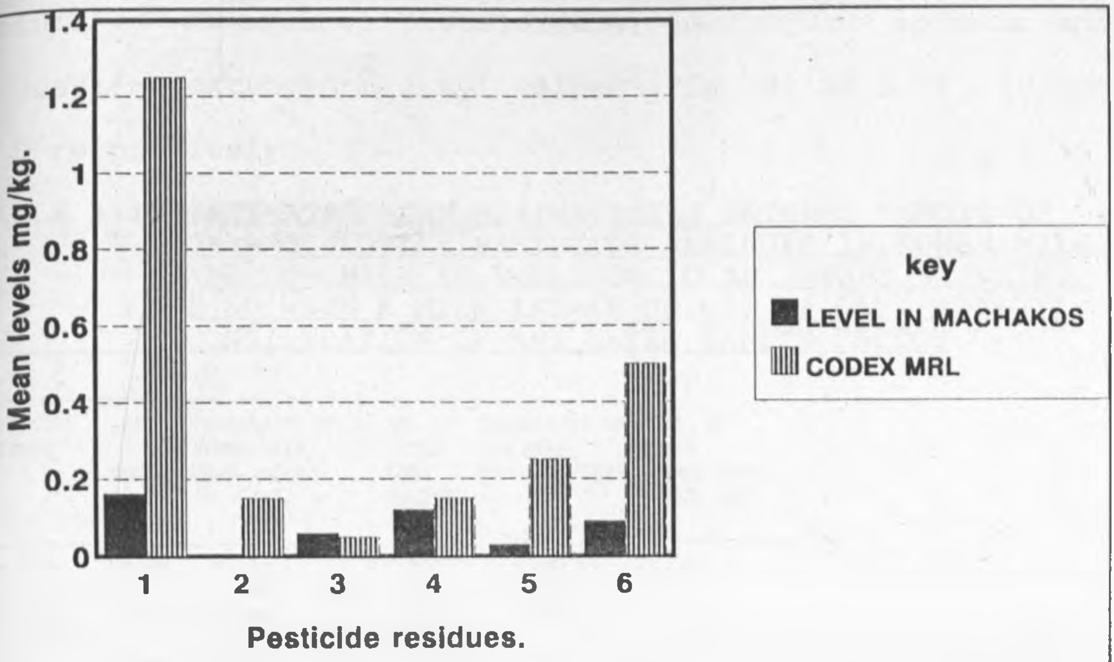


Fig. 6 OC PESTICIDE RESIDUES IN COW MILK COMPARED TO CODEX LIMITS. LEVELS IN mg/kg.

Pesticide residue Key.
1 - total DDT
2 - total heptachlor
3 - total chlordane
4 - total aldrin
5 - lindane
6 - endosulfan

4.2.3 "Dietary intake of some organochlorine pesticide residues by infants.

Table 4.12 gives some calculated Estimated Maximum Daily Intakes (EMDI) of various organochlorine residues by children in Machakos central division. The values of EMDI calculated for human milk of endrin, chlordane, oxychlordane, heptachlor epoxide, lindane and endosulfan were above the acceptable daily intakes (Fig. 7) of these pesticide residues by 40, 4, 9, 70, 1.6, and 6.3 times respectively. In cow milk the EMDI values of chlordane, oxychlordane, heptachlor epoxide and endosulfan exceeded the ADI values (Fig. 8) by 5, 6, 30 and 1.5 respectively.

TABLE 4.12 ESTIMATED MEAN MAXIMA DAILY INTAKES (EMDI) OF ORGANOCHLORINE PESTICIDE RESIDUES IN HUMAN MILK AND COW MILK IN RELATION TO AN INFANT WEIGHING 5 KG WITH A MILK INTAKE OF 655 g/ DAY, WITHOUT CONSIDERATION OF ANY EXTRA SAFETY FACTOR.

PESTICIDE RESIDUE	ADI* mg/kg	CALCULATED EMDI HUMAN MILK MEAN (mg/kg) (n = 147)	NO. OF TIMES EMDI EXCEEDS ADI	CALCULATED EMDI COW MILK MEAN (mg/kg) (n = 21)	NO. OF TIMES EMDI EXCEEDS ADI
SUM DDT	0.02	0.024	0	0.021	0
ALDRIN/	0.01	0.011	0	0.009	-
DIELDRIN	0.01	0.011	0	0.007	-
HEPTACHLOR EPOXIDE	0.0001[a]	0.007	70	0.003	30
OXYCHLOR-DANE	0.001T	0.009	9	0.006	6
LINDANE	0.008B	0.013	1.6	0.004	-
CHLORDANE	0.001T	0.004	4	0.005	5
TOTAL HCH		0.015		0.005	
HCB	0.6	0.042	-	0.002	-
ENDRIN	0.0002	0.008	40	NOT DETECTED	-
ENDOSULFAN	0.008D	0.051	6.3	0.01	1.5

Key.
* FAO/WHO, 1986.
ADI - ACCEPTABLE DAILY INTAKE OF PESTICIDE RESIDUE.
EMDI - ESTIMATED MAXIMUM DAILY INTAKE OF RESIDUE.
T - TEMPORARY VALUE.
B - WHO, 1992b LINDANE.
D- TOTAL ALPHA, BETA ENDOSULFAN AND ENDOSULFAN SULPHATE- WHO, 1984c.
a - PESTICIDE RESIDUES IN FOOD, FAO, 1991.

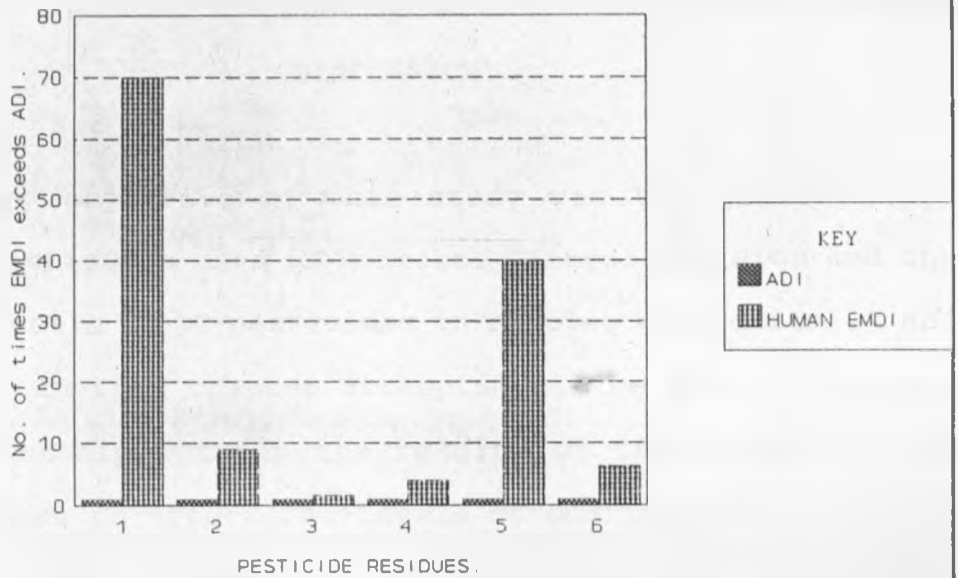


Fig. 7 ESTIMATED INTAKE OF OC'S IN HUMAN MILK COMPARED WITH ADI
MACHAKOS CENTRAL DIVISION, 1991

Pesticide residue Key.

1 - Heptachlor epoxide	3 - Lindane	5 - Endrin
2 - Oxychlordane	4 - Chlordane	6 - Endosulfan

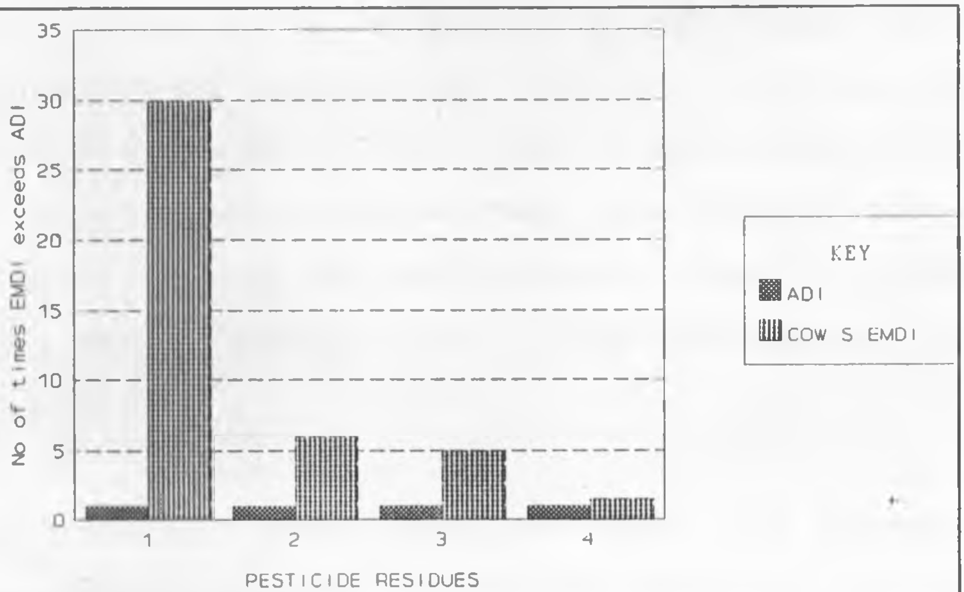


Fig. 8 ESTIMATED INTAKE OF OC'S IN COW'S MILK COMPARED TO ADI
MACHAKOS CENTRAL DIVISION, 1991.

<u>Pesticide residue key</u>.
1 - Heptachlor epoxide 2 - Oxychlordane 3 - Chlordane 4 - Endosulfan

CHAPTER 5.

DISCUSSION.

The primary objective of this study was to determine the types of pesticides used in Machakos Central Division and the residual levels of OC pesticides in samples of breastmilk and cow milk. In this chapter attempts will be made to discuss the relationship between the results of this study to the factors known to affect the levels of OCs in milk.

5.1 TYPES OF PESTICIDES CURRENTLY BEING USED IN THE STUDY AREA.

Data from this study show that, most of the pesticides used were insecticides as is evidenced by the sales from K.G.G.C.U. stores in Machakos town (Table 4.2) and also the varieties used (Table 4.3). Other types of pesticides being used were fungicides (mainly on coffee), herbicides and those used for grain storage and seed dressing. Most of these pesticides were organochlorines, organophosphates or carbamates.

High volumes of the organochlorine pesticides like lindane, BHC (dawa ya mboga), thiodan and agrocide were also sold in the K.G.G.C.U. stores (Table 4.2). Fifty one percent (51%) of the respondants in the study (mothers) used these pesticides in households and 51% on crops/fruit trees in the field. The pesticides reportly used on livestock by 64% of the mothers mainly as dip were organophosphates.

The discussion which follows below will try to relate this section to the levels and types of pesticides detected in milk.

5.2 TYPES OF PESTICIDE RESIDUES FOUND IN BREASTMILK FROM MACHAKOS CENTRAL DIVISION.

5.2.1 "The DDT group".

The mean levels of all the organochlorine pesticide residues reported were generally low and are comparable to levels reported in other countries (Table 2.2). However, compared to the one study done in Kenya (Kanja, 1988), the levels are lower. There was no reported commercial application of DDT in the study area at the time of the study and the last stocks of DDT were reported to have been sold out in K.G.G.C.U. stores in Machakos town in 1988 because DDT was restricted for use in public health. This might explain the relatively low levels of the pesticide in the milk from the study area and the slightly higher levels of this pesticide in older women (22-30 years) as compared to the younger ones. This also, explains the lack of any significant difference ($p < 0.05$) in the mean levels of the DDT group of pesticides between the different agricultural zones (low versus high production), between the different types of pesticide usage on crops/fruit trees in the field, between the two age groups and the parity of the mothers.

However, there was a significant difference ($p < 0.05$) between the mean levels of sum DDT in milkfat from mothers in households treated with pesticides (0.41 mg/kg) as compared to those from households not treated with pesticides(0.13 mg/kg). The active ingredients of the pesticides used in the households w\as not reported by the mothers. De Campos and Olzyna-Marzys (1979) reported that the highest levels of DDT in Guatemalan human milk was due to indoor spraying of DDT for malaria control. The highest mean level of sum DDT in Kenya was reported in Rusinga island (18.73 mg/kg) also due to malaria eradication. It is also very possible that, the higher levels of sum DDT in this study in households treated with pesticides, might have been due to the use of DDT based pesticides for mosquito control.

High use of home and garden pesticides (including non persistent pesticides) has in some investigations been related to increased levels of sum DDT in breastmilk (Jensen, 1983). This could have been the source of high levels of sum DDT found in this study as farmers in the study area were using other pesticides, especially organophosphates, acaricides and on horticultural crops.

The mean levels in human milk of p,p'-DDT or o,p'-DDT were higher than the p,p'-DDE or o,p'-DDE. Contamination of human milk with DDT as compared to DDE reflects relatively recent exposure of the mother to DDT, whereas contamination with DDE reflects either earlier exposure to DDT which has then been metabolized to DDE, or exposure to DDE through consumption of foods of animal origin. The main foods of animal origin consumed by the mothers in the study area were milk and eggs. The higher excretion of o,p'-DDT as compared to other DDT metabolites reflects a possible use of technical DDT in the study area. Technical DDT is more readily excreted and less readily stored than p,p'-DDT because it contains 15-20% o,p'-DDT (WHO, 1979).

5.2.2 "The HCH group".

Lindane (τ-HCH), some HCH acaricides and BHC (dawa ya mboga) were the insecticides reported in use at Machakos central division. Beta-HCH had the highest incidences and concentration in all the samples analyzed. Beta-HCH has a 10-30 times higher ability to accumulate in fat tissues and milk than τ-HCH (Jensen, 1983). Beta-HCH is therefore, the isomer usually found in the highest concentrations in milk. The ratios of the different HCH isomers in breastmilk change from the start of foodchains until excretion in human milkfat, resulting in the more persistent β-HCH being the predominant isomer.

There was a significant difference ($p < 0.05$) in the mean levels of β-HCH and τ-HCH between the low potential production zone and the high potential production zone (Table 4.5). The incidence of these two residues was higher in the high potential production zone, while the mean levels were higher in the low potential production zone.

All the different HCH isomers were detected in human milk in Machakos central division. This indicates that technical grade HCH was used as an insecticide in this area. Technical grade HCH consists of 65-70% α-HCH, 7-10% β-HCH, 14-15% τ-HCH (lindane), 7% δ-HCH, 1-2% E-HCH. Alpha HCH and β-HCH have been used in mixtures with τ-HCH (lindane) as HCH in agriculture and in wood protection. The mothers reported the use of BHC (dawa ya mboga), agrocide, lindane (τ-HCH) and some HCH acaricides in the area.

The mean levels of all the HCH isomers were below the Codex Recommended Maximum Residue Limits. The mean level of β-HCH of 0.189 mg/kg was slightly higher than the highest level reported by Kanja (1988) for Embu (0.164 mg/kg). The levels in this study were, however, comparable to the levels reported for the rest of Kenya (Kanja, 1988) and for other countries (Jensen, 1983; WHO, 1992a and WHO, 1992b). In both Embu and Machakos, the HCH group of pesticides was easily available to the farmers in the local outlets.

5.2.3. "Aldrin, dieldrin and endrin".

In this study, aldrin, dieldrin and endrin were detected in 6%, 1%, and 2% of the samples, respectively. Aldrin is quickly converted to dieldrin in the human body (Hayes, 1982), hence the presence of these residues in human milk indicates exposure to either aldrin or dieldrin or both. More samples contained aldrin (6%) than dieldrin (1%). However, this is expected in areas where there is intense or medium horticulture, since aldrin is applied on certified seedlings for planting. The seedlings are often handled with bare hands during planting and therefore, the pesticide can easily be absorbed through the skin or inhaled. Kanja (1988) also reported a higher incidence of aldrin than dieldrin in Kenya. Higher levels of these pesticides have also been associated with house and garden use of aldrin/dieldrin in a few regions (WHO, 1989b).

The mean levels of aldrin and dieldrin were below the Codex Recommended Maximum Residue Limits, while the mean levels of endrin were above the codex recommended residue limit. However, the mean levels of these residues detected in this study were lower than the values reported by Kanja (1988) and are comparable to values reported in other countries (Jensen, 1983; WHO, 1989b). Endrin is found as an impurity in technical dieldrin.

5.2.4 "Heptachlor, heptachlor epoxide, chlordane and oxychlordane".

The incidence and mean levels of this group of pesticide residues seem to be higher in the high potential production zone as compared to the low potential production zone. The pesticides are mainly used in termite control and their use has been banned in Kenya. Heptachlor epoxide and oxychlordane are very persistent epoxy metabolites of heptachlor and chlordane. Heptachlor epoxide was reported in 63% and oxychlordane in 5% of the samples analyzed.

The mean levels of oxychlordane/chlordane in this study were below the Recommended Codex Maximum Residue Limits and were comparable to levels reported in other countries (Jensen, 1983; WHO, 1984a and WHO, 1984b).

Results of this study show that there was significant difference ($p < 0.05$) between the mean levels of oxychlordane in mother's milk fat from the area where pesticides were applied on crops/fruit trees in the field and those not applied, with levels being lower in the former than the latter. Chlordane has been used as a broad spectrum contact insecticide, mainly for non-agricultural crops and on animals (WHO, 1984a). Mothers reported the use of chlordane (ant killer), although the volumes used were not ascertained. The presence of residues of oxychlordane, transnonachlor and heptachlor epoxide has been associated with chlordane exposure (WHO, 1984a). Technical chlordane is a mixture of insecticidal components α - and τ - chlordane, heptachlor and nona-achlor. Therefore, the higher incidence of heptachlor epoxide (63%) in this study could be attributed to the use of technical chlordane by the mothers.

5.2.5 "Other organochlorine pesticide residues".

The incidences of other organochlorine pesticide residues were low, indicating that they are not a serious pollution problem in the study area.

(a) Endosulfan. Alpha and beta endosulfan were detected in 9% and 2% respectively, of the samples analyzed. Mothers reported using endosulfan (thiodan) in their farms. The pesticide was also reported sold in the area in K.G.G.C.U. stores. Technical grade endosulfan was probably used as it consists of alpha and beta isomers in approximate ratios of 70:30. The results indicate that there were more samples with the alpha isomer than the beta isomer. Endosulfan does not accumulate in food chains and is excreted from the body rapidly (WHO, 1984c). Therefore, from the results it can be concluded that endosulfan was still being used in the study area. No reports of endosulfan in breastmilk have appeared in literature (WHO, 1984c). However, since it is used as a wood preservative and garden pesticide in some countries including

Kenya and at the study area, direct exposure to mothers, infants and children remains a possibility (WHO, 1984c). The mean levels reported in this study are well below the FAO/WHO tolerance levels.

(b) Hexachlorobenzene (HCB). This pesticide was found in only one sample in levels below the Recommended Extraneous Residue Limit of 0.5 mg/kg in milkfat (FAO/WHO, 1982). The presence of HCB could be due to exposure to technical grade HCH. When urine of occupationally exposed workers (apparently to technical-grade HCH in manufacturing processes) was analysed, it was found to contain traces of hexachlorobenzene, apart from **α**-, **β** -, **τ** - and **δ** - HCH, (WHO, 1992a and WHO, 1992b). Hexachlorobenzene was also identified in the blood of the workers (WHO, 1992a and WHO, 1992b).

(c) Methoxychlor. This pesticide residue was detected in only one sample. It is a DDT related insecticide.

5.2.6 "Factors affecting the organochlorine content of breastmilk".

As already mentioned (section 2.3.1), a number of factors have been reported to influence the accumulation of organochlorine pesticide residues in human milk. These factors include maternal age and residence, length of lactation period, mother's diet, number of children (parity), smoking, home use of pesticide, occupation and culture (Jensen, 1983 & WHO, 1979).

5.2.6.1 **Maternal age**.

Although, the narrow age ranges (18-30 years) of the mothers sampled in this study restrict the possibility of indepth investigation of the influence of age on organochlorine residue levels in breastmilk, the trend suggests that there is a positive age corelation. Data in this study show that, milk from older mothers (45%) contained higher levels of oxychlordane, α-endosulfan, sum DDT, total heptachlor/heptachlor epoxide than milk from younger mothers (55%). Milk from younger mothers on the other hand contained higher levels of total HCH and aldrin/ dieldrin than milk from older ones. These findings are in agreement with other studies. Rogan et al. (1986) and Hashemy-Tonkabony et al. (1977) reported higher excretion of DDT and its metabolites by older mothers than younger ones in North Carolina (U.S.A) and Iran respectively. Kanja (1988) demonstrated a positive age relationship with the DDT group and reported no residues of alpha HCH, dieldrin and heptachlor in older mothers as compared to younger ones in Kenya.

Only weak correlations have been reported between the mothers' age and the levels of organochlorine pesticide residues (OCs) in breastmilk (Slorach & Vaz, 1983). The underlying hypothesis seems to be that, older mothers have a longer time to accumulate these lipophilic residues from the environment and through food than younger mothers. Therefore, the higher levels of the residues in older mothers is expected.

In this study, the breastmilk was sampled from mothers who had lived in the study area for over five years to minimize differences from environmental exposure of the subjects. Ninety six percent (96%) of the mothers had lived in the study area for over 17 years, therefore a constant rise in the residue levels with age was expected due to the continued exposure. However, no clear consensus has been reached regarding the factor of maternal age as the divergences might be explained by the existence of compounding factors, such as number of previous childbirths. Furthermore, if the OC levels are expressed on fat weight basis as in this study, the age association may not be very significant because of the variation of fat with the time of sampling (Jensen,1983).

5.2.6.2 **Length of lactation and parity.**

Noren (1983b) reported that when expressed on a fat basis, the levels of p,p'- DDT, p,p'-DDE and PCBs in human milk decrease very little during lactation periods of upto six months. Thus to minimize any differences due to the different times of sampling (post-partum) in this study, milk was sampled at 1 week to 4 months postpartum. Hence this factor was unlikely to have any major effect on the comparability of the data collected from other studies.

In this study 59% of the mothers were breastfeeding their first child, while 41% were breastfeeding their second child. The mean levels of sum DDT, total heptachlor and oxychlordane in human milkfat were higher in milk from mothers breastfeeding their first child as compared to those breastfeeding their second (Table 4.9). However, there was no significant difference ($p > 0.05$) between the mean pesticide residue levels of the various organochlorine pesticides in milk and the mean fat content of milk from mothers breastfeeding their first child and those breastfeeding their second. This is in agreement with the findings from studies with individual mothers by Yakushiji et al. (1979) and Noren (1983a) which showed that the levels of p,p'-DDT, p,p'-DDE and PCBs are higher in human milk from mothers nursing their first child than during subsequent nursing periods. The results of this study are also in agreement with studies where there was no continued exposure to the OCs. The levels of pesticide residues in these studies decreased with the number of previous deliveries by the mother (Kanja, 1988; Slorach and Vaz, 1983). However, in cases where there was continued exposure, the above was not true, for oxychlordane, chlordane, endosulfan aldrin and the HCH group of pesticides.

The results of this study indicate that the mothers were still being exposed to lindane (τ-HCH) and HCH (dawa ya mboga) in the farms. This could explain why there was little difference in the mean levels of these pesticides in relation to the parity of the mothers.

The mean fat content of the milk from mother's breastfeeding their first child was higher than in mothers breastfeeding their second child. Jensen (1983) reported that the parity of the mother influences the level of fat in the milk, with mothers breastfeeding their first child having higher milk fat content than multipara mothers. Since OCs are liphophilic, the higher residue levels in mothers breastfeeding their first child might be explained by the higher fat content.

5.2.6.3 Residence/agricultural activity.

In general, mothers from urban areas have higher sum DDT levels in their milk than mothers from rural areas as long as DDT has not been commercially applied in the area (Jensen, 1983). All the subjects in this study were living in a rural area and no corresponding study was done in an urban area to ascertain whether the above findings are true.

On the other hand, the data from this study indicates that, agricultural activities had an influence on the levels and types of pesticides detected in Milk. The occurence of pesticide residues was more prevalent in breastmilk from mothers in the high potential production zone than those from the low potential production zone. This could probably be due to the fact that, in the high potential production zone where there is generally high agricultural activity, there is higher usage and application of pesticides than in the low potential production zone. However, there were no significant differences between the mean levels of the various OCs in breastmilk from the two agricultural production zones, probably due to the fact that the diet was similar for all the mothers. The diet determines the fat content of milk (Noren, 1983c; Kanja, 1988) in which these liphophilic compounds accumulate.

5.2.6.4 Other factors.

In this study, all mothers were from one ethnic group the Akamba, and 84% of them were housewives. Of these mothers also, 51 % had some primary school level education while 47% had secondary level education. None of the mothers smoked or used tobacco in any form. Therefore, the factors smoking, occupation and culture were not among the known factors that could have influenced the levels of pesticide residues found in their breastmilk. Similarly, although 99% of the mothers were lactovegetarians, very little (quantitatively) milk is actually consumed. In addition, the diets consumed were very much the same. Therefore, correlation between dietary habits and OC levels was not meaningful.

5.3 The types of pesticide residues found in cow milk from machakos central division.

The incidence and mean levels of the pesticide residues detected in cow milk in this study were generally slightly higher than those reported by Maitho (1977) and Kanja (1988) in Central Province, Nakuru, Turkana and Rusinga island in Kenya. However, the mean levels of all the pesticide residues were generally comparable to those reported in other studies (Maitho 1977; Kanja 1988; WHO, 1992a and WHO, 1992b). Low levels of organochlorine pesticides were also reported in non-fatty foodstuffs and feedstuffs by Muinamia (1976). Maitho (1977) reported p,p-DDT (6%), P.P-DDE (47%), lindane (18%), aldrin(23%) and dieldrin (6%) in the 25 samples analyzed.

The mean levels of the different organochlorine pesticides were lower in cow milk than in breastmilk. It has been known for a long time that human milk may contain higher concentrations of DDT (WHO, 1979), heptachlor (WHO, 1984b) and other organochlorine pesticide residues than cow milk in the same country, because human beings are placed at a higher level of most foodchains compared to cattle.

Higher mean levels of o,p'-DDT were found in cow milk as compared to other DDT metabolites. This indicates (as mentioned for breastmilk) that technical DDT may have been used in the area since it is more readily excreted and less readily stored than p,p'-DDT, as it contains 15-20% o,p'-DDT.

All the organochlorine pesticide residues found in breastmilk were also found in cow milk. This could indicate that these residues might be circulating in the environment and in foodchains in the area, hence the common exposure to both man and cattle.

The mean levels of chlordane in cow milk were higher than the Codex Recommended Maximum Residue Limit. Chlordane was reportly being used in the study area. This explains the high incidence of heptachlor epoxide (71%), oxychlordane (24%), chlordane (14%) and heptachlor (19%) in cow milk.

There were higher incidences of τ-HCH as compared to all the other HCH isomers. The notable absence of the more persistent β-HCH might have been due to exposure only to technical HCH, which contains 65-70% α-HCH, 14-15% τ-HCH, 7-10% β-HCH, 7% δ-HCH and 1 - 2% E-HCH. The presence of the HCH-group of pesticide residues in cow milk was due to the continued use of these pesticides in the area, as the pesticides were readily available locally in the study area. Also, the fact that some organochlorine acaricides like toxaphene are still being used in the area might explain the presence of the HCH group residues in cow milk. Keating and Odour (1982) reported that the source of the HCH group of pesticides in butter was due to the use of lindane and BHC dusts in the storage of cereals.

The mean levels of endosulfan reported in this study were below the FAO/WHO tolerance levels (WHO, 1984c). Endosulfan has been detected in cow milk from tobacco growing areas at levels of upto 0.01 mg/litre. It has also been reported in milk of cow's fed on forage crops containing 0.41, 0.70 or 2.25 mg/kg for 21 days (WHO, 1984c).

5.4 ASSESSMENT OF DIETARY INTAKE OF THE VARIOUS ORGANOCHLORINE PESTICIDE RESIDUES.

Assessment of the amount of pesticide to which man can be exposed to daily for a lifetime without injury, is the primary aim of toxicological investigations. These investigations are used to derive Acceptable Daily Intakes (ADI), the maximum daily dosage which during the entire lifetime, appears to be without any appreciable risk on the basis of all the facts known at the time (WHO, 1990).

Dietary intake studies have been used to check whether the acceptance of Codex Maximum Residue Limits (MRLs) could result in situations which the ADI of a pesticide would be exceeded. In cases where such studies are not feasible or where the pesticide has not been in use for long, it is necessary to predict the pesticide residue intake on the basis of the available data (WHO, 1989c).

Out of the several possible indices of food consumption, average daily consumption has been proven accurate and is commonly used (WHO, 1989c). Average daily food consumption values are used in predicting residue intakes to facilitate valid comparison of the estimated maximum daily intake to ADI.

In this study, the average daily milk consumption (by infants) values were used in predicting residue intakes to facilitate valid comparison of the estimated maximum daily intake (EMDI) to ADI for infants. The levels of OCs in milk given in the preceding sections cannot make much practical sense, unless associated with how much is really consumed by the child.

A study in Machakos reported that breastfeeding was unsupplemented for the first one to four months, with a median time of about two months, among a sample of 183 mother-child pairs. It was also found that prolonged breastfeeding is commonly practiced, with a mean duration of

14.7 (+ or - 6.8) months. The same study also reported that children receive most of their nourishment from breastmilk for 5.6 (+ or - 3.1 months). Among most Akamba mothers cow milk was first given as a supplement in the third month, usually by the bottle (INCS, 1983). Therefore, in Machakos a child of 0 - 6 months derives all of its nutrients from breastmilk with some little supplementation with cow milk. Calculation of the possible exposure to infants of this age limit 0 - 6 months to pesticides is therefore based on this information.

Among Akamba mothers breastfeeding was mainly on demand day and night, with feeds nearly every two hours during the first two to three months. At about 6 months of age, infants were breastfed more on the mothers schedule, with about 6 to 7 feeds per 24 hours. The amount of milk delivered per feed was similar at all ages 0 to 17 months i.e 60 millitres (INCS, 1983). Therefore, it can be assumed that at the age of 0 - 6 months the children in the study area consumed 720 millitres of breastmilk with probably very little supplementation with cow milk.

Using the above information and the study data, the EMDI data calculated for human milk of heptachlor epoxide, endrin, oxychlordane, chlordane, endosulfan and lindane were above the acceptable daily intakes of these pesticide residues by

70, 40, 9, 4, 6.3 and 1.6 times respectively (Table 4.12). This data suggests that the infants in the study area are at a higher risk of being exposed to high levels of these residues, especially heptachlor epoxide and endrin, simply by consuming breastmilk. In the case of cow milk the EMDI values of heptachlor epoxide, oxychlordane, chlordane and endosulfan exceeded the ADI values by 30, 6, 5 and 1.5 respectively. Whenever cow milk was given to 0 - 3 months old Akamba infants in Machakos District, it was diluted with a little water, while older infants received undiluted milk (INCS, 1983). Results from this study show that the infants in the study area are at a lower risk of being exposed to high levels of these residues by consuming cow milk, since in most cases it was given as a supplement to breastmilk (INCS, 1983) coupled with its lower pollution load.

As such, the pesticide residues posing the greatest danger to the infants in the study area are heptachlor epoxide and endrin. Other pesticides whose dietary intake was higher than the ADI were endrin, chlordane (oxychlordane), endosulfan and lindane. However, the Estimated Maximum Daily Intakes of the various pesticides are not as high as those shown in other studies (Kanja, 1988 and Jensen, 1983).

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS.

6.1 CONCLUSIONS.

This study has confirmed that organochlorine pesticides had been used and some are still being used extensively for agriculture and veterinary purposes in Machakos central division. It shows that the regulatory (and/or monitoring) measures by the bodies mentioned in section 2.2.1 are not foolproof, since some pesticides which have been banned or restricted for use in Kenya, were found being used. For example, the use of technical grade HCH products containing high concentrations of α- and β-HCH is not justified, since these isomers have no insecticidal action. This shows laxity in the enforcement of the by-laws governing pesticide usage. The results suggest that, organochlorine pesticide residues are circulating in the environment and in foodchains and as such there is a continous potential for human exposure. However, this exposure is low in Machakos central division and is expected to decrease slowly in the coming years, as there were no stocks of some of the pesticides like DDT, aldrin and dieldrin (Table 4.2) in the stores in Machakos central division.

The data from this study demonstrates that all human and cow milk samples from both the low and high agricultural production areas were contaminated with organochlorine pesticides with the incidence being highest in the high agricultural production area. The DDT group and heptachlor/heptachlor epoxide were the major contaminants, though concentrations of their residues in milk from this area are generally low as compared to reports from other studies (Jensen, 1983 ; Kanja, 1988).

The pesticides detected in mothers milk in the highest frequency were p,p' - DDE, heptachlor epoxide, o.p' - DDT, p,p' - DDT, heptachlor and beta HCH. There was an association between the usage of pesticides in the study area (on agriculture or veterinary or households) and the level of the identified pesticides in milk.

Calculation on infant dietary intake of OCs shows that heptachlor/heptachlor epoxide, endrin, chlordane/oxychlordane, endosulfan and lindane in both breastmilk and cow milk are consumed at levels exceeding the ADI by several fold. This is further evidence that infants are exposed to the dangers of pesticide residues through breastmilk.

6.2 RECOMMENDATIONS.

The infants in Machakos central division are exposed to contaminated milk but this is no reason for not promoting the use of breastfeeding. However, every possible effort should be made to decrease dietary and all other types of exposure. Since, use of OCs is banned or restricted (Table 2.2) for use in the country, the bodies concerned with implementation of these regulations, need to be more stringent in order to minimize all types of environmental (and hence dietary) exposure.

There is need to take steps towards reducing the environmental contamination by pesticide residues in general by practicing better agricultural and veterinary practices to avoid the indiscriminate use of pesticides in agriculture not only in Machakos central division, but countrywide.

Since milk from the study area was contaminated with pesticide residues, monitoring programmes should be initiated to ensure that the all the avenues of exposure are properly documented. This study suggests that human and cow milk are suitable indicators for the biological monitoring of environmental contamination by organochlorine pesticides in Machakos Central Division.

Since some organochlorine pesticides are still being used for some restricted programmes like public health (e.g DDT for mosquito control programmes) due to their low costs, more research is needed to establish their effects on the general population in the country.

Monitoring the daily intake of the general population and levels of OCs in breastmilk and cow's from urban areas should also be done to facilitate comparisons with data from other rural areas.

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Appendix 1

Questionaires.

Questionaire 1.

INFORMED CONSENT FORM.

NAME OF MOTHER ....

HOSPITAL I.D. NUMBER ....

I .... hereby consent to participate in the study on levels of organochlorine pesticide residues in breastmilk and cow milk, which has been explained to me by researcher/health staff.

SIGNED .... DATE:...:...:...:
PARTICIPANT (MOTHER).

SIGNED .... DATE:...:...:...:
HEALTH STAFF/RESEARCHER.

QUESTIONAIRE 1 - TO BE COMPLETED DURING THE COLLECTION OF HUMAN MILK SAMPLES IN CENTRAL DIVISION OF MACHAKOS DISTRICT, 1991.

Date .... Questionaire Number ....

Location .... Area ....

Name of interviewer: ....

Section A:

1. Name of mother ....

2. What is your marital status 1= Single _____
   2= Married : :
   3= Divorced :____:

3. What is your age in ....years.

4. What is your educational level? 1= Primary _____
   2= Secondary : :
   3= Above Secondary:___:

5. (If married) What is your educational level of husband?
   1= Primary _____
   2= Secondary : :
   3= Above Secondary:___:

6. How many years have you lived in study area ..... years.

7. What is your occupation? ....

8. (If married) what is the occupation of your husband?

   ....

9. When was your youngest child born? ...:.........:.....:
   Day month year

10. Is it your first or second child? ....

11. Which are the main crops in the area you are living?
    1= Cereals and grains e.g Maize__, Beans__, Pigeon peas__, peas__, etc.

    2= Fresh vegetables e.g. Kale__, Cabbage__, Tomatoes__, Aubergines__, French beans__, etc.

    3= Fruits e.g Oranges__, Lemons__, Plums__, Passion__, Pawpaw__, Guava__, Avocado__, Apples__, etc.

    4= root crops e.g. Cassava__, Arrow roots__, Sweet potatoes__, Potatoes __, Potatoes (English)__, etc.

    5= Cash crops e.g. Coffee__, Cotton__, sunflower__, etc.

12. Do you use (or have you used) tobacco (smoked, chewed, or as snuff)?

1= Yes :___:
2= No :___:

(If yes, ask)

(a) How many times do you smoke per day? ....

(b) For how long have you been smoking? (months or years)

Section B: Pesticides used in the household.

1. Has your house been treated with pesticides during the last year? (e.g. against mosquito, flies, cockroaches, bedbugs, lice etc.)

:___:
:___:

(if yes, ask)
(a) By Whom?

1= mother :___:
2= others :___:

(b) How do you apply?
1= spray :___:
2= springle :___:
3= other specify.................

(c) How often per week? ....

(d) Against what? ....

(e) What is the name of the pesticide? ....

2. (a) Do you use pesticides for stored foods?
1= Yes :___:
2= No :___:

(b) (if yes ask)
Which food and what pesticides(s)

Food	Source of Pesticide	Type of pesticide
....	....	....
....	....	....
....	....	....

3. Do you use pesticides for any crops/fruits trees in the field?

:___:
:___:

(if yes, ask) what pesticide?

Crop/fruit tree	Source of pesticide	Type of pesticide
....	....	....
....	....	....
....	....	....
....	....	....

4. Do you use any pesticide on your livestock? ____

(if yes, ask) What pesticide(s)?

Livestock	Source of Pesticide	Type of pesticide
....	....	....
....	....	....
....	....	....
....	....	....

Section C: Mother's dietary Habits.

1. Which foods do you consume mostly for:-

(a) Breakfast: ....

(b) Lunch ....

(c) Supper ....

(d) Others ....

(check the relevant boxes)

1= Mother eats virtually no food of animal origin including eggs and milk. ____

2= Mother eats mainly vegetarian diet but also milk and eggs. ____

3= Mother eats animal and vegetable foods. ____

4= Mother eats almost exclusively animal foods. ____

2. What is your staple food? ....

3. Do you eat fish?

1= Yes :___:
2= No :___:

(if yes, ask)

How many times do you take fish in the last two weeks?

....

4. What type of milk is consumed mainly in your household?

1= cow milk. :___:
2= goat milk. :___:
3= other specify ....

<u>Section D: Sample Collection</u>.

Breastmilk sample number ....

Date of collection ....

Time of day ....

QUESTIONAIRE 11 - TO BE USED TO GET INFORMATION ON PESTICIDES FROM AGRICULTURAL/LIVESTOCK OFFICERS, SALES OF PESTICIDES (etc) IN THE STUDY AREA.

Section A: Agricultural/livestock Officers.

1. Name of agricultural/livestock officer ______________________

   Location ______________________ Area ______________________

2. What pesticide do you advice farmers to use for stored foods?

Food	Type of Pesticide	Source/ (Local)
....	....	....
....	....	....
....	....	....
....	....	....
....	....	....

3. What pesticides do you advice farmers to use on crop/ fruit trees in the field?

Crop/fruit tree	Type of pesticide	Source (Local)
....	....	....
....	....	....
....	....	....
....	....	....
....	....	....

4. What pesticides are used on livestock in this location/area?

Type of Pesticide	Source
....	....
....	....
....	....
....	....
....	....

Section B: Pesticide available at local shopping centres.

(Inquire and observe) Pesticides available in local shops.

Shopping Centre	Location/area	Type of Pesticide available
....	....	....
....	....	....
....	....	....
....	....	....

Section C: Other pesticide sales outlets in Machakos Town.

Sales outlet	Location	Pesticides available to farmers
1. K.G.G.C.U.	....	
	....	
	....	
	....	
2. Machakos	....	
Cooperative	....	
Union	....	
3. Others	....	
Name	....	