INDOOR LEVELS OF POLYCYCLIC AROMATIC HYDROCARBONS FROM WOOD BURNING IN RURAL HOUSES IN WESTERN KENYA

BY

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ABSTRACT

Western Province of Kenya has prevalence of grass-roofed traditional houses, where cooking is done in open fire places using various biomass types, which leads to accumulation of soot under the roofs. Polycyclic Aromatic Hydrocarbons (PAHs) result from incomplete combustion of organic compounds and are emitted in gaseous phase, which may adsorb on to soot particles, or particulate phase. Exposure to mixtures of PAHs increases incidences of cancer in human populations. The high indoor temperatures probably cause volatilisation of the adsorbed PAHs, leading to higher continuous human exposure to PAHs. Data indicate that cancer cases are rampant in the region, yet no studies had been done to show potential causes of these cancers. The objective of this study was to extract, characterize and quantify the levels of PAHs in accumulated soot in grass-roofed houses in Western Kenya and to determine the variations in concentration of PAHs in the accumulated soot with biomass type used and house age. The houses were classified according to the predominant fuel source (perennial indigenous trees, exotic trees, shrubs and crop residues, and dung), and house age (0-5, 5-10, >10 years). The study design was factorial two, laid out in randomised complete block arrangements with fuel source as the main factor and house age as the second factor. The soot samples, collected from four houses in each sampling unit, were extracted by Soxhlet using dichloromethane, cleaned on silica gel column and analyzed by gas chromatography using open tubular capillary columns with flame ionization detector. Identification of PAHs was based on retention times of authentic standards and verified by gas chromatographic-mass spectral analysis. Quantification was based on peak area responses using the internal standard method and concentrations corrected for recovery. Separation of means and analysis of variance was done using a factorial two in randomized complete block design model. The PAHs levels significantly ($P \leq 0.05$) varied with biomass type in the order: Dung $\geq$ perennial indigenous trees $\geq$ exotic trees $\geq$ shrubs and crop residues. Use of dung and wood from perennial indigenous trees as fuel should be discouraged since they were higher emitters ($P \leq 0.05$) of PAHs. The PAHs emitted during indoor biomass burning significantly accumulated ($P \leq 0.05$) in soot with time. The accumulation patterns of some PAHs varied ($P \leq 0.05$) with variation in biomass type used. These results indicate that indoor burning of biomass fuel emits PAHs which accumulate in the accumulated soot; posing health risks to the inhabitants.

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CHAPTER 1

1.0 INTRODUCTION

1.1 General

Many third world countries use wood, straw, dung, leaves and other combustible materials as energy source for heating and cooking. Predominant, is the use of wood fuel since it is a renewable resource that is generally available in the surrounding at no or low financial cost. Estimates show that globally 50% of all houses and 90% of rural houses use solid fuels. In Kenya it is estimated that more than 85% of the houses use solid fuels, mainly wood, in very poorly ventilated conditions (Smith *et al.*, 2004). The fuels are typically burned in simple inefficient and poorly ventilated houses in traditional three-stone open fire places (Plate 1), which generate large volumes of smoke indoors. The generated smoke contains large amounts of organic materials which are pollutants (Smith, 2002). Usually, the burning is done for several hours each day, most of the times when people are indoors resulting in higher human exposure to smoke pollutants (Plate 2), probably, including polycyclic aromatic hydrocarbons (PAHs), than from outdoor sources (Smith, 2002). Red, teary eyes, running noses, persistent coughing and at times complaints of headaches as a result of exposure to smoke, are usually the identification marks of the people inside the houses during cooking and space warming (Miah *et al.*, 2008). The identities and levels of PAHs emitted as a result of biomass burning under the poorly ventilated conditions inside these houses, if any, are not known.

PAHs are a group of chemicals consisting of hydrogen and carbon in fused–ring structures. They consist of at least two benzene rings, but can contain additional fused rings that are not six-carbon membered. Small amounts of PAHs are found in the environment worldwide (Minnesota Pollution Control Agency, 1998). Anthropogenic sources of PAHs to the environment are more abundant than natural sources. Residential burning of wood is the largest anthropogenic source of most PAHs (Peters *et al.*, 1980; Ramadahl *et al.*, 1982). To obtain the most efficient and cleanest burn from wood, dry seasoned wood should be burned in a manner that allows air flow and abundant oxygen to the greatest amount of burning surface area. Burning a kilogram of wood in a new woodstove produces about 130 grams of carbon monoxide, 51 grams of hydrocarbons (including up to 10 grams of carcinogenic benzene and about 0.3 grams

of the highly carcinogenic PAHs) and 21 grams of fine particulates, (US EPA, 1986; Larson and Koenig, 1993). The traditional house in Western Province of Kenya is either cylindrical or cubical with low walls (about six feet above the ground surface), fairly high and steep grass thatched roofs and tiny wooden windows and door (Plate 3), hence it is generally dark indoors even during day time. Due to poor ventilation in these traditional rural houses, there is usually heavy accumulation of dense black soot under the roofs, over time, as a result of continuous use of various types of biomass fuel for cooking and space warming. It is not known whether PAHs emitted, if any, due to continuous biomass burning inside these traditional houses, also accumulate with time in the accumulated soot.

Plate 1:   Photograph of one of the traditional cooking fire places inside rural houses.

Plate 2: Photograph of the conditions inside rural houses during cooking.

Plate 3:   Photograph of a rural grass-roofed house in Western province of Kenya

Wood is mainly cellulose but it also contains lignin and resins. Lignin is a substance present in cell walls of plants and makes trees woody. Lignin constitutes about 15-25% by weight of woody biomass (Cassidy and Ashton, 2007). White (1986) reported lignin content of up to 33.8%, by mass, in soft wood. Softwoods, on average, generally contain relatively higher percentage, by mass, of lignin than hardwoods when the wood is oven dried, and their gross heat of combustion values are higher than those of hardwoods (White, 1986). Since the average amounts of resins for both soft and hard wood are about the same (White, 1986), it therefore means that hard woods have a higher percentage of cellulose, by mass, than softwoods. The heat content of natural fuels, including biomass, is related to their oxidation capacity, in which carbon atoms generally dominate and overshadow small variations in hydrogen (Susott et al., 1975). During combustion, some biomass burn with flames while others smolder. Smoldering fires lead to emission of higher PAH content than flaming fires due to lack of sufficient oxygen which favors incomplete combustion (Robinson et al., 2008). It is not known how variation in composition of different types of wood affects the oxidation capacity of the woods, the rate of combustion of the woods, as well as the identity and composition of the combustion products of each wood type.

Adsorption and absorption are the primary routes of PAHs association with particles of small diameters (fine particles) and large diameters (coarse particles), respectively (Hays et al., 2003). Studies show that during PAHs distribution, higher and lower molecular weight (MW$\leq$228) PAHs preferentially segregate to fine particles and coarse particles, respectively (Hays et al., 2003; Allen et al., 1996). Information on the exact mechanism of individual PAHs, including those emitted from biomass burning, association with particulate matter is scarce. The variation in PAH-particle association mechanisms with particle size and PAH mass may affect the accumulation patterns of the PAHs, emitted from burning various biomass types, in accumulated soot under the roofs of the traditional rural houses.

Fine particulate matter from residential wood burning is linked to adverse health problems (Larson and Koenig, 1994; Pintos et al., 1998). Considering that PAHs in wood smoke associate with ultra-fine particles, they putatively deposit in nasopharyngeal, trachea-bronchial, and pulmonary zones of the respiratory tract. The degradation of the PAHs adsorbed onto soot particles under the roofs in houses is likely

to be very slow, if any, due to absence of sunlight indoors (Butler and Crossley, 1981). Although PAHs have generally low vapour pressures which decrease with increasing molecular mass, their vapour pressure increases markedly with rise in ambient temperature (Murray *et al* 1974) which in turn affects the distribution coefficients of PAHs between the vapour phase and the particulate phase (Lane, 1989). The relatively high indoor temperatures due to constant heating for long hours, most likely, increase the volatilization of the PAHs adsorbed onto soot particles (Yamasaki *et al.*, 1982), under the roof. This could raise the levels of vapour phase PAHs in indoor air much higher than outdoor levels. Therefore, daily human exposure to PAHs indoors, probably, often far exceeds recommended safe levels (WHO, 1997). The scale of this environmental health problem can be aggravated when the probably, high PAHs concentrations from the wood burning and vapour phase PAHs volatilize from the soot under the roof due to high indoor temperatures (Yamasaki *et al.*, 1982), are combined with the duration of time people spend indoors during cooking. In some cases the cooking houses are also the living houses, thus increasing exposure duration. However, information on the relative accumulation levels of various PAHs in accumulated soot in houses due to continuous burning of various biomass types as fuel is missing.

Western Province in Kenya lies in the tropics and supports a large variety of tree species in the equatorial rain forests and homesteads. According to residents, the region was densely forested in the past but overdependence on wood fuel for cooking and space heating has depleted the forest cover and regrettably, some of the indigenous tree species are becoming endangered. The Kakamega Rain Forest on the eastern part of the province still has a large variety of perennial indigenous trees which are predominantly hardwood but with some softwood species. The residents of this sub-region express preference to use of wood from indigenous hardwood tree species, from the forest, because they burn for a longer time than the softwood species. The southern part of the province is densely populated. The indigenous trees species are extinct in this part and the most prevalent sources of wood fuel in the area are mainly the fast growing exotic trees, predominantly the soft wood species, grown in homesteads, although eucalyptus is also widely grown. Mumias area, on the central part of the province, is mainly an agricultural area where sugarcane is grown on large tracks of land. Barely a few trees are allowed to grow in this area to pave way for the cash crop. The residents in this area

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use dry sugarcane and shrubs due to lack of alternative fuel. Finally, Teso district, to the north-west of the province has a mixture of indigenous perennial trees and shrubs, scattered all over in virgin lands and in home and village forests, as well as some exotic trees. Human settlement is fairly sparse and part of the population is still deeply rooted in their traditional cooking methods of using dry cow dung as fuel for cooking and space warming. The variation in the identity and levels of PAHs emitted due to continuous burning of each of the various biomass types under the indoor conditions in these traditional houses has never been evaluated.

Exposure to mixtures of PAHs increases incidences of cancer in human populations (Grimmer *et al.,* 1991). Exposure to PAHs is the most important single factor in induction of lung tumours and increased incidences of tumours of the urinary bladder, renal, pelvis, mouth, pharynx, larynx and oesophagus (Brune *et al.,* 1981). The carcinogenic potency of PAHs may vary with the route of exposure (Grimmer *et al.,* 1991). The site of tumour development is related to the route of PAH exposure (Wall *et al.,* 1991) but tumours may also develop at distant sites (Grimmer *et al.,* 1991). The Kakamega Hospice is an organisation established in the middle of the year 2009 to document and give physical, psychological, spiritual, as well as bereavement support to medically diagnosed and reported cancer patients, at the Kakamega Provincial General Hospital, and their kin, in the region. According to the data collected by the Hospice between January and June 2010, cancer cases are rampant within the area with cancer of the oesophagus registering the highest number of newly diagnosed and reported cases each successive month (Kakamega Hospice Registry, 2010). Cases of cancer of the cervix, prostate cancer and breast cancer were diagnosed at the hospital and reported every month. Almost all the patients were aged 40 years and above and were either residing or had spent a significant part of their earlier lives in the rural areas (Kakamega Hospice Registry, 2010). Cancer related death cases have also been documented (Kakamega Hospice Registry, 2010). However, potential causes of these cancer incidences are not known.

Studies have shown that respiration immune system in humans is suppressed by exposure to wood smoke (Thomas and Zelikoff, 1999) and cooking with biomass fuels has been associated with chronic obstructive pulmonary disease (Dennis *et al.,* 1996). Although women have low prevalence of smoking in many developing countries, they

experience high rates of chronic obstructive pulmonary disease (Desai *et al.,* 2004). There is also evidence that chronic exposure to biomass smoke causes interstitial lung disease (Ramage *et al.,* 1988; Dhar and Pathania, 1991). In India, adults with continuous exposure to smoke from dung and wood had a significant relative risk for clinically confirmed tuberculosis (Gupta and Mathur, 1997). In Kenya, childhood asthma has been linked to indoor pollution from biomass fuel use (Mohamed *et al.,* 1995). Cases of red, teary eyes, running noses, persistent coughing and at times complains of headaches as a result of exposure to smoke from biomass burning and, probably, PAHs, are common among people inside the traditional rural houses in Western Province of Kenya during cooking and space warming.

Information on levels and identities of PAHs emitted from burning various wood species under the conditions inside the rural houses is scarce. It was therefore necessary to characterize and evaluate the PAHs in these houses. Such a study has not been reported before.

1.2 Statement of the problem

In Kenya, it is estimated that more than 85% of the houses, mostly in the rural areas, use solid fuel, mainly biomass, for space heating and cooking in poorly ventilated conditions (Smith *et al.,* 2004). Most of the houses where biomass is burned in open fire places have grass-thatched roofs with poor ventilation comprising tiny windows and usually single doors. This reduces the movement of air indoors resulting in incomplete combustion of the biomass and hence long durations of human exposure to gaseous PAHs. However, the identity and levels of PAHs emitted due to biomass burning under the poorly ventilated conditions inside these houses, if any, are not known.

The traditional house in Western Province of Kenya is either cylindrical or cubical with low walls, about six feet high from the ground surface, has fairly high and steep grass thatched roofs and tiny wooden windows and a single door (Plate 3), hence it is generally dark indoors even during day time. Due to poor ventilation in these traditional rural houses, there is usually heavy accumulation of dense black soot under the roofs, over time, due to continuous use of biomass as fuel for cooking and space warming. It

is not known whether PAHs emitted, if any, due to continuous biomass burning as fuel inside these traditional houses, also accumulate, with time, in the accumulated soot.

The types of biomass predominantly used as fuel for cooking and space warming in various sub-regions of the Western Province of Kenya vary. This is due to differences in the predominant tree species, agricultural activities as well as cultural practices: The Kakamega Rain Forest on the eastern part of the Province has a large variety of indigenous tree species. In the southern part, Sabatia/Vihiga, the most prevalent source of wood fuel in the area is mainly the fast growing exotic trees grown in homesteads. In Mumias, on the central part of the province, the residence use dry sugarcane and shrubs due to lack of alternative fuel. In Teso, to the north-western part, a section of the population is still deeply rooted in their traditional cooking methods; use of dry cow dung as fuel. It is not known how variation in the composition and modes of combustion of the various types of biomass fuel under the indoor conditions in these traditional houses affect the identity and composition of the PAHs emitted from burning of each the biomass type.

Further, the PAH-particle association modes vary with particle size and PAH mass (Hays *et al.*, 2003; Venkataraman *et al.*, 2002). It is not known if pattern of accumulation, due to household age, of each of the PAHs emitted from indoor biomass burning, varies with the predominant biomass types used as fuel

1.3 Hypotheses

1) Burning of biomass emits PAHs, but the PAHs do not deposit onto soot particles under the roofs of the traditional rural houses.

2) The levels of PAHs in accumulated soot do not vary significantly with the age of the house.

3) The levels of emitted PAHs do not significantly vary with variation in type of biomass fuel used, even though there is variation in composition and in the mode of combustion.

4) The patterns of accumulation of PAHs emitted from indoor biomass burning are homogenous due similarity in burning conditions, hence their interactions are not significant.

1.4 Objectives

1.4.1 Main objective

The main objective of this research was to characterize and quantify the levels of various PAHs in accumulated soot from selected houses with grass thatched roofs in Western Province of Kenya and to assess the levels of accumulation and distribution of the PAHs in soot in these houses with time.

1.4.2 Specific objectives

The specific objectives of this research were:

1. To characterize PAHs in soot samples from houses with grass thatched roofs in Western Province of Kenya, which predominantly use biomass fuel for space heating and cooking.

2. To determine the variation in indoor levels of PAHs in the region due to variations in biomass types predominantly burned.

3. To determine the variation in levels of PAHs in soot, resulting from biomass burning in the rural houses, with the age of the house.

4. To examine if the pattern of possible accumulation of PAHs due to age varies with the predominant biomass types used as fuel.

1.5 Justification

Biomass burning products depend on the type of biomass, while human exposure level to biomass combustion products, including PAHs, is a factor of the architecture of the house (Miah *et al.,* 2008). The traditional house in Western Province of Kenya has poor ventilation indoors. This exposes the inhabitants of the houses to smoke and volatile compounds (probably, including PAHs) which are produced from indoor biomass burning during cooking and space warming.

Data collected by the Kakamega Hospice, at the Kakamega Provincial General Hospital, indicate that cancer cases are rampant in the region with cancer of the oesophagus registering the highest number of newly diagnosed and reported cases each successive month (Kakamega Hospice Registry, 2010). Almost all the cancer patients either resided or had spent a significant part of their earlier lives in the rural areas. Cancer related death cases have also been documented by the Hospice (Kakamega

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Hospice Registry, 2010), yet no studies had been done to show potential causes of these cancer cases.

It was therefore necessary to characterize and evaluate the indoor levels of PAHs in traditional rural houses in Western province of Kenya which predominantly use biomass fuel for space heating and cooking, as this could be one reason for the high cancer incidences.

1.6 Significance of the study

Presently, there no guidelines in Kenya, either by the National Environmental Management Authority (NEMA) or the Ministry of Environment and Natural Resources or the Ministry of Public Health and Sanitation or Ministry of Health, on the levels of PAHs released to the environment. The findings also present an important basis upon which NEMA and other environmental conservation groups can push for legislation on levels PAHs emission to the environment. The findings of this study also highlight a new front towards which these ministries should consider channeling their efforts in the fight against cancer.

1.7 Scope and Limitation

The scope of this study was to characterize and quantify PAHs in soot samples from houses with grass thatched roofs in Western province of Kenya, which predominantly use biomass fuel for space heating and cooking. The study sought to establish if there is any significant variation in indoor levels of PAHs in the region due to variation in wood types or dung predominantly burned.

The study aimed to establish if there was accumulation of PAHs in the soot in these houses with time and hence determine the relative accumulation levels of various PAHs in the soot. For this reason, only those houses in which cooking and space heating using wood fuel exclusively and continuously, without a break, and in which soot had never been removed since the first day of wood burning were included in this study.

There was no control or restriction on use of biomass to exclusively one type. Possible contamination could not be isolated. It was also not possible to determine the exact amount of wood or dung used by each house annually. Since the houses are mainly occupied by nuclear families, it was assumed they use approximately the same

quantity annually. It was not possible to determine the levels of individual PAHs in samples in which their concentrations were below the minimum detection limit.

2.0 LITERATURE REVIEW

2.1 Definition and identity

Polycyclic aromatic hydrocarbons (PAHs) refer to a large class of organic compounds with two or more fused aromatic rings (Lee *et al.*, 1976a). PAHs contain only carbon and hydrogen atoms i.e. unsubsituted parent PAHs and their alkyl-substituted derivatives. PAHs are sometimes also referred to as polycyclic organic matter, polynuclear organic matter or polynuclear aromatic compounds, (Minnesota Pollution Control Agency, 1998). More than 100 PAHs have been identified in the atmospheric particulate matter (Lao *et al.*, 1973; Lee *et al.*, 1976a) and in emissions from coal-fired residential furnaces (Grimmer *et al.*, 1985). About 200 PAHs have been identified in tobacco smoke (Lee *et al.*, 1976b; 1981). Some common PAHs with their respective molecular formulae and relative molecular mass are shown in Table 1.

Table 1: Examples of PAHs with their formulae and molecular masses.

Compound	Molecular Formula	Molecular Mass
Naphthalene	$C_{10}H_8$	128.2
Acenaphthylene	$C_{12}H_8$	152.2
Acenaphthene	$C_{12}H_{10}$	154.2
Fluorene	$C_{13}H_{10}$	166.2
Anthracene	$C_{14}H_{10}$	178.2
Phenanthrene	$C_{14}H_{10}$	178.2
1-Methylphenanthrene	$C_{15}H_{12}$	192.3
Fluoranthene	$C_{16}H_{10}$	202.3
Pyrene	$C_{16}H_{10}$	202.3
Benzo[a]fluorene	$C_{17}H_{12}$	216.3
Benzo[b]fluorene	$C_{17}H_{12}$	216.3
Benzo[ghi]fluoranthene	$C_{18}H_{10}$	226.3
Cyclopenta[cd]pyrene	$C_{18}H_{10}$	226.3
Benz[a]anthracene	$C_{18}H_{12}$	228.3
Benzo[c]phenanthrene	$C_{18}H_{12}$	228.3
Chrysene	$C_{18}H_{12}$	228.3
Triiphenylene	$C_{18}H_{12}$	228.3
5-Methylchrysene	$C_{19}H_{14}$	242.3
Benzo[b]fluoranthene	$C_{20}H_{12}$	252.3
Benzo[j]fluoranthene	$C_{20}H_{12}$	252.3
Benzo[k]fluoranthene	$C_{20}H_{12}$	252.3
Benzo[a]pyrene	$C_{20}H_{12}$	252.3
Benzo[e]pyrene	$C_{20}H_{12}$	252.3
Perylene	$C_{20}H_{12}$	252.3
Anthanthrene	$C_{22}H_{12}$	276.3
Benzo[ghi]perylene	$C_{22}H_{12}$	276.3
Indeno[1,2,3-cd]pyrene	$C_{22}H_{12}$	276.3
Dibenz[a,h]anthracene	$C_{22}H_{14}$	278.4
Coronene	$C_{24}H_{14}$	300.4
Dibenzo[a,e]pyrene	$C_{24}H_{14}$	302.4
Dibenzo[a,h]pyrene	$C_{24}H_{14}$	302.4
Dibenzo[a,i]pyrene	$C_{24}H_{14}$	302.4
Dibenzo[a,l]pyrene	$C_{24}H_{14}$	302.4

(Source: Community Bureau of Reference, 1992).

2.2 Properties of PAHs

PAHs generally have high melting and boiling points hence all PAHs are solids at room temperature. They have low vapour pressures which decreases with increasing molecular mass (IARC, 1983). These affect the adsorption of individual PAH on to

particulate matter in the atmosphere and their retention on the particulate matter (Thrane and Mikalsen, 1981). Table 2 below shows physical properties of some polycyclic aromatic hydrocarbons.

Table 2: Some physical properties of some polycyclic aromatic hydrocarbons.

Compound	Colour	Melting point (^oC)	Boiling point (^oC)	Vapour pressure (Pa at 25^oC)	Log Koc Values
Naphthalene	White	81	217.9	10.4	2.99
Acenaphthylene	White	92-93	nf	8.9×10^{-1}	3.61
Acenaphthene	White	95	279	2.9×10^{-1}	3.53
Fluorene	White	115-116	295	9.0×10^{-2}	3.79
Anthracene	Colourless	216.4	342	8.0×10^{-4}	4.14
Phenanthrene	Colourless	100.5	340	1.6×10^{-2}	4.17
1-Methylphenanthrene	Colourless	123	354-355	nf	nf
Fluoranthene	Pale Yellow	108.8	375	1.2×10^{-3}	4.82
Pyrene	Colourless	150.4	393	6.0×10^{-4}	4.78
Benzo(a)fluorene	Colourless	189-190	399-400	nf	nf
Benzo(b)fluorene	Colourless	213.5	401-402	nf	nf
Benzo(ghi)fluoranthene	Yellow	128.4	432	nf	nf
Cyclopenta(cd)pyrene	Orange	170	439	nf	nf
Benzo(a)anthracene	Colourless	160.7	400	2.8×10^{-5}	5.50
Chrysene	Colourless	253.8	448	8.4×10^{-5}	5.45
Triphenylene	Colourless	199	425	nf	nf
5-Methylchrysene	Colourless	117.1	458	nf	nf
Benzo(b)fluoranthene	Colourless	168.3	481	6.7×10^{-5}	5.39
Benzo(j)fluoranthene	Yellow	165.4	480	2.0×10^{-6}	nf
Benzo(k)fluoranthene	Pale Yellow	215.7	480	1.3×10^{-8}	5.59
Benzo(a)pyrene	Yellowish	178.1	496	7.3×10^{-7}	5.63
Benzo(e)pyrene	Pale Yellow	178.7	493	7.4×10^{-7}	nf
Perylene	Yellow/Colourless	277.5	503	nf	nf
Anthracene	Golden yellow	264	547	nf	nf
Benzo(ghi)perylene	Pale yellow	278.3	545	1.4×10^{-8}	6.08
Indeno(1,2,3-cd)pyrene	Yellow	163.6	536	1.3×10^{-8}	6.08
Dibenzo(a,h)anthracene	Colourless	266.6	524	1.3×10^{-8}	6.33
Coronene	Yellow	439	525	2.0×10^{-10}	nf
Dibenzo(a,e)pyrene	Pale yellow	244.4	592	nf	nf
Dibenzo(a,h)pyrene	Golden yellow	317	596	nf	nf
Dibenzo(a,i)pyrene	Greenish yellow	282	594	3.2×10^{-10}	nf
Dibenzo(a,l)pyrene	Pale yellow	162.4	595	nf	nf

"nf" denotes "not found in literature"
(Source: IARC, 1983)

The vapour pressure of PAHs, however, increases markedly with rise in ambient temperature (Murray *et al.,* 1974), which affects the distribution coefficients, between gaseous and particulate phases (Lane, 1989). PAHs in the environment exist mostly in the vapour and particulate phases. Due to high indoor temperatures in houses, the PAHs adsorbed onto soot particles probably vaporise (Yamasaki *et al.,* 1982), increasing their levels in vapour phase.

PAHs have very low solubility in water, especially those with high molecular weight. However, they are soluble in many organic solvents such as n-hexane, methanol, cyclohexane, dichloromethane (DCM) and toluene among others (IARC, 1983; ATSDR, 1990; Lide, 1991) and are therefore extracted, cleaned and stored in organic solvents.

2.3 Sources of PAHs in the environment
2.3.1 Natural occurrence of PAHs

Forest fires and volcanoes are the main natural sources of PAHs in the environment (US National Research Council, 1983; Baek *et al.,* 1991). For instance in Canada, about 2000 tones of PAHs per year are attributed to natural forest fires (Environmental Canada, 1994). Benzo(a)pyrene, benzo(e)pyrene, perylene and phenanthrene have been identified in coal (Neff, 1979; Anderson *et al.,* 1986). Chrysene and dibenzo(a,h)anthracene have also been identified in the greenish-yellow fluorescent curtisite from the surface vents of the hot springs at Skagg Springs, California, USA and in the bituminous mercury ore , idrialite, from Idria, Yugoslavia (West *et al.,* 1986). In Kenya, no major natural source has been identified since forest fires are rare and there are no coal deposits. The most probable major PAHs sources in the country are the anthropogenic sources.

2.3.2 Anthropogenic sources of PAHs

Major anthropogenic sources of PAHs in the environment include combustion of fossil fuels, automobile engine exhausts (Herlan, 1982; Lalah and Kaigwara, 2005), atmospheric fall-out of fly-ash particulates, coal tar and other coal processing wastes, accidental spills from oil tankers and ships (Carmicheal *et al.,* 1990), tobacco and cigarette smoke, refuse and waste incineration, coal gasification and liquefaction

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processes, creosote and other wood preservation wastes (Anderson *et al* 1986), petrochemical industrial effluents (Franck and Stadelhofer, 1987; Collin and Hoke, 1985; Griesbaum *et al.,* 1989), forest and prairie fires, rural and urban sewage sludge, municipal waste water discharges run-off, smoke, char-broiled or pan-fried foods, river-borne pollution, commercial and pleasure boating activities and domestic wood burning (ATSDR, 1995a). In rural areas the most probable major anthropogenic source of PAHs may be residential biomass burning in houses for cooking and space warming.

2.3.2.1 Emission of PAHs due to incomplete combustion

Polycyclic aromatic hydrocarbons (PAHs) form as a result of incomplete combustion of organic compounds. The primary source of PAHs in air in developing countries is the incomplete combustion of wood and fuel for residential heating (ATSDR, 1995a). The PAHs are found in gasoline or diesel motor vehicle exhaust, by-products of open fires or refuse burning, coal tar, coal tar pitch, coke oven emissions, creosote, mineral oils, bitumen, industrial smoke and soot, tar or smoke condensates and charcoal-broiled foods (ATSDR, 1995a). Estimates reveal that motor vehicles are a major source of atmospheric PAHs emission (Van Metre *et al.,* 2000; Nielson, 1996) in urban environment. Domestic heating in general and residential wood combustion, in particular, are another major source of PAHs to both urban and rural outdoor air (Menichini, 1992). In a 1996 estimate of the inventory of toxic chemical emissions to the air, approximately 60% of all the PAHs emissions from point, area and mobile sources in the Great Lakes states in USA were attributed to residential wood burning (Great Lakes Commission, 2000). In Kenya it is not known how much PAHs, either in the atmosphere or indoors, in both rural and urban environments, are attributable to residential wood burning.

2.3.2.2 Domestic residential burning of biomass

Biomass fuels are at the low end of the energy ladder in terms of the combustion efficiency, heat content and cleanliness (Smith and Liu, 1994). Smoke and soot from biomass combustion produces a large number of health damaging air pollutants which include PAHs (Bruce *et al.,* 2000). Numerous PAHs including acenaphthene, acenaphthylene, fluorene, phenanthrene, anthracene, 1-methylphenanthrene,

fluoranthene, pyrene, benzo(a)fluorene, benzo(g,h,i)fluoranthene, benzo(i)phenanthrene, cyclopenta(c,d)pyrene, benzo(c)anthracene, chrysene plus triphenylene, benzo(b)fluoranthene, benzo(e)pyrene, benzo(a)pyrene, prylene, indeno(1,2,3-c,d)perylene, benzo(g,h,i)perylene and anthanthrene have been detected in atmospheric emissions from straw burning residential stoves (Ramdahl and Moeller, 1983). Analysis for the PAHs content of soot from open fires, under laboratory conditions, gave 3-240 ppm benzo(a)pyrene, 2-190 ppm chrysene, 2-100 ppm benzo(a)anthracene, 1-77 ppm indeno(1,2,3-c,d)pyrene, 2-39 ppm benzo(e)pyrene, 1-29ppm benzo(g,h,i)perylene, 1-18 ppm coronene,1-14 ppm perylene and 1-12 ppm anthracene (Cretney et al., 1985). In Western Province of Kenya, various types of biomass are burned in open fire places, as fuel, in poorly ventilated conditions in the traditional rural houses. The identity and levels of the PAHs and other health damaging combustion products emitted from biomass burning under these conditions is not known.

Burning conditions are apparently the major determinants of emissions and are much more important than the kind of wood burnt (Ramdahl et al., 1982). In developing countries where large proportions of houses rely on biomass fuels for cooking and space heating, the concentrations of PAHs tend to be highest indoors (Bruce et al., 2000). The fuels are burned in simple inefficient and poorly ventilated house cooking stoves or open fire places, which generate large volumes of smoke indoors (Smith, 2002). The organic component of smoke is partly PAHs molecules which are precursors to soot formation. This leads to heavy accumulation of soot under the roofs of the houses. The cooking stoves or open fires are used for several hours, which raise indoor temperatures, each day at times when people are indoors. The vapour pressure of PAHs increases markedly with rise in ambient temperature (Murray et al., 1974) and this in turn affects the distribution coefficients between the gaseous phase and particulate phase PAHs (Lane, 1989). The high indoor temperatures probably cause an increase in volatilization of the adsorbed PAHs from soot particles (Yamasaki et al., 1982) and this raises the levels of vapour phase PAHs in indoor air resulting in high exposure to gaseous PAHs, including highly carcinogenic ones, than from outdoor sources (Smith, 2002). It is not known if the PAHs emitted from the continuous

biomass burning in the houses accumulate in the accumulated soot, under the roofs, with time.

The most significant form of biomass burning in Western province of Kenya is wood burning. Wood is a renewable resource that is generally in ample supply in Kenya, although the country is facing an imminent severe deforestation. Burning a kilogram of wood, for instance in a new wood-stove, produces about 130 grams of carbon monoxide, 51 grams of hydrocarbons (including up to 10grams of carcinogenic benzene, 21grams of fine particulates and about 0.3 grams of highly carcinogenic PAHs), (US EPA, 1986; Larson and Koenig, 1993). The levels of biomass combustion products emitted from biomass burning in open fire places under the indoor conditions in the traditional rural houses are not known.

2.3.2.3 Effect of lignin on burning of biomass

Wood is mainly cellulose but it also contains lignin. Wood also contains resins, turpenes and other substances which can be extracted with solvents such as ethanol/benzene (ASTM, 1984), generally referred to as extractives. Wood from which these extractives have been removed is therefore referred to as 'extractive-free' wood, while 'unextracted' wood is wood on which no extraction has been done. The heat content of natural fuels, including biomass, is related to their oxidation capacity, in which carbon atoms generally dominate and overshadow small variations in hydrogen (Susott *et al.,* 1975). Cellulose has a gross heat of combustion value of 8000 Btu/lb (Baker 1983), whereas lignin has a gross heat of combustion value of 15000 to 16000 Btu/lb (Howard, 1973). Lignin is a substance present in cell walls of plants and makes trees woody and it constitutes about 15-25% by weight of woody biomass (Cassidy and Ashton, 2007); although lignin content of up to 33.8%, by mass, in softwood has been reported (White, 1986). Softwoods, on average, generally contain relatively higher percentage, by mass, lignin than hardwoods when the wood is oven dried (Table 3), and their gross heat of combustion values are higher than those of hardwoods (Table 3). This is primarily caused by their difference in lignin contents since the average amount of resins for both is about the same (White, 1986). The rates of burning of various biomass types in the poorly ventilated conditions in rural houses are expected to vary. It is not known how variation in composition of different types of biomass affects the

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oxidation state of the biomass, the rate of combustion of the biomass, as well as the identity and composition of the combustion products of each biomass type. Information on levels and identities of PAHs emitted from burning various biomass types is scarce.

During combustion, some biomass burn with flames while others smolder. Smoldering fires in biomass leads to emission of much higher PAH content than flaming fires due to lack of sufficient oxygen which favors incomplete combustion (Robinson *et al.*, 2008). Dry cow dung, used as fuel in some of the traditional rural houses, smolders, while wood burns with a flame during cooking and space warming. The variation in the relative levels of PAHs emitted due to the variation in the modes of burning of the various biomass types, under the indoor conditions of the traditional rural houses, is unknown.

Table 3: Percentage by mass of lignin and extractives, and the gross heat of combustion values for both unextracted and extractive-free woods of some softwoods and hardwoods.

Species	% of oven-dried wood		Gross Heat of Combustion Values (Btu/lb)	
	Lignin	Extractives	Unextracted wood	Extractive-free wood
Softwoods				
Engelmann spruce	26.9	2.4	8,650	8,570
Western red-cedar	30.8	7.8	8,890	8,800
Southern pine	26.8	5.4	8,890	8,610
Redwood	33.8	8.2	9,120	8,940
Hardwoods				
Maple	21.7	5.3	8,450	8,450
Yellow-poplar	20.5	3.8	8,440	8,360
Red oak	22.5	6.4	8,590	8,410
Basswood	18.1	6.7	8,590	8,290

Btu/lb – British thermal units per pound
[Source: White, 1986]

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2.4 Distribution and fate of PAHs

2.4.1 Distribution of PAHs between media

The distribution of PAHs in the environment depends on their physicochemical properties. These include;

i) *Aqueous solubility*: PAHs are hydrophobic compounds with very low solubility in water (McVeety and Hites, 1988). Their solubility in water tends to decrease with increasing molecular mass. Thus airborne PAHs adsorbed onto particulate matter result in effective wet deposition but gaseous PAHs are removed from the air only to a minor degree (McVeety and Hites, 1988). Inside the rural house, wet deposition does not take place because the grass thatched roofs are water-tight. As a result the gaseous PAHs within the houses are likely to persist for a much longer time than outdoors increasing the possibility of longer human exposure to high levels of vapour phase PAHs indoors than outdoors.

ii) *Vapour pressure*: The vapour pressure of PAHs under environmental conditions is very low and tends to decrease with increasing molecular mass. The maximum at room temperature is 10.4 Pa (naphthalene) and the calculated minimum is 3×10^{-12} Pa [dibenzo(a,i)pyrene] (Sonnefeld *et al.,* 1983). This characteristic affects the adsorption of individual PAHs on to particulate matter in the atmosphere (Thrane and Mikalsen, 1981). Vapour pressure, however, increases markedly with increase in ambient temperature (Murray *et al.,* 1974) which additionally affects the distribution coefficients between gaseous and particulate phases (Lane, 1989). The high indoor temperatures in houses probably increase volatilisation of adsorbed PAHs from soot particles (Yamasaki *et al.,* 1982), which is likely to raise the levels of gaseous PAHs in indoor air. The levels of adsorbed PAHs on accumulated soot, under the roofs of the traditional grass-roofed houses, due to continuous burning of biomass as fuel is not known

2.4.2 Distribution of PAHs in the gaseous phase.

Polycyclic aromatic hydrocarbons (PAHs) are emitted to the atmosphere where they are transported in the vapour phase until they are adsorbed onto particulate matter (Baker and Eisenreich, 1990). The distribution of a substance between air and particulate matter under atmospheric conditions depends on the lipophilicity, vapour

pressure and aqueous solubility of the substance (McVeety and Hites, 1988). Generally, PAHs with 2-4 aromatic rings occur in the vapour phase and rarely exist in adsorbed state, whereas PAHs consisting of more aromatic rings exist mainly in adsorbed state (Hoff and Chan, 1987; McVeety and Hites, 1988; Baker and Eisenreich, 1990). PAHs emitted during biomass burning, if any, are likely to be adsorbed onto particles like fly-ash and soot that are also formed during combustion.

PAHs are widely distributed in the environment, probably because they can travel in air for long distances without significant degradation (McVeety and Hites, 1988). For instance, PAHs emitted from the United Kingdom are distributed in air up to Norway and Sweden during winter without undergoing degradation (Bjorseth and Lunde, 1979). Washout ratios calculated from measurements in rain and snow in the area of Northern Lake Superior, during one year showed that airborne PAHs adsorbed onto particulate matter result in effective wet deposition. Gaseous PAHs are removed only to a minor degree because they are highly hydrophobic (McVeety and Hites, 1988). The gaseous PAHs in indoor air are expected to persist for much longer time since the air movement is more restricted indoors than outdoors. As a result, there is very low dispersal of the PAHs indoors, which probably leads to high human exposure, as well as increase the possibility of adsorption of the vapour phase PAHs on the particles of accumulated soot under the roofs of the traditional grass-thatched houses.

2.5 Environmental fate of PAHs

2.5.1 Volatilization

The rates of removal and volatilization of PAHs are strongly dependent on environmental conditions such as ambient temperatures and wind velocity (Klopffer *et al.*, 1982). Although PAHs are released into the environment mainly in air, considerably higher concentrations are found in aqueous media and particulate phases such as soot samples because of low vapour pressure (Klopffer *et al.*, 1982). Under similar conditions, 30% of naphthalene volatized from the soil after 48 hours, but there was negligible loss of PAH with three or more rings (Park *et al.*, 1990). Volatilization of adsorbed PAHs from soot particles depends on ambient temperatures (Yamasaki *et al.*, 1982). On average, indoor temperatures in the traditional grass-roofed houses are usually higher than those outdoors due to continuous burning of biomass in open fire

places as fuel. Hence the rate of volatilization of PAHs, especially the low molecular weight ones, adsorbed on to accumulated soot in these houses, is likely to be higher than that outdoors.

2.5.2 Adsorption onto particulates.

The bulk of PAHs in the environment are tied to organic matter (Karickhoff *et al.*, 1979). The vapour phase and adsorbed phase partition of these PAHs vary with ambient temperature (Yamasaki *et al.*, 1982). The PAHs which arise from residential wood burning most probably get adsorbed onto soot particles which accumulate under the roofs of the traditional grass-thatched houses. Respirable particulate matter has been linked to human morbidity and mortality (Pope, 2000; Samet *et al.*, 2000). The particles deposit in the human respiratory tract zones, trancheo-bronchial, pulmonary, nasopharyngeal and thoracic zones, depending on size of the particles (Balashazy *et al.*, 1999; Tsuda *et al.*, 2002). The PAH content on the accumulated soot particles under the roofs of the traditional rural houses, as a result of continuous burning of biomass as fuel, is not known. Determination of this is essential from the human health standpoint.

2.5.3 Transformation and Degradation
2.5.3.1 Biodegradation

PAHs can be lost from the environment through microbial breakdown, photo-oxidation and volatilization (Smith *et al.*, 1996). PAHs are usually adsorbed strongly to organic particles, which reduce bioavailability. This greatly slows down biological degradation process (Luitgen and Piet, 1983; Environment Canada, 1994). Bio-degradation is the major mechanism for removal of PAHs from the soil. The rate of degradation depends on several factors which include characteristics of the soil and its microbial population, properties of the PAHs present, temperature, pH, oxygen content, soil type, nutrients and presence of other substances that can act as co-metabolites (Sims and Overcash, 1983). The microbial population in soot under roofs is almost zero due to the dry conditions, thus removal of PAHs adsorbed on to soot particles under the roofs in rural houses through bio-degradation is expected to be very slow.

2.5.3.2 Photodegradation

Photodegradation plays a major role in removal of PAHs in air and water, but to a very low extent in soils and sediments due to low light intensity (Park *et al.*, 1990). In natural waters, photodegradation of PAHs takes place only in the upper few centimetres of the aqueous phase. PAHs are photodegraded in air and water by two processes; direct photolysis by light with a wavelength <290nm, and indirect photolysis by at least one oxidizing agent such as OH·, O_x, $(x = 2$ to $3)$, and NO_x, $(x = 1$ to $2)$, in air and ROO· radicals in water (Wild and Jones, 1993). However under tropical environmental conditions, PAHs of high molecular mass, i.e. those with more than four aromatic rings are almost completely adsorbed onto fine particles which reduce the degradation rate remarkably (Hoff and Chan, 1987; McVeety and Hites, 1988; Baker and Eisenreich, 1990).

Degradation half-lives of 3.7-30 days have been reported for the reaction of NO_x with various PAHs adsorbed on soot. The degradation is much slower in the absence of sunlight (Butler and Crosley, 1981). PAHs in wood smoke and gasoline exhaust do not degrade significantly during winter in the extreme northern and southern latitudes owing to low temperatures and low angle of the sun (Kamens *et al.*, 1986). The degradation rate however increases with increase in the temperature and angle of the sun, and further with increasing humidity (Kamens *et al.*, 1991). Information on degradation PAHs in tropical environment is scarce. Indoors, degradation of PAHs is expected to be very low due to absence of sunlight.

2.6 Human exposure

The main sources of non-occupational human exposure pathways to PAHs are through active and inactive inhalation of compounds in tobacco smoke, wood smoke and soot from open fires and cooking using wood and dung as fuel, contaminated air from emissions from vehicle traffic, industrial plants and coal burning, and tobacco smoking (ATSDR, 1995b). Others include ingestion of contaminated food and drinking water, ingestion of house dust and dermal contact with coal tar in cosmetics and shampoos (ATSDR, 1995b). Soot particles often fall back from the roofs as tiny particles in the air indoors, on to food, drinking water and on beddings as insects, bats, birds and flies fly and patch on them under the roof. This, most probably, leads to

increased human exposure to, and, intake of the toxic PAHs indoors through inhalation of the air borne descending particles and ingestion, besides directly inhaling air contaminated with vapour phase PAHs from burning biomass. The level of PAHs in accumulated soot in traditional rural grass-roofed houses, which would indicate level of human exposure, is not known.

In developing countries, the greatest PAH exposure is through use of wood, straw, dung and leaves for heating and cooking by almost all houses (Bruce *et al.*, 2000). The fuels are typically burned in simple inefficient and poorly ventilated house cook-stoves or open fire places, which generate large volumes of smoke indoors (Smith, 2002). The organic component of smoke is partly PAH molecules which are precursors to soot formation. This leads to heavy accumulation of soot under the roofs of the houses. Moreover the cook-stoves or open fires are used for several hours each day and at times when people are indoors, which raises indoor temperatures. The vapour pressure of PAHs increases markedly with rise in ambient temperature (Murray *et al.*, 1974) and this in turn affects the distribution coefficients between the gaseous phase and particulate phase PAHs (Lane, 1989). The high indoor temperatures probably causes increase in volatilization of the adsorbed PAHs from soot particles (Yamasaki *et al.*, 1982) and this raises the levels of vapour phase PAHs in indoor air resulting in much higher exposure to gaseous PAHs, than from outdoor sources (Smith, 2002). The health problems associated with exposure to PAHs are well documented in literature (Thomas and Zelikoff, 1999; ATSDR, 1995a; Grimmer *et al.*, 1991; Gupta *et al.*, 1991; Brune *et al.*, 1981). The indoor levels of PAHs in these traditional rural houses are not known.

2.7 Human exposure limits

The US EPA has classified PAHs as probable human carcinogens with sufficient evidence from animal studies (US EPA, 1994). As a result, the World Health Organisation (WHO), the European Union (EU) and some countries have set ambient air quality limits and/or guideline values for PAHs (Rubailo and Oberenko, 2008). Table 4 below gives the non-mandatory ambient air quality limits and/or guide values for the PAHs.

Table 4: Non-mandatory ambient air quality standards for PAHs.

Country	Limit Value (ng/m^3)	Annual Average Guide Values (ng/m^3)
Australia	nvs	1.0
Belgium	1.0	0.5
Croatia	2.0	0.1
Germany	nvs	10.0
India	nvs	5.0
Netherlands	1.0	0.5
France	0.7	0.1
Italy	1.0	Nvs
Sweden	nvs	0.1
WHO	nvs	1.0
EU*	6.0	Nvs

* - To be met in the year 2010; nvs – No value set
[Source: Rubailo and Oberenko, 2008]

2.8 Toxicity of PAHs

2.8.1 Acute and sub-acute toxicity

Polycyclic aromatic hydrocarbons (PAHs) generally have a low degree of acute toxicity to humans (ATSDR, 1995a). Single doses of PAHs have moderate to low toxicity with LD_{50} values generally less than 100mg/kg body weight after intravenous injection and less than 500 mg/kg body weight after oral administration. For instance, naphthalene has shown dose-dependent abnormalities in the bronchial region (Clara cells) and depletion of pulmonary glutathione, which protects against the toxicity of xenobiotics within 6 h of administration (Honda *et al.*, 1990). Cytotoxicity to the olfactory epithelium with necrosis in the oral cavity has been associated with administration of naphthalene by injection (Plopper *et al.*, 1992). Oral administration of anthracene has been associated with increased carboxylesterase activity in the intestinal mucosa by 13% (Nousiainen *et al.*, 1984). Death due to myelotoxicity has been reported after daily oral administration of benzo(a)pyrene at 120 mg/kg body weight to mice daily for four days (Legraverent *et al.*, 1983). Oral administration of

benzo(a)pyrene and benzo(a)anthracene at 150 mg/kg, and phenanthrene at 100 mg/kg body weight daily for four days showed suppressed carboxylestrase activity in the intestinal mucosa (Nousiainen *et al.,* 1984). The PAHs emitted as a result of biomass burning in the traditional rural houses of Western Kenya, most probably get adsorbed on to the particles of the accumulated soot under the roofs. The soot particles often fall back from the roofs as tiny particles in the air inside, on to food, drinking water and on beddings as insects, bats, birds and flies fly and patch on them under the roof. This probably leads to increased exposure to and intake of the toxic PAHs indoors through inhalation of the air borne descending particles and ingestion, besides inhaling smoke directly from burning wood. The identity and levels of PAHs content of these soot particles is not known.

2.8.2 Sub-chronic and chronic toxicity

Studies show that effects of chronic exposure to PAHs involve primarily the pulmonary, gastrointestinal, renal and dermatological systems (Gupta *et al.,* 1991), though the most significant end point of chronic PAH exposure is cancer (ATSDR, 1995a). Other evidence indicate that exposure to PAHs, especially benzo(a)pyrene, which is found in biomass smoke, cause immune suppression in humans and can increase the risk of infections and diseases (Kong *et al.,* 1994; Hardin *et al.,* 1992; Thomas and Zelikoff, 1999).

Mice exposed to benzo(a)pyrene at 500 mg/kg body weight per day, showed symptoms of laboured respiration, drooping eyelids, unkempt appearance, decrease in erythrocyte in females, decreased haemoglobin concentration and increased total serum bilirubin level in females and significant increases in absolute and relative spleen, kidney and liver weights were observed (US EPA, 1989).

Cooking with biomass fuels has been associated with chronic obstructive pulmonary disease (Dennis *et al.,* 1996). Although women have low prevalence of smoking in many developing countries, they experience high rates of chronic obstructive pulmonary disease (Desai *et al.,* 2004). There is also evidence that chronic exposure to biomass smoke causes interstitial lung disease (Ramage *et al.,* 1988; Dhar and Pathania, 1991). In India, adults with continuous exposure to smoke from dung and wood had a significant relative risk for clinically confirmed tuberculosis (Gupta and

Mathur, 1997). In Kenya, childhood asthma has been linked to indoor pollution from biomass fuel use (Mohamed *et al.*, 1995).

2.8.3 Reproductive effects, embryotoxicity and teratogenicity

Polycyclic aromatic hydrocarbons (PAHs) can cross the placenta and induce adverse effects on the embryo and foetus. Benzo(a)anthracene, benzo(a)pyrene, dibenzo(a,h)anthracene and naphthalene are embryotoxic (Rigdon and Neal, 1965). Benzo(a)pyrene also has adverse effects on female fertility, reproduction and post-natal development (Rigdon and Neal, 1965). For instance, benzo(a)pyrene metabolites formed in the mother cross the placenta causing foetal death or malformation (Shum *et al.*, 1979). Ovarian volume, weight and follicle numbers reduced following administration by injection at single dose of 10µg of benzo(a)pyrene into the ovary of mice after two weeks. A single dose of intraperitoneal injection of benzo(a)pyrene reduced fertility and destroyed primordial oocytes of mice in dose dependent ratio (Mattison *et al.*, 1980). In India, women using biomass fuels during pregnancy have been reported to have an excess risk of 50% of having stillbirth (Mavalankar *et al.*, 1991). Exposure to biomass smoke during pregnancy has also been linked to low birth weight (Boy *et al.*, 2002) and prematurity (Bobak, 2000). The rural population in the Western Province of Kenya use biomass (wood or dry cow dung) as fuel, in open fire places, in poorly ventilated conditions. This generates large volumes of smoke, which probably includes PAHs, indoors. Determination of the identity and relative levels of PAHs emitted in these houses due to biomass burning is an important step in evaluating the health risks the inhabitants of these houses are exposed to.

2.8.4 Mutagenicity

Deoxyribonucleic Acid (DNA) binding adducts of benzo(a)pyrene have been found in human cell systems (Moore *et al.*, 1982; Harris *et al.*, 1984). Formation of DNA adducts was inhibited in the presence of scavengers of active oxygen species like super oxide dimutase, catalase and citrate-chelated ferric iron indicating that reactive oxygen such as super oxide, OH˙ radicals and singlet oxygen may be involved in DNA binding of PAH (Bryla and Weyand, 1991). Benzo(a)pyrene at a total dose of 10mg/kg body weight induced gene mutations in mice (Davidson and Dawson, 1976). In a study

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of activity of PAHs in short-term tests, only three (anthracene, fluorene and naphthalene) of the 33 PAHs considered, were inactive in all the tests, while 16 had mutagenic effects, 8 showed tendency to mutagenic activity (Community Bureau of Reference, 1992). Table 5 gives a classification of the 33 PAHs tested based on their genotoxicity.

Table 5: Classification of PAHs according to their genotoxicity.

Compound	Genotoxicity
Acenaphthene	(?)
Acenaphthylene	(?)
Anthanthrene	(+)
Anthracene	-
Benz[a]anthracene	+
Benzo[b]fluoranthene	+
Benzo[j]fluoranthene	+
Benzo[ghi]fluoranthene	+
Benzo[k]fluoranthene	+
Benzo[a]fluorene	(?)
Benzo[b]fluorene	(?)
Benzo[ghi]perylene	+
Benzo[c]phenanthrene	+
Benzo[a]pyrene	+
Benzo[e]pyrene	+
Chrysene	+
Coronene	(+)
Cyclopenta[cd]pyrene	+
Dibenz[a,h]anthracene	+
Dibenzo[a,e]pyrene	+
Dibenzo[a,h]pyrene	(+)
Dibenzo[a,i]pyrene	+
Dibenzo[a,l]pyrene	(+)
Fluoranthene	+
Fluorene	-
Indeno[1,2,3-cd]pyrene	+
5-Methylchrysene	+
1-Methylphenanthrene	+
Naphthalene	-
Perylene	+
Phenanthrene	(?)
Pyrene	(?)
Triphenylene	+

+, positive; -, negative; ?, questionable; ( ), result derived from small database

Source: Community Bureau of Reference (1992).

The synergetic effects of exposure to PAH mixtures on DNA is not documented. Exposure to biomass fuel combustion products has been linked to increase in the frequency of cytogenetic alterations in blood lymphocytes of exposed populations (Musthapa *et al.*, 2004). Exposure to biomass fuel smoke is reported to lead to greater levels of DNA damage than exposure to liquefied petroleum gas combustion products (Panday *et al.*, 2005). It is expected that biomass fuel burning, under the poorly ventilated conditions inside the traditional rural houses, emits PAH mixtures including those classified by US EPA as probable mutagens.

2.8.5 Carcinogenicity

Many individual PAHs are carcinogenic to animals and humans. Exposure to mixtures of PAHs increases incidences of cancer in human populations (Grimmer *et al.*, 1991). PAHs produce tumours both at site of exposure and at distant sites. The carcinogenic potency of PAHs may vary with the route of exposure (Grimmer *et al.*, 1991). Exposure to PAHs is an important factor in induction of lung tumours and increased incidences of tumours of the urinary bladder, renal, pelvis, mouth, pharynx, larynx and oesophagus (Brune *et al.*, 1981). The site of tumour development is related to the route of PAH administration. For instance, dermal application induces skin tumours, inhalation and intratracheal instillation result in lung tumour; subcutaneous injection result in sarcomas and oral administration induces gastric tumours (Wall *et al.*, 1991). Tumour induction is however not restricted to the obvious sites. For example, lung tumours have been observed after oral administration or subcutaneous injection of benzo(a)pyrene to mice and liver tumours following intraperitoneal injection (Wall *et al.*, 1991). Table 6 gives the classification of PAHs based on their carcinogenicity. However, humans are almost always exposed to mixtures of PAHs predominantly adsorbed onto inhalable particles such as fine soot particles. Information on the synergetic effects on carcinogenicity of the PAHs in the mixtures is scarce. It is expected that biomass burning as fuel in open fire places emits a mixture of PAHs.

Table 6: Classification of PAHs according to their carcinogenicity.

Compound	Carcinogenicity
Acenaphthene	(?)
Acenaphthylene	No studies
Anthanthrene	+
Anthracene	-
Benz[a]anthracene	+
Benzo[b]fluoranthene	+
Benzo[j]fluoranthene	+
Benzo[ghi]fluoranthene	(-)
Benzo[k]fluoranthene	+
Benzo[a]fluorene	(?)
Benzo[b]fluorene	(?)
Benzo[ghi]perylene	-
Benzo[c]phenanthrene	+
Benzo[a]pyrene	+
Benzo[e]pyrene	?
Chrysene	+
Coronene	(?)
Cyclopenta[cd]pyrene	+
Dibenz[a,h]anthracene	+
Dibenzo[a,e]pyrene	+
Dibenzo[a,h]pyrene	+
Dibenzo[a,i]pyrene	+
Dibenzo[a,l]pyrene	+
Fluoranthene	(+)
Fluorene -	-
Indeno[1,2,3-cd]pyrene	+
5-Methylchrysene	+
1-Methylphenanthrene	(-)
Naphthalene	(?)
Perylene	(-)
Phenanthrene	(?)
Pyrene	(?)
Triphenylene	(-)

Key: +, positive; -, negative; ?, questionable; (), result derived from small database
Source: Community Bureau of Reference, (1992).

Four studies, one in Japan (Sobue, 1990) and three in China (Gao *et al.,* 1987; Liu *et al.,* 1993; Ko *et al.,* 1997) identified an association between biomass fuel use and lung cancer in women. Two studies in Brazil showed strong relationship between exposure to wood smoke and upper aero-digestive tract cancers (Franco *et al.,* 1989;

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Pintos *et al.,* 1998). In Western Province of Kenya, data collected by Kakamega Hospice, an organisation established in middle of the year 2009 to document and give physical, psychological, spiritual and bereavement support to medically diagnosed and reported cancer patients, at the Kakamega Provincial General Hospital (PGH), and their kin, in the region, between January and June, 2010 (Table 7), shows that cancer cases are rampant in the area with cancer of the oesophagus registering the highest number of diagnosed and reported cases each successive month. New cases of cancer of the cervix, prostate cancer and breast cancer cases were also diagnosed at the hospital and reported every month (Kakamega Hospice Registry, 2010). Almost all the patients were aged 40 years and above and either resided or had spent a significant part of their earlier lives in the rural areas. Cancer related death cases have also been documented, although the total number of cancer patients and cancer- related death in Kenya is unknown. However, potential causes of these cancer incidences are not known.

Table 7:   Cancer cases diagnosed at Kakamega PGH between January-June 2010.

Month	Type of cancer					Death
(Year 2010)	Oesophagus	Cervical	Prostate	Breast	Others	
January	4	5	1	4	1 (Anal)	0
February	10	4	1	4	1 (Bone)	0
March	11	2	0	3	1 (Eye)	1
April	17	3	1	4	0	3
May	15	3	2	3	0	0
June	13	6	5	1	2 (Stomach)	3

Source: Kakamega Hospice Registry, (2010).

CHAPTER 3

3.0 METHODOLOGY

3.1 Area of Study

Figure 1 are maps showing the sampling sites; figure 1a shows the distribution of districts in the Western Province of Kenya and the sampling areas, while figure 1b shows the distribution of the provinces in Kenya.
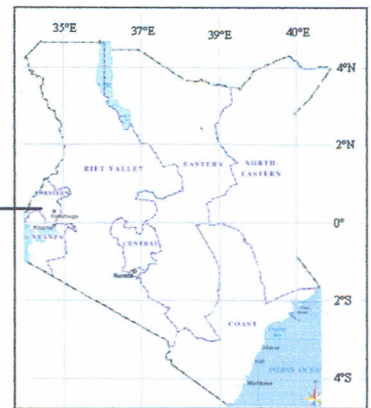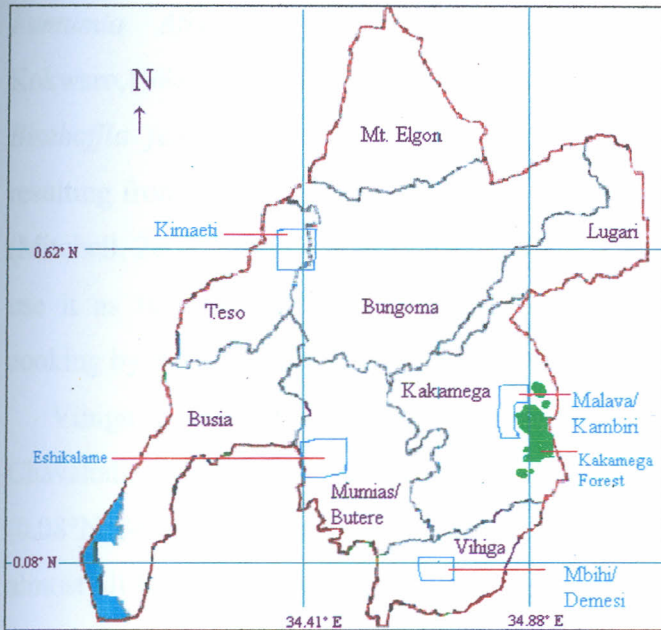

Figure 1a: Map of Western province of Kenya showing the sampling sites.

Figure 1b: Map of Kenya.

Figure 1: Maps showing the sampling areas

Western province in Kenya spans from Busia, (0.08°N, 34.12°E) to Mount Elgon, (1.12°N, 34.60°E) on the west, Kaimosi, (0.13°N, 34.91°E) to Turbo (0.64°N, 35.01°E) on the east and from Maseno, (0.01°N, 34.60°E) on the south to Mount Elgon (1.12°N, 34.60°E) on the north (Figure 1). The region supports a large variety of tree species in the equatorial rain forest (Kokwaro, 1988) and homesteads (Kindt et al., 2006). The people in the rural areas of this region are generally poor and there is prevalence of the grass-roofed traditional houses with tiny wooden windows and doors. Cooking and space warming is usually done in open fire places inside these houses, wood being the main form of fuel used.

The Kakamega Rain Forest lies within this region running from around Kaimosi, (0.13°N, 34.91°E) through Kambiri, (0.34°N, 34.84°E) and ends in Malava, (0.39°N, 34.86°E). This forest has a variety of perennial indigenous tree species which are a mixture of hardwoods and softwoods. The major tree species in the forest include; *Prunus Africana, Olea capencis, Markhania lutea, Croton megalocarpus, Maesopsis eminii, Entandrophragma angolense, Celtis midlbraedii, Aningeria altissima, Antiaris toxicaria, Diospyros abyssinica, Harungana madascarensis, Cordia Africana, Funtumia Africana* and *Trichilia emetica* (Mitchell, 2004; KIFCON,1994; Kokwaro,1988), among others. *Eucalyptus saligna, Cuprssus lustitanica, Pinus patula, Bischoffia javanica* and *Khaya anthotheca* have also been planted in the open spaces resulting from human encroachment and are strictly protected by the forest authorities (Mitchell, 2004). The population in these areas usually collect wood from the forest and use it as fuel (Mitchell, 2004). The hardwood species are generally preferred for cooking by the population because they burn for a longer time than the softwoods.

Vihiga and Sabatia areas, on the southern part of western province, span from Chavakali, (0.12°N, 34.73°E) to Nyang'ori (0.02°N, 34.74°E) and from Mahanga (0.08°N, 34.76°E) to Gisambai (0.13°N, 34.76°E). The areas are densely populated and almost all the perennial indigenous tree species are extinct in the sub-region. The most prevalent trees in these areas are the fast-growing exotic species mainly softwood which are planted in homesteads. These include *Grevillea robusta, Euphorbia tirucalli, Tithonia diversifolia, Thevelia peruviana, Leucaena leucocephala, Tarminallia browni, Mangitera Indica, Eriobotrya japonica, Balanites aegyptica, Passiflora edulis* and *Citrus Sinensis* (Kindt et al., 2006) among others. *Eucalyptus saligna* and *Eucalyptus globulus* (Kindt et al., 2006) are also grown in the sub-region, but according to the residents, they are usually preserved for building and timber.

Mumias on the central part of the province lies between Shianda (0.3021°N, 34.6066°E) and Buhuyi (0.36°N, 34.38°E), from Musanda (0.25°N, 34.50°E) to Makunda (0.48°N, 34.52°E). The area is mainly agricultural, where sugarcane grown on large tracks of land is used as raw materials in the Mumias Sugar Factory. Barely a few trees are allowed to grow in this area to pave way for the cash crop. As a result, the population here use the dry sugarcane residues and shrubs as fuel.

Finally, Teso to the north-west of Western province of Kenya spans from Chwele (0.73°N, 34.57°E) to Silisia (0.53°N, 34.36°E), and from Mt. Elgon (0.59°N, 34.99°E) to Kimaeti (0.59°N, 34.48°E). Human settlement in this sub-region is fairly sparse and hence there is still prevalence of the perennial indigenous tree species and shrubs in sections of virgin land spread across the sub-region. There are also some exotic tree species planted in some homesteads in the sub-region. However, a section of the rural population here is still deeply rooted in their traditional cooking methods. They use dry cow dung as fuel, in open fire places inside the houses, which simply smoulder with barely any flames. Due to the tedious exercise of lighting these balls of dry dung, the fires burn continuously even when no cooking is taking place.

3.2 Study design

A factorial two layout in a randomised complete block experimental design was used in this study with the predominant biomass type used in the sub-region as the main factor and the age of the house as the sub-factor. The experiments were set in a 4x3 arrangement with 4 replications. Only houses with grass thatched roofs within the sampling regions were included in this research. Also only houses exclusively using wood or dung as fuel in their entire lifespan for cooking and space warming were used in the study.

The houses with grass thatched roofs were classified into three according to the predominant wood type in the sub-region. In addition houses which predominantly use dry dung as fuel were included in the study for comparison,

Class 1: Houses in Malava/ Kambiri near Kakamega rain forest which predominantly use wood from perennial indigenous tree species, mostly hardwood, collected or harvested from the forest.

Class 2: Houses in Vihiga/ Sabatia, away from the rain forest, which predominantly use wood from the exotic fast-growing tree species, mostly softwood, grown in homesteads.

Class 3: Houses in Mumias, within the sugarcane growing belt, which predominantly use dry shrubs and crop residues as fuel.

Class 4: Houses in Teso where dry dung is predominantly used as fuel.

Houses in each class were further categorized according to the approximate ages of the houses: 0-5 years, 5-10 years and more than 10 years. This classification therefore formed the sampling units.

3.3 Sampling

3.3.1 Sampling sites

i) Kambiri in Malava - From (0.34°N, 34.84°E) to (0.34°N, 34.86°E), between (0.33°N, 34.85°E) and (0.38 °N, 34.84°E).

ii) Mbihi/Demesi in Vihiga/Sabatia - From (0.07°N, 34.71°E) to (0.09°N, 34.71°E), between (0.08°N, 34.68°E) and (0.08°N, 34.73 °E).

iii) Eshikalame in Mumias- From (0.32°N, 34.44°E) to (0.32°N, 34.47°E), between (0.27°N, 34.42°E) and (0.30°N, 34.36°E).

iv) Kimaeti in Teso- From (0.63°N, 34.43°E) to (0.63°N, 34.46°E), between (0.57°N, 34.44°E) and (0.64°N, 34.44°E).

3.3.2 Sample collection

Soot samples were collected from under the roofs of four randomly selected houses in each sampling unit into a clean aluminium foil, by hand in gloves, wrapped in black polythene bags, and labelled appropriately. The samples were then refrigerated at temperatures below -4°C until extraction was done. Table 8 below gives the number of samples collected from each site and the labelling. During sampling, protective gloves, goggles and mouth and nose masks were worn.

Table 8: Number of samples per site and labelling.

Biomass type / Age of House	Perennial indigenous Wood (HW)	Fast growing Exotic Wood (SW)	Shrubs and Crop Residues (SHCR)	Dry Dung (DD)
0 – 5 Years [3]	4 HW3	4 SW3	4 SHCR3	4 DD3
5 – 10 Years [2]	4 HW2	4 SW2	4 SHCR2	4 DD2
>10 Years [1]	4 HW1	4 SW1	4 SHCR1	4 DD1
Total number of samples = 48				

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3.4 Experimental

3.4.1 Extraction of PAHs from soot samples and purification

Extraction was based on the method used by Lalah and Kaigwara (2005), modified as explained below. PAHs were extracted from the soot samples in soxhlet apparatus with folded pre-extracted filter papers loaded with 30g dry weight of soot samples by refluxing with 300 mls of dichloromethane (≥99.8% purity), DCM, for 8 hours (Lalah and Kaigwara, 2005), with a fill-empty rate of two per hour. The DCM extract was then reduced in a rotary evaporator fitted with a suction pump to 50 mls, and 2 mls of this was then transferred into a pre-cleaned 24 cm long x 2.2 internal diameter column packed with silica gel (230-400 µm mesh GC grade) using HPLC grade n-hexane and anhydrous sodium sulphate added on the top to a height of 2 cm above the silica gel. Elution was achieved with HPLC grade n-hexane (Lalah and Kaigwara, 2005) followed by toluene/methanol (1:6 v/v) (Jonker and Koelmans, 2002). The toluene/methanol fraction was then reduced to 1 ml in a rotary evaporator, under pressure, for GC analysis (Lalah and Kaigwara, 2005). Each sample was extracted and purified three times and labelled appropriately. Preliminary analysis by GC –FID showed that the hexane fraction did not contain any PAHs.

The samples were stored in glass vials wrapped in aluminium foil and kept in a deep freezer at temperatures below -4°C at all stages until GC analysis (US EPA, 1990; US NIOSH, 1994).

3.4.2 Preparation of standards and samples for GC analysis

The samples were prepared for GC-analysis according to the US EPA method 8100/8015 (US EPA 1997a). Dodecane was used in this study as the internal standard. The spiking solution was prepared by dissolving 5 µl of dodecane stock solution (density 0.75g/ml) in 1ml dichloromethane (≥99.8% purity) and the mixture thoroughly shaken. Each of the 1ml cleaned samples was then spiked with 1µl of the internal standard solution. Calibration standards of 5 concentration levels; 0.1 ppm, 0.5 ppm, 1 ppm, 5 ppm and 10 ppm, respectively, were prepared from the authentic PAHs standards mix stock solution, from Dr. Ehrenstorfer, Augsburg, Germany, in HPLC grade n-hexane and 1µl of the internal standard spiked into 1ml of each of the calibration standards made. The standard samples were then stored in glass vials with

teflon-lined screw caps, wrapped in aluminium foil and kept in a deep freezer (at <10°C).

2,4-dinitrophenlyhydrazine (2,4-DNPH) was chosen as the surrogate standard (UNEP, 1992). One gram of GC-grade 2,4-DNPH was dissolved in 2 mls concentrated sulphuric acid and the solution made up to 1litre with distilled water. The solution was further diluted by dissolving 25 mls of the solution in distilled water to 1litre and 2 mls of this final solution added to 30 g of soot sample, thoroughly mixed and kept in a dark, dry cabinet for 24 hours before being subjected to same analytical procedure as the rest of the samples. This recovery study was replicated three times.

3.4.3 GC-Analysis of the samples, characterisation and confirmation of the PAHs
3.4.3.1 Analysis by GC-FID

The analysis of the samples was based on the method used by Lalah and Kaigwara (2005). The spiked samples, including the calibration standards and recovery study samples were analysed using a Shimadzu GC-17A version 3 instrument with a flame ionisation detector (FID). Separation was achieved using a DB-5 capillary column, 30m long x 0.25 mm internal diameter (ID) and a film thickness of 0.25µm. The GC was run in splitless injection mode with a total flow rate of 3ml/min, linear velocity of 27.1 cm/sec and a pressure of 77kPa using N_2 as the carrier gas. The oven temperature was maintained at 60°C for 1 minute, then increased to 180°C at the rate of 3.0°C/min and maintained there for 2 minutes. Finally it was then increased to 300°C at the rate of 5.0°C/minute and maintained there for 2 minutes. The injector temperature was set at 250°C and the column temperature at 60°C. Only 1µl of each sample was injected into the GC for analysis

3.4.3.2 Analysis by GC-MS

The analysis of the samples was done according to the US EPA method 8270C (US EPA 1997b). The samples were analysed using Agilent Technologies 7890A GC-MS system and separation achieved using a HP-5MS, (5% methyl silox), (30 m × 250 µm × 0.25 µm) column. The injector was set in the splitless mode with a total flow rate of 10.2 ml/min, septum purge flow rate of 3 ml/min and a pressure of 8.8271 psi using N_2 as the carrier gas. The oven equilibration time was 1min. The oven temperature was

maintained at 35°C for 5 min, and then increased to 280°C at the rate of 10°C/min and maintained there for 10.5 min. Finally the temperature was increased to 285°C at the rate of 50°C/min and maintained there for 9.9 min. The heater temperature was set at 250°C. Only 1μl of sample was injected for analysis.

3.4.3.3 Characterisation of PAHs and confirmation

The PAHs were characterised basing on the method used by Lalah and Kaigwara, (2005). The PAHs were identified by comparing peak retention times on the chromatograms of authentic standards with the retention times indicated on the gravimetric certificate supplied with authentic standards (Lalah and Kaigwara, 2005), bought from Dr. Ehrenstorfer, Germany. The PAHs in the extracts were then identified by comparing the peak retention times in the chromatograms of the extracts, with those in the chromatograms of the authentic standards. The authentic standards were also analysed by GC-MS and the retention times of the analytes in the samples in GC-MS analysis matched with the retention times of the PAHs in the authentic standards mix for further identification.

The spectra of the identified analytes were generated and confirmation of identity of the analytes was done according to US-EPA method 8270C (US EPA, 1997b). This involved calculation, by the data system analysis, of a similarity index, match factor or purity between the unknown spectrum and library (reference) spectra using NIST/EPA/NIH MASS SPECTRAL LIBRARY (NIST 05) and NIST MASS SPECTRAL SEARCH PROGRAM Version 2.0d.

3.4.4 Determination of relative response factors (RRF)

The relative response factors of the analytes were determined according to the US EPA method 8100/8015 (US EPA 1997a). The peak area responses of the identified analytes were tabulated against their concentration and that of the internal standard, and the relative response factors (RRF) for each of the PAHs calculated using the equation 1 below.

$$RRF = \frac{A_S \times C_{IS}}{A_{IS} \times C_S} \qquad (1)$$

Where,

A_S = Peak Area for the target PAH measured

A_{IS} = Peak Area for the internal standard

C_{IS} = Concentration of the internal standard

C_S = Concentration of the target PAH

3.4.5 Quantification of PAHs

Those samples whose analyte concentrations were above the linear range were diluted and the dilution factor (D) noted (Lalah and Kaigwara, 2005). Quantification of the PAHs concentration was then done using the internal standard method, according to the US EPA method 8100/8015 (US EPA 1997a). The equation 2 below was used in the calculations:

$$\text{Concentration } (\mu g/g) = \frac{A_S \times W_{IS} \times D \times F}{A_{IS} \times RRF \times W_S} \qquad (2)$$

Where,

A_S = Peak Area for the analyte in the sample.

A_{IS} = Peak Area for the internal standard in the sample.

W_{IS} = Amount (μg) of internal standard (dodecane) added to the sample.

D = Dilution factor if dilution was made on the sample prior to analysis. If no dilution was made, D = 1, dimensionless.

W_S = Weight of soot extracted, in g, dry weight.

F = Factor if the samples are split in half for analyses.

All the samples had a split factor (F) of 25 since only 2ml of the 50ml extracted were taken for clean-up.

The percentage recovery 2,4-DNPH was calculated using equation 3 below.

$$\text{\% Recovery of 2,4-DNPH} = \frac{\text{Amount of (µg/g) in the Spiked sample} \;\_\_\; \text{Amount (µg/g) in unspiked sample}}{\text{Amount (µg/g) spiked}} \times 100 \qquad (3)$$

The mean percentage recovery rate was then used to correct all the PAHs concentrations calculated for recovery.

3.4.6 Statistical analysis of data

Separation of means and analysis of variance (ANOVA) of the data was done using the MSTAT-C programme for a two factor randomized complete block design (Appendix 1). Statistical analysis was based on a 4x3 layout. In some cases the levels of some PAHs in soot samples from houses that predominantly use shrubs and crop residues were below the minimum detection limit (MDL). These include naphthalene, phenanthrene and dibenzo(a,h)anthracene. In such cases a 3x3 arrangement was used in statistical analysis (Appendix 2). In this study, analyte concentration below the MDL was assumed to have a concentration of zero. The PAHs whose concentration was below the detection limit in more than 20% of the samples were excluded from statistics. These include benzo(k)fluoranthene, benzo(b)fluoranthene and indeno(1,2,3-c,d)anthracene.

The data was then subjected to the ANOVA F-test ($P\leq0.05$) (Gomez and Gomez, 1983; Motulsky, 1999; Hopkins, 2000), to test the hypothesis. Pair-wise comparisons of the means were done using the LSD test (Gomez and Gomez, 1983) to determine if the variation in the mean concentration of each PAH in the various biomass types and in different house age groups was significant. The interactions in the patterns of accumulation of the PAHs from various wood types with time were also assessed.

3.4.7 Data transformation

Analysis of variance of the data in the original scale gave relatively large variation coefficients. The data was then subjected to the logarithmic transformation, $X^1 = Ln(X+1)$: where, X is the concentration of PAH in µg/g of soot and Ln is the natural logarithm (Hopkins, 2000; Osborne, 2002). This was to achieve homogeneity in the

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variance of the experimental errors and to ensure that the errors are normally distributed (Gomez and Gomez, 1983; Motulsky, 1999; Hopkins, 2000; Osborne, 2002).

The data in the logarithmic scale was then subjected to ANOVA, with MSTAT-C programme, for a two factor randomized complete block design to separate the means (Appendix 3 and Appendix 4) and further subjected to the F-test (P≤0.05) (Gomez and Gomez, 1983; Motulsky, 1999; Hopkins, 2000) and the LSD-test (P≤0.05) (Gomez and Gomez, 1983) as the data in the original scale. The results from the comparison of means in the transformed data were then used to make inferences about the data in the original scale (Gomez and Gomez, 1983; Hopkins, 2000).

CHAPTER 4

4.0 RESULTS AND DISCUSSION

4.1 Identity and confirmation of PAHs in soot

Fifteen out of the target sixteen US EPA priority carcinogenic PAHs were identified in soot samples using retention times from soot samples by GC-FID analysis (Figure 2) and confirmed by GC-MS spectra (Appendix 5) by marching the GC-MS spectra of each of the analyte in the sample extracts with the spectra in the GC-MS internal reference library. These were naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo(a)anthracene, chrysene, benzo(a)pyrene, dibenzo(a,h)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene and indeno(1,2,3-c,d)anthracene. Previous studies on PAHs emitted during residential biomass burning, (Jenkins *et al.*, 1996; Hueglin *et al.*, 1997; McDonald *et al.*, 2000; Zou *et al.*, 2003) characterised PAHs in the on-flight particulate phase or in indoor dust and gaseous phase wood combustion products. This study characterized PAHs in accumulated soot under the roofs of the rural houses.

A number of the lower molecular weight PAHs were found in the particulate phase, a finding that was also reported by Zou et al., (2003). According to Jenkins *et al.*, (1996), lower molecular weight PAHs (molecular weight ≤ 200) are generally formed and detected in gaseous phase. The results of this study indicate that the PAHs emitted in gaseous phase during wood combustion probably cool and deposit onto soot particles as the smoke from wood burning in these houses take a long time to find its way through the grass thatched roofs due to poor ventilation and limited movement of air indoors. This is in agreement with Venkarataman *et al.* (2002) who suggested the possibility of surface adsorption of lower molecular weight PAHs on to particulate matter. The findings of this study indicate that accumulated soot under the roofs of rural houses is an indoor repository for PAHs, emitted during residential burning of biomass fuel.

The relatively high indoor temperatures is likely to cause vaporization of the adsorbed lower molecular weight PAHs, increasing their vapour phase concentration in indoor air, even at times when no cooking or space heating is taking place, and hence exposing the people indoors to much higher levels of gaseous carcinogenic PAHs than outdoors.

Figure 2 shows some of the chromatograms of obtained from gas chromatography: Figure 2a is a chromatogram of an extract of soot from houses that predominantly use shrubs and crop residues, figure 2b is a chromatogram of an extract of soot from houses that predominantly use wood from perennial indigenous trees, figure 2c is a chromatogram of an extract of soot from houses that predominantly use dry cow dung, while figure 2d is a chromatogram of an extract of soot from houses that predominantly use wood from the fast growing exotic trees.
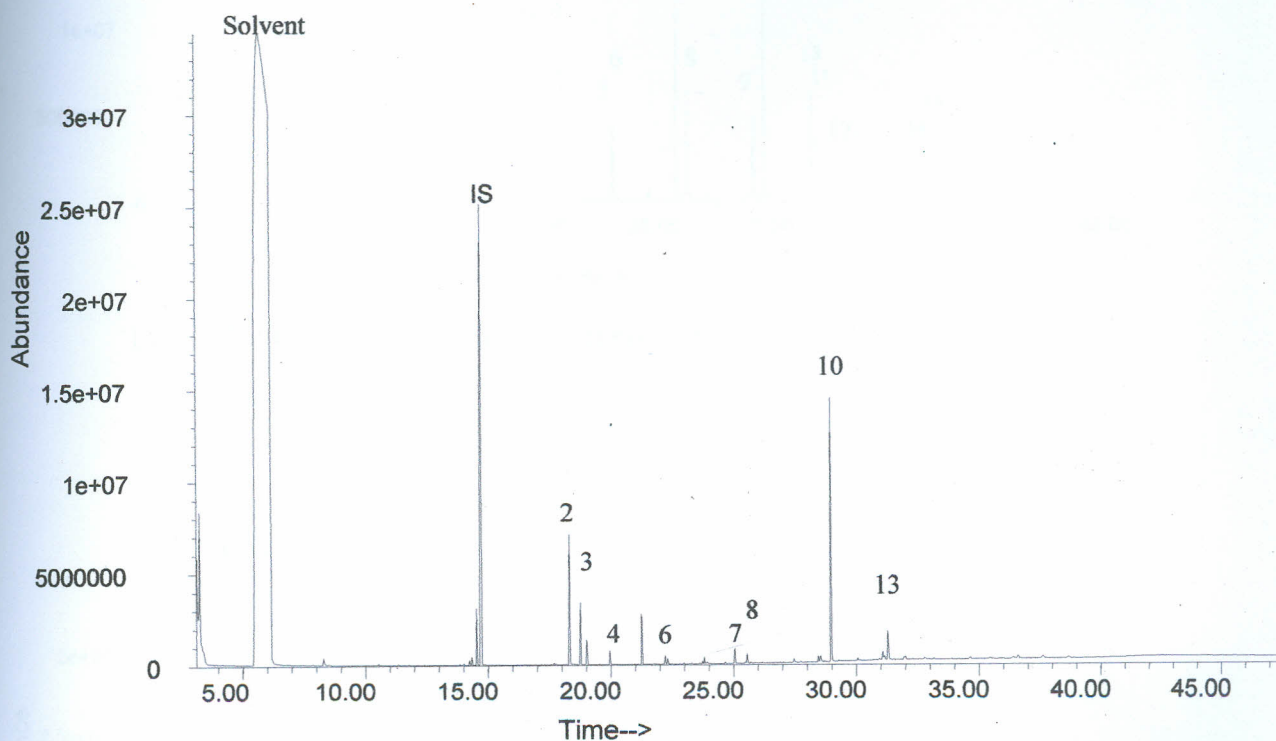

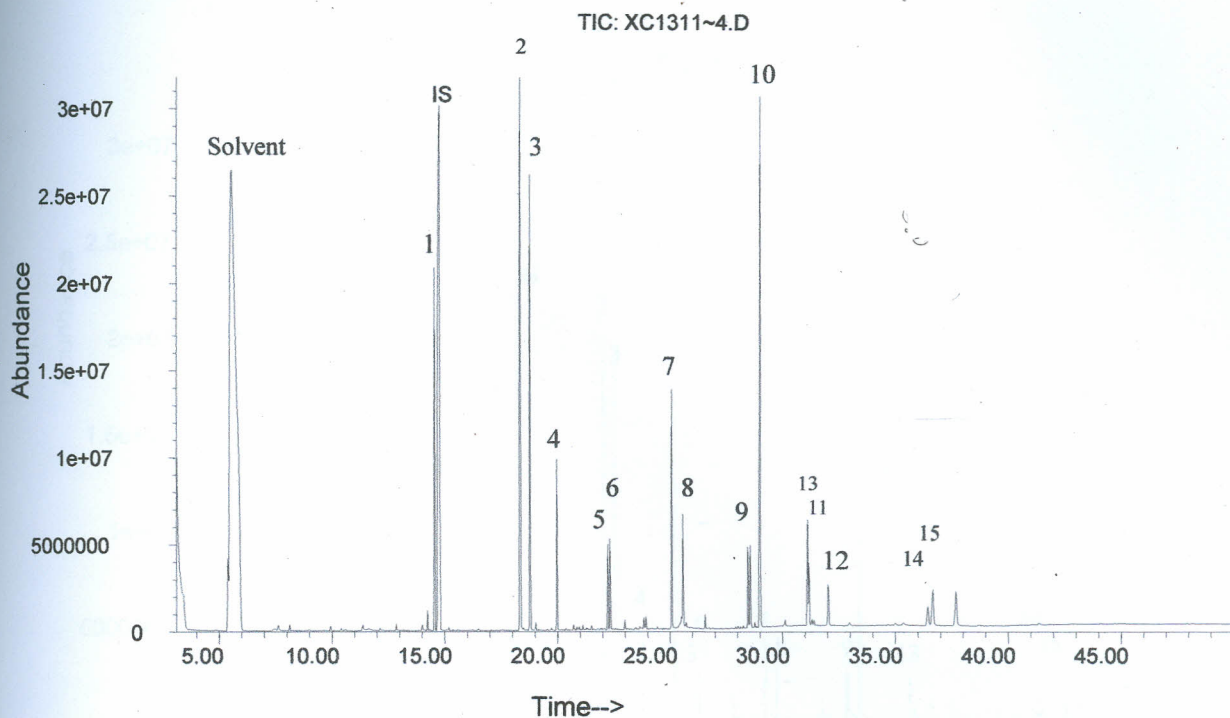Figure 2a: GC chromatogram of sample SHCR1A1 (Mumias)

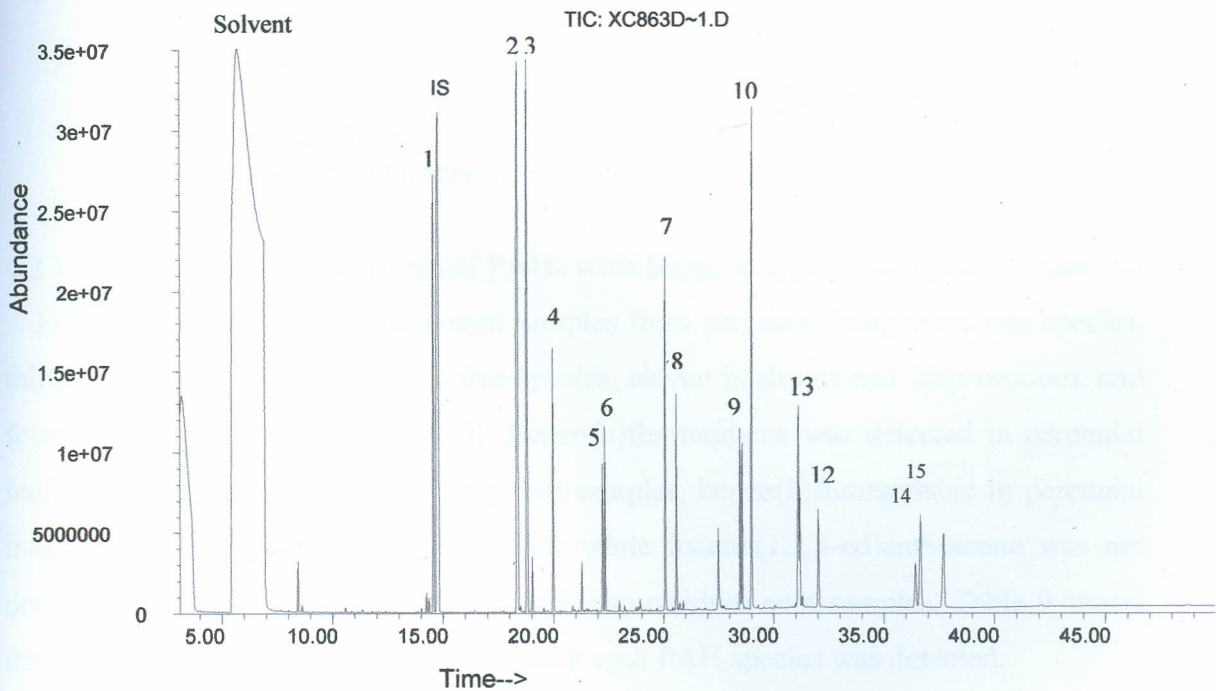Figure 2b: GC chromatogram of sample HW2B3 (Malava/Kambiri)

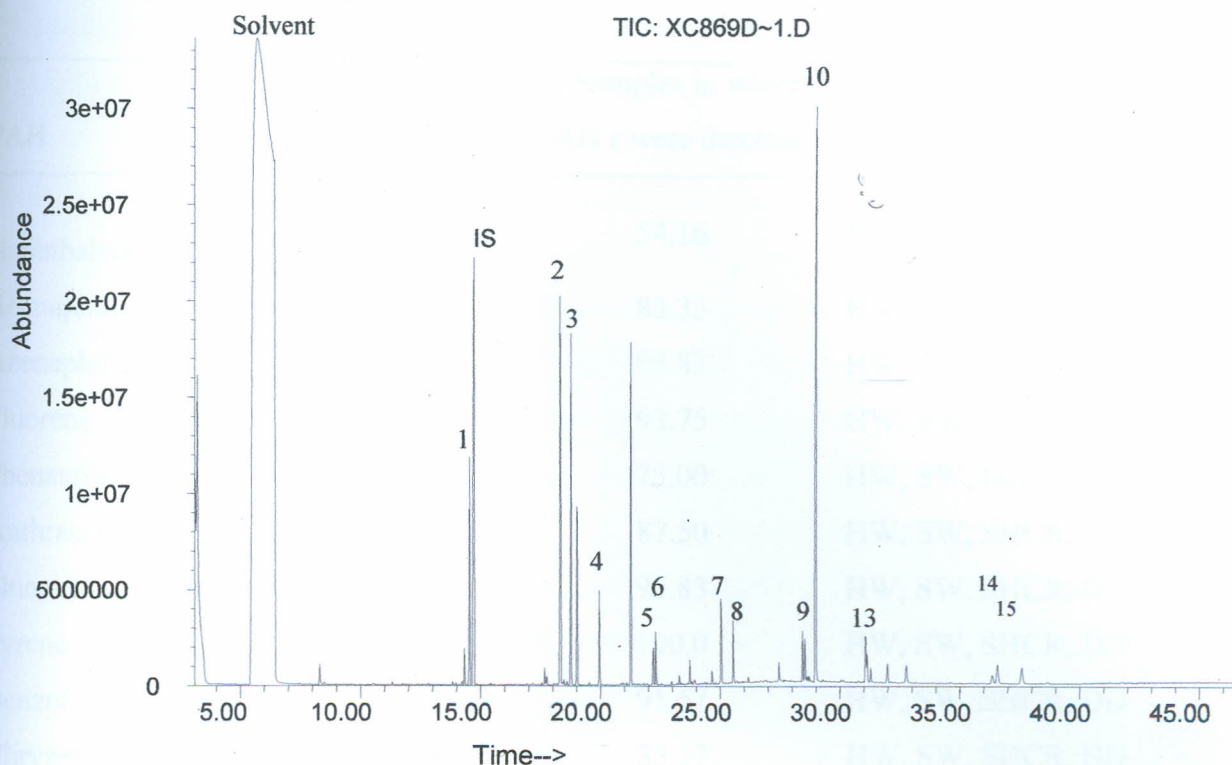Figure 2c: GC chromatogram of sample DD3C1 (Teso)

Figure 2d: GC chromatogram of sample SW1A2 (Vihiga/Sabatia)

Figure 2: GC chromatograms for some of the samples; IS = Internal standard; 1 = naphthalene; 2 = acenaphthylene; 3 = acenaphthene; 4 = fluorene; 5 = phenanthrene; 6 = anthracene; 7 = fluoranthene; 8 = pyrene; 9 = benzo(a)anthracene; 10 = chrysene; 11 = benzo(b)fluoranthene; 12 = benzo(k)fluoranthene; 13 = benzo(a)pyrene; 14 = indeno(1,2,3-cd)anthracene; 15 = dibenzo(a,h)anthracene.

4.2 Variations in concentrations of PAHs with biomass types and age of houses

Fifteen PAHs were detected in soot samples from perennial indigenous tree species, thirteen in the fast-growing exotic tree species, eleven in shrubs and crop residues, and fourteen in dry cow dung (Table 9). Benzo(b)fluoranthene was detected in perennial indigenous tree species and dry dung soot samples, benzo(k)fluoranthene in perennial indigenous tree species soot sample only while indeno(1,2,3-cd)anthracene was not detected any of the soot of the shrubs and crop residues soot samples. Table 9 shows the percentages of the soot samples in which each PAH species was detected.

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Table 9: Percentage of soot samples in which each PAH was detected.

PAH	Number*	% Samples in which PAH s were detected	Biomass type in which PAH is detected
Naphthalene	1	54.16	HW,SW,DD
Acenaphthylene	2	83.33	HW, SW, SHCR, DD
Acenaphthene	3	95.83	HW, SW, SHCR, DD
Fluorene	4	93.75	HW, SW, SHCR, DD
Phenanthrene	5	75.00	HW, SW, DD
Anthracene	6	87.50	HW, SW, SHCR, DD
Fluoranthene	7	95.83	HW, SW, SHCR, DD
Pyrene	8	100.0	HW, SW, SHCR, DD
Benzo(a)anthracene	9	91.67	HW, SW, SHCR, DD
Chrysene	10	83.33	HW, SW, SHCR, DD
Benzo(b)fluoranthene	11	2.08	HW
Benzo(k)fluoranthene	12	12.50	HW, DD
Benzo(a)pyrene	13	89.58	HW, SW, SHCR, DD
Indeno(1,2,3-c,d)anthracene	14	16.67	HW, SW, DD
Dibenzo(a,h)anthracene	15	60.42	HW, SW, DD

HW-Perennial indigenous tree species; SW- Fast growing exotic tree species; SHCR- shrubs and crops residues; DD- dry cow dung.

* PAHs peaks labelled in figure 2.

These results revealed that the identities of the PAHs emitted from residential burning of biomass as fuel depended on the biomass type used. Previous studies, (Jenkins *et al.*, 1996; Hueglin *et al.*, 1997; Rogge *et al.*, 1998; Oanh *et al.*, 1999; McDonald *et al.*, 2000; Zou *et al.*, 2003; US EPA, 1998) have also reported that the characterisation of PAHs emitted from different biomass burning in both gaseous and particulate phases depend mainly on biomass type used. Exposure to PAHs from residential biomass fuel burning is a significant health hazard. The toxicity and

carcinogenic potency of various PAHs are documented, but the synergetic effects on carcinogenicity of the PAHs in the mixtures are unknown. Records from the Kakamega Hospice (Table 7) showed that cases of cancer patients were rampant in the region (Kakamega Hospice Registry, 2010). The results show that, of the four biomass types considered in this study, use of wood from indigenous tree species as fuel exposes people indoors to the highest number of carcinogenic PAHs.

Benzo(a)pyrene, which is both carcinogenic and mutagenic (Community Bureau of Reference, 1992) and commonly used as a biomarker of PAH pollution, was detected across all the four biomass types in this study. These results indicate that biomass burning as fuel in open fire places in the traditional rural houses is a major source of PAH pollution in the rural areas. While in this study benzo(ghi)perylene was not detected in all the samples collected, Lobscheid and Mckone (2004) reported high amount of benzo(ghi)perylene from wood combustion in external combustion boilers.

Table 10 shows the variations in levels of PAHs in accumulated soot under the roofs of the traditional rural houses in Western Province of Kenya due to variations in biomass types predominantly used as fuel for cooking and space warming and age of houses. The PAHs whose concentrations were below the minimum detection limit in more than 20% of the samples analysed (Table 9), were not included in the analysis of variance. These were benzo(b)fluoranthene, in 2.08% of samples, benzo(k) fluoranthene in 12.50% of samples and indeno(1,2,3-c,d)anthracene in 16.67% of the samples. The variations in the concentrations of twelve PAHs in soot from houses, of various ages, that use various biomass types were therefore analysed.

Table 10: Variations in levels (µg/g of soot) of PAHs due to wood types and age of houses.

PAH	Age (Years)	Wood Type				Mean (Age)
		HW	SW	SHCR	DD	
Naphthalene	0–5	1.014	0.000	bdl	1.939	0.985
	5–10	9.294	2.547	bdl	10.996	7.612
	› 10	15.928	11.706	bdl	16.690	14.775
	Mean (wood type)	8.746	4.751	bdl	9.880	
	CV (%)		63.250			
	LSD(P≤0.05)		8.344			8.344
Acenaphthylene	0–5	4.503	5.064	0.000	11.805	5.342
	5–10	7.532	24.004	1.984	25.895	14.851
	› 10	11.721	15.879	4.510	47.263	19.843
	Mean (wood type)	7.916	14.982	2.165	28.321	
	CV (%)		54.120			
	LSD(P≤0.05)		9.382			6.576
	Interaction		12.496			
Acenaphthene	0–5	17.206	7.613	0.387	46.670	17.969
	5–10	58.807	26.982	0.989	105.028	47.952
	› 10	122.424	50.507	1.778	176.820	87.882
	Mean (wood type)	66.146	28.367	1.052	109.506	
	CV (%)		65.530			
	LSD(P≤0.05)		43.644			51.113
	Interaction		58.127			
Fluorene	0–5	2.274	1.166	0.156	10.722	3.580
	5–10	18.591	18.587	0.404	33.473	17.764
	› 10	22.372	29.156	1.141	92.229	36.224
	Mean (wood type)	14.412	16.303	0.567	45.475	
	CV (%)		41.110			
	LSD(P≤0.05)		10.248			12.002
	Interaction		13.650			
Phenanthrene	0–5	3.965	0.900	bdl	5.568	3.477
	5–10	7.513	2.034	bdl	5.717	5.088
	› 10	13.487	5.580	bdl	13.353	10.807
	Mean (wood type)	8.321	2.838	bdl	8.212	
	CV (%)		74.190			
	LSD(P≤0.05)		8.416			8.416
Anthracene	0–5	10.326	3.243	0.212	20.227	8.502
	5–10	62.023	21.057	0.984	70.465	38.633
	› 10	68.835	41.070	5.173	149.441	66.130
	Mean (wood type)	47.062	21.79	2.123	80.044	
	CV (%)		75.920			
	LSD(P≤0.05)		37.236			43.608
	Interaction		49.598			

PAH	Age (Years)	Wood Type				Mean (Age)
		HW	SW	SHCR	DD	
Fluoranthene	0–5	10.298	1.199	0.639	15.666	6.950
	5–10	9.267	6.425	0.667	31.105	11.866
	› 10	22.612	13.789	1.943	53.796	23.035
	Mean (wood type)	14.059	7.138	1.083	33.522	
	CV (%)		39.760			
	LSD(P≤0.05)		7.205			8.439
	Interaction		9.597			
Pyrene	0–5	12.432	9.020	1.142	38.660	15.313
	5–10	61.929	13.126	2.445	60.523	34.506
	› 10	104.503	31.954	7.053	134.559	69.517
	Mean (wood type)	59.621	18.033	3.547	77.914	
	CV (%)		57.820			
	LSD(P≤0.05)		29.880			34.993
	Interaction		39.799			
Benzo(a)anthracene	0–5	2.997	6.376	0.538	13.134	5.761
	5–10	14.754	15.084	0.923	64.919	23.920
	› 10	40.405	21.063	2.128	162.643	56.560
	Mean (wood type)	19.386	14.174	1.196	80.232	
	CV (%)		68.310			
	LSD(P≤0.05)		25.508			29.872
	Interaction		33.975			
Chrysene	0–5	22.229	6.787	0.439	24.074	13.382
	5–10	37.516	17.708	1.411	52.475	27.278
	› 10	44.069	31.222	6.953	72.256	38.625
	Mean (wood type)	34.604	18.573	2.934	49.602	
	CV (%)		57.040			
	LSD(P≤0.05)		19.582			22.933
Benzo(a)pyrene	0–5	8.642	7.021	0.373	29.865	11.475
	5–10	21.603	13.405	1.154	101.260	34.355
	› 10	61.640	45.566	1.479	194.213	75.725
	Mean (wood type)	30.629	21.997	1.002	108.446	
	CV (%)		38.220			
	LSD(P≤0.05)		20.119			23.562
	Interaction		26.798			
Dibenzo(a,h) Anthracene	0–5	1.641	0.000	bdl	3.486	1.709
	5–10	2.143	0.203	bdl	3.903	2.083
	› 10	4.802	0.449	bdl	9.533	4.928
	Mean (wood type)	2.862	0.217	bdl	5.641	
	CV (%)		77.330			
	LSD(P≤0.05)		3.948			3.948

HW-Perennial indigenous trees; SW- Fast growing exotic trees; SHCR- shrubs and crops residues; DD- dry cow dung; LSD – least significant difference; CV - coefficient of variance; bdl – below detection limit.

4.2.1 Variance of PAHs concentrations in soot

Factorial analysis of variance of the data gave relatively high coefficients of variation (CV %) as shown in Table 10; an indication of heterogeneous variance of the experimental errors. This high variability within experimental units could have resulted from the grouping of the houses in large age clusters: 0-5 years, 5-10 years and more than 10 years. For instance, in the cluster 5-10 years, houses which are 5 years old may have been sampled with those which are 9-10 years old as replicates. Similarly, in the cluster more than 10 years old, houses which are 11 years old may have been sampled with those which are more than 18 years old as replicates. The high variability might have also been caused by variation in composition of the different types of biomass (White, 1986), used as fuel, which affects the oxidation state of the biomass (Susott *et al.,* 1975), and probably the rate of combustion of each of the various biomass types, as well as the identity, composition and levels of the PAHs emitted. As a result some biomass types emitted much higher levels of PAHs than others over the same period of time.

In this study, the various PAHs concentration range within replicates showed a linear relationship to the mean concentrations of the PAHs in the replicates, that is, the range increased proportionally with the mean. This was an indication that the experimental errors had heterogeneous variance that is functionally related to the mean (Gomez and Gomez, 1983). One of the assumptions of parametric statistics is that the data being compared have constant/equal variance, that is, experimental errors in the data have common or homogenous variance (Gomez and Gomez, 1983; Hopkins, 2000; Osborne, 2002). Hence two means can not be compared when the variances are significantly different. The second assumption is that the experimental errors are normally distributed (Gomez and Gomez, 1983; Hopkins, 2000; Osborne, 2002). Hence, transformation of the data was necessary to achieve homogeneity in the variance of the experimental errors and to normalize their distribution.

When the data was subjected to the natural logarithmic transformation and subjected to ANOVA, the resulting data showed much lower coefficient of variation percentages (Table 11); an indication that the variance of experimental errors in the transformed data was homogenous and the errors normally distributed. Table 11 shows the variation of the logarithm transformed data due to biomass type and house age.

Table 11: Variations in levels [log (X+1) µg/g of soot] of PAHs due to wood types and age of houses in the logarithm transformed data

PAH	Age (Years)	Wood type				Mean (age)
		HW	SW	SHCR	DD	
Naphthalene	0–5	0.545	0.000	bdl	1.027	0.524
	5–10	2.234	0.000	bdl	2.451	1.562
	› 10	2.798	1.614	bdl	2.758	2.390
	Mean (wood type)	1.859	0.538		2.078	
	CV (%)		38.350			
	LSD(P≤0.05)		1.005			1.005
	Interaction		0.990			
Acenaphthylene	0–5	1.623	1.441	0.000	2.478	1.386
	5–10	1.756	3.185	0.949	3.200	2.273
	› 10	2.522	3.073	1.437	3.855	2.722
	Mean (wood type)	1.967	2.567	0.795	3.178	
	CV (%)		32.210			
	LSD(P≤0.05)		0.890			1.042
Acenaphthene	0–5	2.542	1.618	0.306	3.816	2.071
	5–10	4.022	3.194	0.673	4.662	3.138
	› 10	4.731	3.023	0.979	5.057	3.447
	Mean (wood type)	3.765	2.612	0.653	4.512	
	CV (%)		26.710			
	LSD(P≤0.05)		1.001			1.173
Fluorene	0–5	1.120	0.668	0.000	2.422	1.053
	5–10	2.889	2.955	0.310	3.537	2.423
	› 10	3.004	3.371	0.735	4.520	2.907
	Mean (wood type)	2.338	2.332	0.348	3.493	
	CV (%)		15.520			
	LSD(P≤0.05)		0.429			0.503
Phenanthrene	0–5	1.874	0.632	bdl	1.865	1.457
	5–10	1.733	1.084	bdl	1.993	1.604
	› 10	2.488	1.873	bdl	2.610	2.324
	Mean (wood type)	2.032	1.196		2.156	
	CV (%)		31.060			
	LSD(P≤0.05)		0.829			0.829
	Interaction		1.202			
Anthracene	0–5	2.367	1.085	0.187	2.953	1.648
	5–10	4.137	3.042	0.497	4.152	2.957
	› 10	3.353	3.731	1.525	4.896	3.376
	Mean (wood type)	3.286	2.619	0.736	4.000	
	CV (%)		26.100			
	LSD(P≤0.05)		0.902			1.056

PAH	Age (Years)	HW	SW	SHCR	DD	Mean (age)
			Wood type			
Fluoranthene	0–5	2.317	0.768	0.494	2.793	1.593
	5–10	1.932	1.956	0.469	3.687	2.011
	› 10	3.043	2.678	1.032	4.244	2.749
	Mean (wood type)	2.431	1.801	0.665	3.575	
	CV (%)			23.580		
	LSD(P≤0.05)			0.628		0.759
	Interaction			0.864		
Pyrene	0–5	2.488	2.263	0.717	3.679	2.287
	5–10	4.063	2.629	1.187	4.081	2.990
	› 10	4.605	3.410	2.059	4.842	3.729
	Mean (wood type)	3.719	2.768	1.321	4.200	
	CV (%)			11.990		
	LSD(P≤0.05)			0.467		0.548
	Interaction			0.623		
Benzo(a)anthracene	0–5	1.315	1.669	0.396	2.541	1.480
	5–10	2.735	2.754	0.515	4.170	2.544
	› 10	3.676	3.029	1.131	5.046	3.221
	Mean (wood type)	2.575	2.484	0.681	3.919	
	CV (%)			20.920		
	LSD(P≤0.05)			0.656		0.769
	Interaction			0.874		
Chrysene	0–5	3.110	1.339	0.000	3.157	1.901
	5–10	3.646	2.401	0.669	3.189	2.476
	› 10	3.728	3.434	2.055	4.258	3.369
	Mean (wood type)	3.494	2.391	0.908	3.535	
	CV (%)			37.930		
	LSD(P≤0.05)			1.072		1.490
Benzo(a) Pyrene	0–5	2.230	1.742	0.000	3.393	1.841
	5–10	3.060	2.630	0.574	4.566	2.707
	› 10	3.765	3.786	0.799	5.271	3.405
	Mean (wood type)	3.018	2.719	0.458	4.410	
	CV (%)			17.610		
	LSD(P≤0.05)			0.607		0.710
Dibenzo(a,h) Anthracene	0–5	0.945	0.000	bdl	1.297	0.747
	5–10	1.009	0.179	bdl	1.108	0.765
	› 10	1.735	0.356	bdl	2.274	1.455
	Mean (wood type)	1.230	0.178		1.560	
	CV (%)			59.820		
	LSD(P≤0.05)			1.039		1.039

HW-Perennial indigenous tree species; SW- Fast growing exotic tree species; SHCR- shrubs and crops residues; DD- dry cow dung; LSD – least significant difference; CV - coefficient of variance; bdl – below minimum detection limit.

4.2.2 Variation in levels of PAHs with biomass type

The concentrations of low molecular weight (MW) PAHs (MW≤228) were above the minimum detection limit in a large percentage, 83.33-100%, of the soot samples, across all the biomass types under investigation, except naphthalene, 54.16%, and phenanthrene, 75%, whose concentration in shrubs and crop residues soot samples were below the minimum detection limit (Table 9). This is consistent with the findings of Rogge *et al.,* (1998) and Venkataraman *et al.,* (2002), who reported that low molecular weight (202 ≤ MW≤ 228) PAHs predominate wood smoke profiles regardless of the combustion appliance design, fuel class or combustion environment. The mean concentrations range (in µg/g of soot) of these low molecular weight PAHs (MW≤228) in soot samples from the traditional rural houses were; 4.751 – 9.880 µg/g naphthalene, 2.165 – 28.321µg/g acenaphthylene, 1.052 – 109.506 µg/g acenaphthene, 0.567 – 45.475 µg/g fluorene, 2.838 – 8.212 µg/g phenanthrene, 2.123 – 80.044 µg/g anthracene, 1.083 – 33.522 µg/g fluoranthene, 3.547 – 77.914 µg/g pyrene, 1.196 – 80.232 µg/g benzo(a) anthracene and 2.934 – 49.602 µg/g chrysene (Table 10).

Soot samples from the houses that use shrubs and crop residues, gave the least mean concentrations for all the PAHs while those from houses that use dry dung gave the highest mean PAHs concentrations. These particle phase concentrations of the lower molecular weight (MW≤202) PAHs, naphthalene, acenaphthylene, acenaphthene, fluorene, anthracene and phenanthrene were much higher than would be expected, with the concentration range of some almost the same as that of the PAHs that usually predominate the particle phase (202 ≤ MW≤ 228). In previous studies high concentrations of these PAHs were reported mainly in gas-phase. Kamens *et al.,* (1986) reported that PAHs with MW≤ 202 partition between gas and particle phase. Hays *et al.,* (2003) reported that gas-phase PAHs adsorbed on to uncoated quartz filter media causing a trend of higher concentrations of lower molecular weight particle-phase PAHs. This implies that the lower molecular weight gaseous phase PAHs in smoke, from biomass fuel burning as fuel, most likely, adsorb on to the surface of the accumulated soot particles deposited under the roofs of the traditional rural houses, as the smoke slowly finds its way through the grass thatched roofs, hence increasing the particle phase concentration of the PAHs.

The concentration of benzo(a)pyrene was generally high, 1.002 – 108.446 µg/g of soot, across all the biomass types used in this study (Table 10). This was consistent with Hays et al (2003) who reported that benzo(a)pyrene is a major component of, and predominate the particle phase PAHs, in wood smoke, regardless of the combustion appliance design, fuel class or combustion environment. The mean concentrations of dibenzo(a,h)anthracene was low, in the range of 0.217 – 5.641 µg/g and below the minimum detection limit in shrubs and crop residues soot samples. The concentrations of benzo(b)fluoranthene, benzo(k)fluoranthene and indeno(1,2,3-cd)pyrene were generally below the minimum detection limit in most samples but just above the detection limit in some hardwood, softwood and dry dung soot samples (Table 10). PAH growth during biomass combustion is a multi-step process dependent on pyrolysis, progressive aromatization and surface reaction yields (Violi et al., 1999). The polymerization reactions of 2- and 3-ring monomeric PAHs, in rich high temperature combustion, control formation of high molecular weight PAHs ($MW \geq 252$) and soot (Violi et al., 1999). The results of this study indicate that the conditions in which the various types of biomass burn in the open fire places in the traditional rural houses, probably, does not support these polymerization reactions to a large extent, leading to formation of low levels of the high molecular PAHs across all the biomass types considered. This study has shown that biomass burning, as fuel, in these poorly ventilated grass thatched rural houses is a major source of particulate matter and associated PAHs.

In general, of the four biomass types in this study, burning of dry cow dung as fuel emitted the highest levels ($P \leq 0.05$) of PAHs, followed by wood from the perennial indigenous tree species. Wood from the exotic fast-growing tree species emit lower levels ($P \leq 0.05$) of PAHs than wood from the perennial indigenous tree species while burning of shrubs and crop residues as fuel was found to emit the least amount ($P \leq 0.05$) of PAHs (Table 11). The differences in levels of naphthalene, acenaphthene, phenanthrene, fluoranthene, pyrene, chrysene and dibenzo(a,h)anthracene were significant ($P \leq 0.05$) across all the biomass types. The levels of acenaphthylene, fluorene, anthracene, benzo(a)anthracene and benzo(a)pyrene emitted by the perennial indigenous tree species were generally higher than those from the fast growing exotic tree species, but their differences were not significant at $P \leq 0.05$ (Table 11). These

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results show that the levels of PAHs emitted vary with the biomass type (wood type or dry cow dung) used as fuel.

Previous studies have also reported that PAH emissions were mainly dependent on fuel types and burning conditions (Rogge et al., 1998; Oanh et al., 1999; McDonald et al., 2000; Zou et al., 2003). Dry cow dung burn with smoldering fires which leads to emission of higher levels of PAH content, than wood fuel which burn with flaming fires, due to lack of oxygen which favors incomplete combustion (Robinson et al., 2008). A previous study has reported that hard wood generally has lower lignin and higher cellulose percentage composition by mass than soft wood (White, 1986). This variation in lignin and cellulose composition of the different types of wood, probably affects the oxidation state of wood from perennial indigenous trees, mostly hardwoods slowing their rate of combustion which leads to emission of higher levels of PAHs ($P \leq 0.05$) than wood from the fast-growing exotic trees which are mostly soft wood. Under slow burning conditions, emission levels of individual PAHs species from wood burning was higher than those under fast burning conditions, (Zou et al., 2003). Of the four biomass types, use of shrubs and crop residues as fuel gave the cleanest ($P \leq 0.05$) burn in terms of emission of PAHs (Tables 11 and 12). The levels of naphthalene, phenanthrene and dibenzo(a,h)anthracene were below the detection limit (MDL) in houses that predominantly use shrubs and crop residues as fuel. The levels of the other PAHs were much lower ($P \leq 0.05$) than those of the other biomass types in this study (Table 11). Shrubs and crop residues undergo a fast burn with large flaming fires which leads to more complete combustions, hence emit very low levels of PAHs. This means that shrubs and crop residues present the least health risks to the inhabitants of the rural houses, in terms of indoor PAH exposure levels, when used as fuel for cooking and space warming. Their use as fuel should therefore be encouraged. The use of dry cow dung and wood from these perennial indigenous tree species as fuel should be discouraged as they are higher emitters ($P \leq 0.05$) of PAHs.

4.2.3 Variation of PAHs concentration with age of house

The soot from houses more than 10 years old gave the highest concentrations of the various PAHs quantified in this study, followed by that from houses 5 – 10 years old. Soot from houses of ages between 0-5 years gave the least PAHs concentrations (Table

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10). The mean concentrations range (in µg/g of soot) of the PAHs in soot samples from the traditional rural houses were; 0.985 – 14.775µg/g naphthalene, 5.342 – 19.843 µg/g acenaphthylene, 17.969 – 87.882 µg/g acenaphthene, 3.580 – 36.224 µg/g fluorene, 3.477 – 10.807 µg/g phenanthrene, 8.505 – 66.517 µg/g anthracene, 6.950 – 23.035 µg/g fluoranthene, 15.313 – 69.517 µg/g pyrene, 5.761 – 56.560 µg/g benzo(a) anthracene, 13.382 – 38.625 µg/g chrysene, 11.475 – 75.725 µg/g benzo(a)pyrene and 1.709 – 4.928 µg/g dibenzo(a,h)anthracene (Table 10). The trend in concentrations of benzo(b)fluoranthene, benzo(k)fluoranthene and indeno(1,2,3-cd)pyrene could not be established because they were below the minimum detection limit in more than 20% of the samples (Table 9). There are no previous reports on quantities of PAHs on accumulated soot inside houses. The findings of this study have shown that accumulated soot under the roofs of houses in an indoor is a major repository for PAHs, including the carcinogenic ones, emitted during residential burning of biomass fuel.

The levels of naphthalene, acenaphthylene, acenaphthene, fluorene, anthracene, fluoranthene, pyrene, phenanthrene, benzo(a)anthracene and benzo(a)pyrene in houses more than 10years old were significantly higher ($P \leq 0.05$) than those in houses which were 0 – 5 years old (Table 11), while the levels of naphthalene, fluorene, anthracene, benzo(a)anthracene, pyrene and benzo(a)pyrene in houses of ages between 5 – 10 years were significantly higher ($P \leq 0.05$) than those in houses of ages between 0 – 5 years (Table 11). Though the levels of chrysene and dibenzo(a,h)anthracene increased with the age of the houses, the differences between their mean concentrations in the houses of the various age groups were not significant at $P \leq 0.05$ (Table 11). In general, the mean concentrations of various PAHs in accumulated soot under the roof of the grass thatched rural houses were found to increase ($P \leq 0.05$) with the age of the houses.

Information on environmental accumulation or fate of PAHs from point sources is rare. However, both Douben (2003) and Tuvikene (1995) reported that PAHs tend to accumulate in sediments where they may be present at levels thousands of times higher than those in the overlying water, while Sharma and Mcbean (2001) reported accumulation of PAHs in snow-packs during winter, especially in the urban environment, as a result of wet deposition process. The results of this study indicate accumulation of PAHs indoors, including the lower molecular weight ones ($MW \leq 202$), usually formed in gaseous phase (Jenkins et al., 1996), in the particulate phase. This

implies that the accumulated soot underneath the roofs of the traditional rural houses act as an indoor reservoir for PAHs, by accumulating both particulate phase and gaseous phase PAHs formed as a result of continuous burning of wood and dung by the rural population as fuel, for cooking and space warming, over the years.

Biomass fuel, wood and dung, burning indoors is done daily in open fire places releasing the combustion products, including PAHs, inside the poorly ventilated houses. As the smoke, which includes PAHs, slowly finds its way through the grass thatched roofs, the PAHs particles physically adsorb on to the accumulated soot (Venkarataman *et al.*, 2002). Fate process for PAHs such as photolysis and biodegradation either do not occur at all or are severely impeded by the dark and dry indoor conditions of these houses. This implies that the rate of PAHs deposition in the accumulated soot under the roofs of the traditional rural houses is faster than their rate of removal through any of the fate process, leading to accumulation.

The vapour pressures of PAHs generally decrease with increase in molecular weight (IARC, 1983). The vapour pressure of the lower molecular weight PAHs range from naphthalene, 10.4Pa, fluoranthene, 1.2×10^{-3} Pa to chrysene, 8.4×10^{-5} Pa (Table 2). The health problem results from the probable vaporization of these adsorbed PAHs, which include lower molecular weight PAHs (MW<202.3) usually formed in vapour phase (Jenkins *et al.*, 1996), due to the high indoor temperatures (Yamasaki *et al.*, 1982), as a result of wood and dung burning open fires places. This, in addition to the daily emission of PAHs directly from the burning biomass during cooking, may lead to higher continuous human exposure to higher vapour phase PAHs concentration, including the highly carcinogenic ones, indoors than outdoors or in those houses using cooking stoves with ventilation by flues, hoods or chimneys and improved combustion efficiency, even at times when no cooking is taking place. The health risk problem is compounded by the observation that most of the cooking houses also serve as the living houses yet air movement indoors is very low due to poor ventilation.

According to the data collected by the Kakamega Hospice (Kakamega Hospice Registry, 2010), cancer cases are rampant within the area with cancer of the oesophagus registering the highest number of newly diagnosed and reported cases each successive month (Table 7). New cases of cancer of the cervix, prostate cancer and breast cancer cases are also diagnosed at the Kakamega PGH and reported every month. Death cases

amongst these patients from cancer have also been documented (Table 7). There is very high probability of continuous human exposure to much higher levels of both vapour phase PAHs, and particulate phase PAHs, through inhalation of falling fine soot particles, in these rural houses, even during times when no cooking or space warming is taking place, than outdoors. While this study can not conclusively establish a relationship between these relatively high indoor levels of carcinogenic PAHs, emitted during burning of biomass as fuel, and the many cases of cancer patients diagnosed in the region, the possibility of a link between the two can not be simply ignored.

4.2.4 Variation in the pattern of accumulation of PAHs in soot with predominant biomass type used

The results of this study show that continuous burning of each of the biomass types considered in the study, as fuel in open fire places inside the traditional rural grass thatched houses, leads to accumulation of PAHs in the accumulated soot under the grass-thatched roofs (Section 4.2.3). Significant interaction in accumulation patterns of the various PAHs in the accumulated soot, as a result of burning various biomass types, wood from perennial indigenous trees and fast-growing exotic trees, shrubs and crop residues and dry cow dung, was set at $P \leq 0.05$. The interaction in the patterns of accumulation of naphthalene, phenanthrene, fluoranthene, pyrene, and benzo(a)anthracene, from predominantly burning the various biomass types, was significant ($P \leq 0.05$) (Table 11). This means that these PAHs accumulate in the accumulated soot faster when some biomass types are predominantly used than others. This indicates that the pattern of accumulation of each of these PAHs vary with variation in biomass type predominantly used for cooking and space warming.

Previous studies (Hays *et al.*, 2003; Venkataraman *et al.*, 2002), on the mechanisms of individual PAHs, including those emitted from biomass burning, association with particulate matter gave an inconsistent trend, which pointed to multiple mechanisms of PAH-particle associations, including adsorption and absorption. Hays *et al.*, (2003) indicated that the surface areas of particles seemed highly sensitive to the PAH mass, which affects both adsorption and absorption processes. During PAHs distribution, higher and lower molecular weight PAHs preferentially segregate to fine particles and coarse particles respectively (Hays *et al.*, 2003; Allen *et al.*, 1996). This

variation in PAH-particle association mechanisms with particle size and PAH mass probably causes variation in the accumulation patterns of some of the PAHs emitted from burning various biomass types in accumulated soot under the roofs of the traditional rural houses.

The interaction in the patterns of accumulation of acenaphthylene, acenaphthene, fluorene, anthracene, chrysene, benzo(a)pyrene and dibenzo(a,h)anthracene were not significant at $P \leq 0.05$ (Table 11). These results indicate that the accumulation patterns of these PAHs from biomass burning in open fire places in the traditional rural houses is homogeneous across all the biomass types considered in the study.

Thermal breakdown and alteration of the cellulose polymer and steam-stripping distillation are among the particle formation mechanisms of biomass combustion (Simoneit *et al.,* 1999). The fuel type and combustion conditions therefore influence the particulate matter mass distributions (Purvis *et al.,* 2000). Higher temperatures likely limit particle size partitioning of organics, including soot particles, lessen agglomeration, and shift the mass distributions to smaller diameters. High ventilation can also help to encourage and sustain an intense flaming condition, which in turn produces a large concentration of smaller diameter particles (Hays *et al.,* 2003). The biomass burning inside the houses is done in poorly ventilated conditions. This similarity in biomass burning conditions probably has an effect on the distribution in the size of the particulate matter, emitted during biomass burning, which has a homogenous effect, across all the biomass types, on the pattern of accumulation of some of the PAHs in the accumulated soot under the roofs.

CHAPTER 5

5.0 SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

5.1 Summary

- This was the first study to characterise and quantify PAHs in accumulated soot. Fifteen out of the target sixteen US EPA priority carcinogenic PAHs; naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo(a)anthracene, chrysene, benzo(a)pyrene, dibenzo(a,h)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene and indeno(1,2,3-c,d)anthracene, were identified from soot samples .

- The type of fuel used; wood type, dung or shrubs and crop residues, has a significant effect ($P \leq 0.05$) on emission levels of PAHs. Burning of dry cow dung as fuel emitted the highest levels ($P \leq 0.05$) of PAHs, followed by wood from the perennial indigenous tree species. Wood from the exotic fast-growing tree species emit lower levels ($P \leq 0.05$) of PAHs than wood from the perennial indigenous tree species while burning of shrubs and crop residues as fuel was found to emit the least amount ($P \leq 0.05$) of PAHs.

- The soot from houses more than 10 years old gave the highest concentrations of various PAHs quantified in this study, followed by that from houses $5 - 10$ years old. Soot from houses of ages between 0-5 years gave the least PAHs concentrations.

- The accumulation pattern of acenaphthylene, acenaphthene, fluorene, anthracene, chrysene, benzo(a)pyrene and dibenzo(a,h)anthracene in the accumulated soot inside the traditional rural houses is homogeneous across all the biomass types considered in the study, while patterns of accumulation of naphthalene, phenanthrene, fluoranthene, pyrene, and benzo(a)anthracene in the accumulated soot inside the traditional rural houses vary with variation in biomass type predominantly used as fuel, as evidenced by the significant interaction effects.

5.2 Conclusions

- Indoor burning of biomass in open fires places in rural houses during cooking and space warming emit PAHs, including the highly carcinogenic ones, which get adsorbed onto soot particles that form under the grass-thatched roofs. This pause possible health risks to the inhabitants of these houses.

- The levels of PAHs emitted from biomass burning vary with the biomass type (wood type or dry cow dung) used as fuel. The use of dry cow dung and wood from these perennial indigenous tree species as fuel should be discouraged because they are higher emitters ($P \leq 0.05$) of PAHs.

- The concentrations of various PAHs in accumulated soot under the roof of the traditional rural houses increased ($P \leq 0.05$) with the age of the houses. This accumulated soot under the roofs of the traditional rural houses is a major repository for PAHs, including the carcinogenic ones, emitted during residential burning of biomass fuel.

- Naphthalene, phenanthrene, fluoranthene, pyrene, and benzo(a)anthracene accumulate in the accumulated soot faster when some biomass types are predominantly used than others, in the order; dung $\geq$ perennial indigenous trees $\geq$ exotic trees $\geq$ shrubs and crop residues. Accumulation patterns of acenaphthylene, fluorene, acenaphthene, anthracene, benzo(a)pyrene, dibenzo(a,h)anthracene and chrysene, did depend on the biomass type used.

5.3 Recommendations

- ➤ Programmes should be put in places to encourage the population to adopt cooking stoves with ventilation by flues, hoods or chimneys and improved combustion efficiency instead of open fires places, to reduce emission and release of PAHs indoors.

- ➤ People should be encouraged to use the shrubs and fast growing exotic tree species for cooking and space heating since they emits lower levels of PAHs during combustion than the perennial indigenous trees and dry cow dung. Use of cow dung and wood from the perennial indigenous trees indoors should be discouraged.

➤ The population should also be encouraged to adopt house design change to improve ventilation; increase the number of window openings in the kitchen, provide gaps between the roof and walls of the house, and the importance of having a kitchen separate from the living houses.

➤ People should be encouraged to increase the ventilations in the houses to encourage and sustain intense flaming conditions during biomass burning, which in turn produces a large concentration of fine particles. This would reduce the rate of PAHs accumulation indoors, since lower molecular weight PAHs, which dominate PAHs emitted from biomass burning, preferentially associate with coarse particles.

5.4 Suggestions for future studies

• A study should be done to evaluate the levels of indoor vapour phase levels of various PAHs in these houses during cooking, when no cooking is taking place, during the day when doors and windows are open, and at night when doors and windows are closed.

• A study should be done to establish if there is any evidence of a link between the relatively high indoor levels of PAHs, emitted during burning of biomass fuel, and the rampant cases of cancer patients in the region.

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