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Review

Essentials of ecotoxicology in the tanning industry

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Pollution within the tanning Industry has been a great concern with efforts recently directed towards mitigating the emerging challenges. The most inspiring is the development of novel ecotoxicological diagnostic tools which are considered rapid, cost effective and reliable. In the tanning industry, particular attention is directed towards use of chromium, salts, chlorinated phenols among others which are recognised as potential pollutants especially within the developing world where environmental restriction is still low. Therefore the review attempts to underscore the need to comprehend about these pollutants and the appropriate diagnostic approaches required. The application of biosensors and dehydrogenase is discussed to evaluate the toxicity levels and provide inherently the bioavailability status of the contaminants. Indeed while the diagnostic tools are crucial and opportune, other measures to ensure cleaner production within the tanning industry needs to be addressed to disalienate the notion that the tanning industry is still a major source of pollution.

Key words: Ecotoxicology, effluents, leather, pollutants.

INTRODUCTION

The world production of hides and skins is approximated at 1303 million pieces, while in Africa it is about 183 million pieces. Share wise therefore Africa has 14% of the world production. If compared to the world scenario it depicts 0.47% share to the world (Mwinyihija, 2011). The global leather industry is currently valued at more than $75 billion (Mwinyihija, 2011). The worldwide production of the leather is expected to have grown to 2.4 billion square metres by the end of 2000 (Komma, 1996). The leather and leather products industries have witnessed considerable shifts in the location of tanning and leather manufacturing to developing countries where production costs are lower and environmental regulations less stringent (Muchie, 2000). Indeed, while production of leather and fur products has decreased between 1975 and 1992 in developed countries, it has increased in the developing world from 4.4% (1975 – 1985) to 5.3% (1985 –1992) (UNIDO, 1992). Demand for leather and leather goods was also expected to rise, despite slowdowns as a result of the 1998 Asian financial crises (Muchie, 2000), while demand grows at 0.6% for the developed countries, the projected rise for developing countries is 1.1% (CCP, 1998), Mwinyihija, 2011.

When this performance is adjudged against the environmental impact the tanning industry also referred to as the leather sector exerts to the ecosystems, one wonders about the logical use of techniques that challenges negatively to the ecosystems. However emergence of sophisticated technologies in industrial, waste and water treatment techniques and new lifestyles means that the way the environmental media (terrestrial, aquatic and atmospheric systems) are imparted will differ at any point and place in time. This therefore poses a challenge in managing newer threats associated with stressors that are exposed to such environmental media. Of a major concern among the environmental media is the aquatic system that forms the basis of all living systems (Baumgartner, 1996; Sweeting, 1994, Mwinyihija et al., 2005, 2006a) and the terrestrial systems that supports such existence. For example the demand for clean water as a resource has risen with the increasing urbanisation (domestic supplies and recreation), industrialisation and agricultural intensification of the last century. Ironically, industrialisation and agricultural activities are the major contributors to pollution in the atmosphere, terrestrial and aquatic systems (SEPA, 1999; Mwinyihija et al., 2005). Soil contamination can be localised (e.g. related to industrial sites) or diffuse (the result of deposition of a pollutant over a wide area). More over the environmental threats could be related to physical or sensory attributes such as waste mound hills or resultant odour.

LEATHER SECTOR

To comprehend on the essentials of ecotoxicology it is important to review the basis of the leather sector and explore the production chain. Thus the transformation of animal hides and skins into leather primarily involves five main phases; flaying, preservation (whenever applicable) to arrest putrefaction; removal of hair and flesh, and tanning depending on the final leather products targeted through the application of chemical agents. The aim eventually is to render the raw material to be non-putrescible and enhance its durability (Mwinyihija, 2010).

Pollution from the leather processing industries has a negative long-term impact on both the ecosystem health and functionality; and economic growth potential of a country irrespective of the immediate profit accruals intended. Development of the industry in Africa for example does not match the technical know-how and capacity in protecting and predicting the environmental impacts related to the industry. Cleaning up of such environment will require expenditure of funds, which could have promoted positive and sustainable development. Tanning has thus been deemed as one of the largest polluters in the world.

Tanning pollutants and its effect to the ecosystem

Tanning Industry involves the following processes in brief, linked closely with pollution:

Soaking

The objective in carrying out soaking at this stage is to wash the rawstock from debri, dirt and other related physically bound materials (e.g. mostly insecticides, salts ($NaCl_2$) and other preservatives). Primarily and very important (other than washing) the purpose also at this stage is to rehydrate the hides and skins to replace lost moisture during curing to allow for the next level's process involved with application of various chemical to allow for permeation of such intended chemicals, during subsequent processes.

Liming

After the hide and skins is rehydrated, the material is ready to move to the next stage where the use of alkaline medium is undertaken e.g. lime. The aim at this stage is to remove the hair, flesh and splitting up of the structural protein e.g. fibre bundles by chemical and physical means (Ramasami et al., 1991). In this process Na_2S is added to facilitate in dehairing (Flaherty, 1959). It is estimated that for processing 1ton of raw skins weight of skins before soaking the input in a typical input audit processing(kg) of lime is 100 with an output of 12.3, while Na_2S has an input of 35 with an output of 18.3 (Thanikaivelan, 2000).

Deliming, bating and pickling

For these processes weak organic acids, digestive enzymes and inorganic acids respectively are used to remove the lime, digest and remove the non-structural proteins and eventually bring the pH to a level that will enhance the tanning process.

Chrome tanning

Nearly 90% of all leather produced is tanned using Cr salts (Stein, 1994) especially in the developed world. 8% of the basic chromium sulphate salt is used for conventional tanning. It binds with the collagenous protein to convert to leather. This conversion renders the material non putrescible.

To understand these fundamental processes which are, but a few, of the total that are found in processing of leather, form the vital point of intervention in pollution control. Main pollutants found during leather processing include, $NaCl_2$ and pesticides, strong alkalines and sulphides, inorganic residual compounds, dissolved matter and chromium salts. Chlorinated phenols are important compounds to be investigated due to the various mixtures used in the tanning industry and their ecotoxicity potential.

In another approach of tanning, vegetable tannin are used mostly to retan leather to impart certain specific properties desired or could be used alone in producing leather especially at the rural tanning level by the use of plant material (this could be tree barks and pods which are commonly used in Africa). Mostly according to the tanning sciences, the tannin materials are derived from plants and consist of condensed or hydrolysable tannins (Zywicki et al., 2002). In the East African region, wattle and certain species of acacia (e.g. in arid and semi arid areas) is extensively used as a tannin material. Previous efforts to study the polyphenolic structures of condensed tannins have been hampered by the fact that the structure rapidly transforms during the tanning process to yet unknown products (Zywicki et al., 2002).

Other related ecotoxicological approaches

Biosensors: There is growing need for quick, cheap and reliable bioassays for toxicity testing of compounds. The demand for such a necessity emanates from the continued escalation of pollution load related to elevated anthropogenic activities (i.e. agricultural, urbanisation and industrial activities). Although there is zeal to use cleaner technologies in various processing techniques in the manufacturing sector, contamination of various eco-systems in the world continue to be conspicuous. There is need at this juncture to understand two terms used frequently in environmental sciences that is *pollution* and *contamination* more clearly. Pollution is a term that defines the state of the anthropogenic introduction of substances into the environment causing hazard to human, living resource, damage to structure, and eco-systems. Contamination while used synonymously with pollution demonstrates the concentration leverage of a substance when it exceeds its natural occurrence but not with the set standard threshold. Thus pollutants are chemicals that cause environmental harm (Harrion, 2001) in contrast to contaminant which does not necessarily impact an evident harm.

Therefore the study of these contaminants and pollution in the environment require novel ecotoxi-cological techniques in understanding and predicting the impact of such chemicals on terrestrial and aquatic systems. Manly, 2000 defines ecotoxicology as the study of toxic effects of substances on the biotic and abiotic components of the biosphere, especially on populations and communities within defined ecosystems. However it is imperative to understand that ecotoxicology is a discipline within a wider field of environmental toxicology. Thus bioassays that integrate toxicity tests developed to examine the effects or impacts of chemicals in a broad range of ecosystems using various species are presently the modern preferred ecotoxicological techniques. Indeed bioassay may be used to determine both short term (acute) and long term (chronic) impact of pollutants, as well as being used to study the mode of action and routes of transport through the ecosystems. Bioassays are therefore described as tests determining or estimating the effects of biologically active substances under standardised and reproducible conditions.

To that effect, emerging novel techniques for ecoto-xicological analysis has included biosensors. Mwinyihija (2010) found out that efficiency, accuracy, rapidity, convenience and on-line monitoring are some of the advantages conferred by the use of biosensors over other forms of biomonitoring. Vo-Dinh and Cullum, 2000 defined biosensors as a combination of a bioreceptor, biological component, and a transducer as the detector. The interaction of the analyte with the bioreceptor has the functional characteristic of producing an effect which is measured with a transducer. The transducer then con-verts the information into measurable effect such as an electrical or optical signal also referred to as bio-luminescence. Moreover biosensors have been reported to provide reliable technique of measuring biological effects (e.g. acute and chronic physiological toxicity, genotoxicity, immunotoxicity and endocrine toxicity) and the concentration of specific analytes which are difficult to detect and are important contaminants of water, waste, soil or air (e.g. surfactants, chlorinated hydrocarbons, sulphophenyls carboxylates, sulphonated dyes, fluorescent whitening agents, naphthalensulphonates, carboxylic acids, dioxins, pesticides and metabolites). The protocols for the biosensor (e.g. *E. coli* HB101 pUCD 607) in flow chart as indicated in Figure 1.

During the research period *lux*-marked bacteria biosensors (Mwinyihija, 2010) (Figure 2) offered a powerful way forward for rapid screening of the effect of environmental variables on the fate and toxicity of the pollutants.

Dehydrogenase: Mwinyihija (2010) provides an in depth use of Dehydrogenase test as a tool in an Ecotoxicological diagnosis especially for the river sediments impacted by the tanning industry for the first time. The technique is a simple and inexpensive test used to determine the degree of toxicity by measuring the microbial activity through the production of the enzyme dehydrogenase. Dehydrogenase activity (DHA) tends to be a common and suitable test made to quantify the impact of heavy metals on soil micro-organism. The mea-surement of enzyme concentrations may be used as an indirect measure of the soil microbial activity. This may serve as a supplement to biomass measurement. This is because DHA is assumed to be proportional to microbial respiration (Stevenson, 1959; Thalman, 1968; Skajins, 1973; Frankenberger and Dick, 1983). The sensitivity of the particular enzyme to the pollutant depends on the success of such a related study. Dehydrogenase is an enzyme which is unspecific in its activity unlike urease. Zn and Cu additions were found to decrease the activity of acid phosphatase and urease activity (Tyler, 1976) and amylase (Ebregt and Boldewijn, 1977). Dehydrogenase functions in living cells while phosphatase is an extracellular enzyme. Adverse effects on the metabolic functioning of the cell would result in reduction of Dehydrogenase activity (DHA). Compara-tively, reductions in phosphate activity would not be as noticeable as those of Dehydrogenase when the microbial metabolic function reduces.

The mode of action for the dehydrogenase encompasses a theorem where an endocellular enzyme Dehydrogenase facilitates in the transfer of hydrogen and electrons from organic compounds to appropriate electron acceptors during the initial oxidation of the substrate (Skujins, 1978).Normally when electrons pass along a chain of intermediate carriers, Oxygen act as the final electron acceptor. In the dehydrogenase test tetrazolium salts (e.g. triphenyltetrazolium chloride (TTC) and 2-(4-iodophenyl)-3-(4-nitrophenyl)-5-phenyl tetrazolium chloride) act as the terminal electron acceptors in the anaerobic environments (Trevors et al., 1981). Banefield et al. (1977) suggested that in O_2 depleted conditions aerobic as well as anaerobic dehydrogenase use TTC as an electron acceptor. In effect TTC is reduced to TPF (triphenyltetrazolium

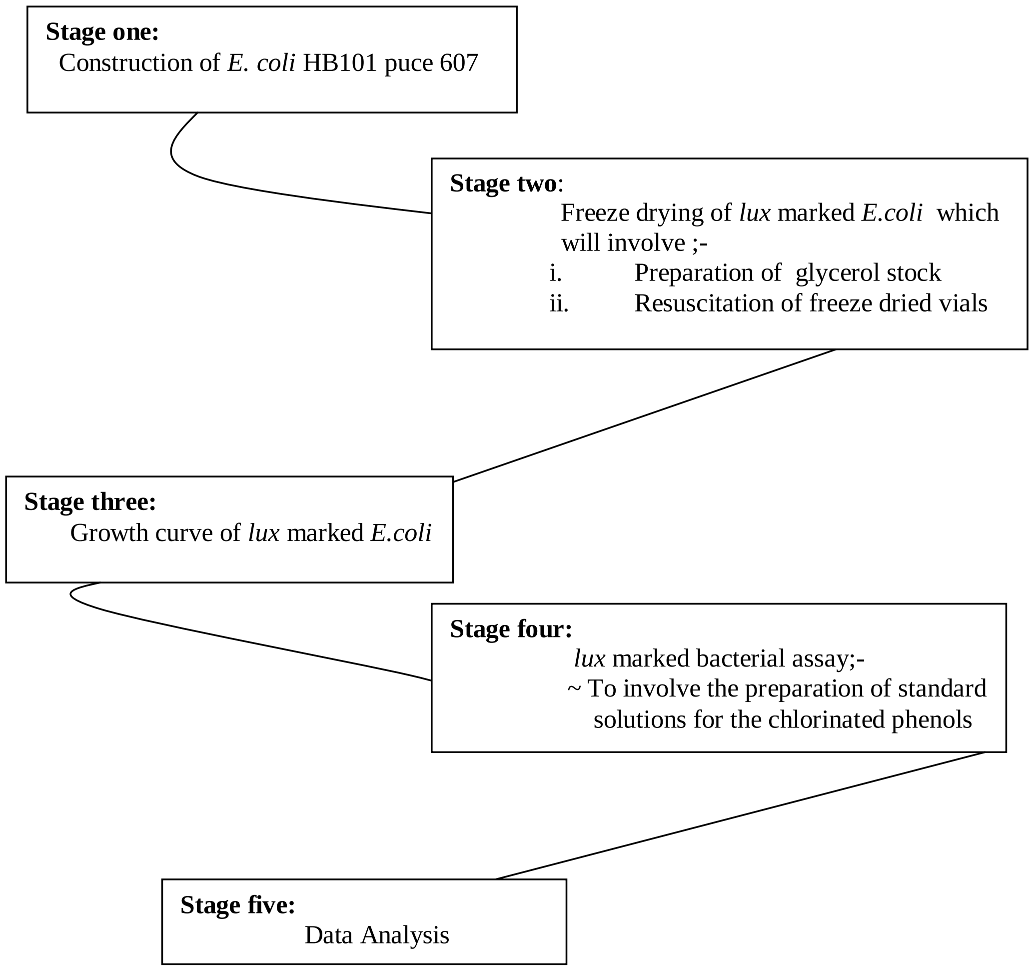

Figure 1. Protocols To assess lux-marked *E. coli*'s response in the form of toxic response and examine the effect of exposure time on toxicity of pollutants.

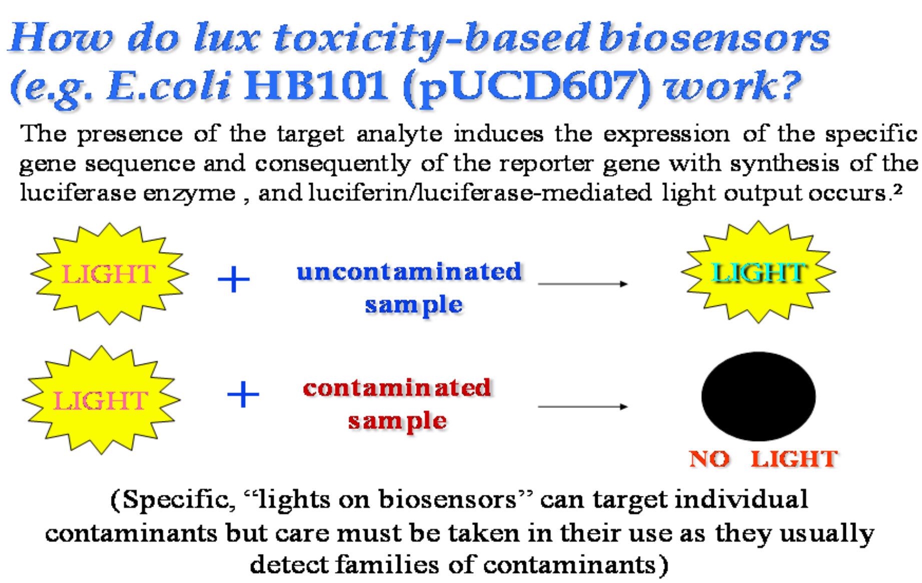

Figure 2. Illustration of how *lux* toxicity based biosensors work.

formazon) a red insoluble compound. Ethanol is used to dissolve the precipitate and calorimetrically measured by a spectrometer.

i. Application of the DHA as an ecotoxicological tool

The Dehydrogenase test has been used to determine the effects of heavy metals present in sewage sludge (during the study a reduction of the DHA in metal contaminated soils was observed suggesting a decline in microbial activity in polluted soils) on the soil microbial activity (Cenci and Morozzi, 1979; Ruhling and Tyler, 1973; Doelman and Haanstra, 1979; Schinner et al., 1980; Brookes et al., 1984). Similarly DHA was used to determine the microbial biomass of the soil (Malkommes, 1988; Wilke and Keuffel, 1988). A correlation between the DHA and the biomass of the soil microflora was established (Beck, 1984) and also other processes involving nitrification (Skujins, 1973). Recently it was used as a tool to ascertain river health impacted by the tanning Industry (Mwinyihija et al., 2006a; Mwinyihija, 2010).

However there are limitations through use of Dehydrogenase Tests. For example Dehydrogenase in the presence of Cu (unlike other enzymes such as urease, invertase and cellulase which) causes a reduction in TPF (triphenyltetrazolium formazon) absorbance (Chander and Brookes, 1991). The presence of alternate electron acceptors in the soil may cause a problem in comparing the DHA of different soil types due to a large proportion of O_2 uptake which remains unaccounted for (Burns,1978) (Somerville et al., 1987). DHA inhibition may also be caused by humic acid (Pflug and Ziechman, 1981) or soils with high inorganic nitrogen (Trevors, 1984) and long incubation periods.

In retrospect ways of addressing these shortcomings have been developed. For example competition of DHA with other electron acceptors in the soil may be addressed by using INT (2-p-iodophenyl-3-p-nitrophenyl-5-phenyl-tetrazolium chloride) instead of TTC (Benefield et al., 1977; Mwinyihija, 2006b). INT has a rapid response, high sensitivity and compete well with O_2 for liberated electrons. INT as an electron acceptor is pH sensitive with optimum result at pH 4.8 (Benefield et al., 1977). DHA as a test may further be enhanced by shortening the incubation period from 24 to 6 hours and by adding yeast extract (where a 53% increase in DHA was observed) (Rossel and Tarradellas,1991), good correlation between short term respiration and DHA (Beck ,1984) and lack of correlation occurring between DHA and microbial numbers especially in nutrient starved soils ,may be due to the presence of low metabolism, dormant or dwarf forms of organisms(Marshall, 1980, Roszack and Colwell, 1987). DHA as a test estimates the total potential activity of the microflora rather than its total effective activity.

Fundamentals of applying ecotoxicological tools for diagnosis

There is increasing movement towards the use of biological techniques for the monitoring of environmental pollution by industry and regulators a like (McHenry, 1995; Mwinyihija, 2010; 2009; 2006a). This will further strengthen capacity in quality control where detrimental and beneficial aspects of the environment influenced by the tanning industry will be monitored more efficiently and effectively.

The fundamentals of the study by Mwinyihija (2010) related to the tanning industry and mostly centred on; characterising the chemistry of effluents of Kenyan tanning industry, developed toxicity testing procedures for risk assessment and also undertook field based assessment of environmental impact of chromium. The manipulation linked to toxicity for the purpose of remediation will include additive techniques such as; purging, ion exchange resin extraction, pH adjustment and filtration through activated charcoal. Subsequently the use of whole cell microbial biosensors as a way forward for monitoring environmental contamination by heavy metals (e.g. Cr for this study) (Knight et al., 1999) and organic contaminants (e.g. chlorinated phenolics) as well as toxicity in soils and water contaminated by industrial effluents (Brown et al., 1996) including river sediments and tannery dust (Mwinyihija et al., 2005, 2006b; Mwinyihija, 2010)

Selected potential pollutants of the tanning industry

The following areas form important part of this review to identify impact areas of the tanning industry effluent on soils and water;

$NaCl_2$: Salt as a curing agent in the primary level of preservation is an inorganic chemical that strongly has been identified as a pollutant. In a study in Egypt, $NaCl_2$ concentration varied between 40, 000 to 50, 000 mg/l in the effluent discharge in the tannery under study (Hafez et al., 2002). Salinity or ionic strength can cause a small decrease in the solubility of non-polar organic compounds (e.g. Naphthalene, Benzene, toluene etc) through a process known as salting-out effect (Pepper et al., 1996). This is a very critical point of interest in the tanning industry and its one of the most important pollutant that need to be addressed urgently.

Organic matter: Most of these will include biodegradable organic matter e.g. Proteins and Carbohydrates. Their main problem is the depression of the dissolved oxygen content of stream waters caused by their microbial decomposition. Their impacts are primarily the loss of

Table 1. Summary of various Cr oxidation numbers, type and environmental behaviour.

Valency	Environment behaviour	Remarks
Cr	Unstable	
Cr^{1+}	Unstable	
Cr^{2+}	Readily oxidised to Cr_3 but stable only in the absence of any oxidant	Active under anaerobic condition
Cr^{3+}	Most stable	Considerable energy required to convert to lower or higher states
Cr^{4+}	Forms Unstable intermediate reactions to trivalent and oxidation states	Exhibits this phase during oxidation and reduction
Cr^{5+}	Unstable intermediate	Observed during oxidation and reduction
Cr^{6+}	In acidic conditions demonstrates very high positive redox potential and unstable in the presence of electron donors	Strongly oxidizing

dissolved oxygen, which is detrimental to aquatic organisms. Secondly, their effect is on dissolved oxygen that is consumed by aerobic microbial oxidation of the waste; anaerobic decomposition becomes prominent and releasing noxious gases (Pepper et al., 1996).

Chromium salts as important tanning industry ingredients: Chromium basic sulphate is the most widely used tanning substance today. The exhausted bath coming from the chromium tannage contains about 30% of the initial salt and is normally sent for cleaning up (Cassano et al., 2000). Here chromium salts are entrained in the sludge creating serious problems for their disposal (Gauglhofer, 1986). Chromium can exist in several chemical forms displaying oxidation numbers from 0 to VI. Trivalent and hexavalent forms are most stable in the environment exhibiting different environmental behaviour as depicted in Table 1.

In nature, Cr originates from weathering or rock, wet precipitation and dry fallout from the atmosphere and run off from the terrestrial systems (Kotas and Stasicka, 2000). In rivers and lakes the Cr concentration is usually limited to 0.5 – 100 nM (Handa, 1988; Kaczynski and Kieber, 1992), in seawaters it varies from 0.1- 16 nM. Tanning industry can contribute highly to the increase of Cr concentration if located near the water systems. In nature Cr exists in its two stable oxidation states, Cr^{3+} and Cr^{6+}. The presence and ratio between these two forms depend on various processes; Chemical and photochemical, redox transformation, precipitation/ dissolution. Campanella et al. (1996) described the Cr^{3+} in oxygenated aqueous solution as predicted by thermodynamic calculations on the stable species at pH $\leq$ 6 whereas pH $\geq$ 7 the CrO_4^{2-} ion as predominate under anoxic and suboxic condition, trivalent Cr should be the only form (Kotas and Stasicka, 2000).

The nature and behaviour of various Cr forms found in wastewater can be quite different from those present in natural water because of altered physico-chemical condition of the effluents originating from various industrial sources (Kotas and Stasicka, 2000). The presence and concentration of Cr forms in effluents depends on Cr compounds applied during processing, pH, and organic and / or inorganic waste coming from the material processing ((Kotas and Stasicka, 2000).

Cr^6 will dominate in wastewater from metallurgical industry, metal finishing industry (Cr hard plating), refractory industry and production or application of pigments (chromate colour pigments and corrosion inhibition pigments). Cr_3 dominates from tannery, textile (printing, dyeing), decorative plating industry waste. In reference to the tanning industry Cr_3 in the effluents is the most expected form but with redox reactions occurring in the sludge, an increase in hexavalent form can occur.

Under slightly acidic or neutral pH, condition in this type of wastewater the poorly soluble $Cr(OH_3)$.aq should be the preferred form, but a high content of organic matter originated from hide/skin material processing is effective in forming soluble Cr^3 complexes (Stein et al., 1994; Walsh and Halloran, 1996).

In soils an increase in local Cr concentration originates from fallout and wash out of atmospheric Cr containing particles as well as from the chrome bearing sludge and refuse from industrial activity (Kotas and Stasicka, 2000). Cr^3 adsorption into humic acids renders it insoluble, immobile and unreactive. This process is the most effective within the pH range of 2.7 – 4.5 (Walsh, 1996). Other macromolecular ligands behave similarly. In contrast mobile ligands such as citric acid, diethylene triamine pentaacetic acid (DTPA) and fulvic acid form soluble Cr^{3+} complexes, which mediate its relocation and oxidation to Cr^{6+} in soils (James and Bartlett, 1983; James, 1996). Dechromification is thought as being of vital importance because without it, theoretically all atmospheric oxygen could be a threat to life on earth (James and Bartlett, 1983).

Whilst in the atmosphere Cr present in the atmosphere originates from anthropogenic sources, which account for 60-70%, as well as from natural sources, which account for the remaining 30-40%. Industrial activities still remain the major source of pollution to the atmospheric systems. Other could be natural sources like volcano eruptions and erosions of soil and rocks ((Kotas and Stasicka, 2000). Sea salt particles and forest wild fires do not seem to be important sources of Cr. Average atmospheric concentrations of this metal are, 1 ng/m^3 in rural to 10 ng/m^3 in polluted urban areas. The amount of Cr at any particular time depends on the intensity of industrial processes, proximity to the sources, the amount of Cr released and meteorological factors (Kotas and Stasicka, 2000).

Cr forms mostly ionic compounds, their vapour could be neglected and one may assume that gaseous Cr species do not exist at ambient atmospheric temperatures and Cr is present in the atmosphere in form of particles and droplet aerosols. On the other hand Cr from sources releasing the element in lager particles (particle diameter varies: 0.2-50 µm (Kotas and Stasicka, 2000) is deposited locally and can migrate through individual particular environmental media. The size of particles is of importance for consideration of Cr toxicity: Friess et al. (1989) found that only the particles of diameters from 0.2 to 10 µm are respirable, and that their retention in the lung can pose carcinogenic risk.

Chlorinated phenols related to the tanning industry

The curing and storage phase of the hides and skins utilises various types of insecticides and antimould chemical compounds. All these including the bacteriostats used in the tanning industry are eventually spewed out into the tannery effluents. Most of these compounds belong to the chlorophenols, which enter the environment through several pathways (Steirt and Crawford, 1985). It was reported by Escher et al. (1996) that phenolic compounds exert toxic effect on microorganisms disrupting energy transduction either by uncoupling oxidative phosphorylation or inhibiting electron transfer. Substituted phenols act by destroying proton gradient by transporting protons back across the membranes and/or inhibiting electron flow by binding to specific components of the transport chain Escher et al. (1996).

Chlorophenols are routinely determined by chemical analysis. The disadvantage of this method is the inability to determine the concentration of the chlorophenols present, in effect providing no information on availability of the compounds to the biomass. Further, chemical analysis does not provide information in the toxicity of the compounds and factors that affect toxicity. In retrospect biological assessment provide sensors that can identify the toxicity of chlorophenols in a variety of environmental matrices. It assesses the impact of any environmental factor on the toxicity of these compounds much quickly and easily.

During previous study by Mwinyihija (2010) *lux* marked *E. coli* was used as a biosensor. It was responsive to a wide variety of chlorophenols depending on the number and position of chlorine substitution. The advantages associated with the use of this biosensor as an appropriate Ecotoxicological diagnostic tool, was that they are rapid, reliable tools for the toxicity testing and easy in assessing environmental factors associated with the pollutants (Mwinyihija et al., 2006b; Mwinyihija, 2009)

Perspective of ecotoxicology on public health and ecology

Adopting of this perspective of study assisted in investigating the occupational hazards within the tanning industry e.g. understanding the impact levels of contaminated tannery dust. Indeed an in-depth qualitative assessment of the risk of human exposure to dust was made throughout a commercial Kenyan tannery using this technique and reported upon (Mwinyihija et al., 2006c). The success of applying this novel technique also for the first time, was obtaining information which demonstrated the high-risk points in the processing line and identifying the dust sampling regimes developed. A support study by Mwinyihija et al. (2005) using an optical set-up using microscopy and digital imaging techniques was used to determine dust particle numbers and size distributions. The results showed that chemical handling was the most hazardous (12 mg m^{-3}). A Monte Carlo method was used to estimate the concentration of the dust in the air throughout the tannery during a working day. Thus toxicity studies related to the ecology could also engage other methodologies apart from bioassays. This method also proved beyond reasonable doubts that Ecotoxicological diagnosis could also provide mitigation of occupational risk within the tannery workers environment.

The effect on pollution of terrestrial and aquatic ecosystem carries a positive correlation to human health as a component of the environmental matrix.

Soil and effluent samples characterisation as an important ecotoxicological approach

Characterisation of soil and effluent samples from the tanning sector is fundamental in understanding the primary sources of toxicity in both terrestrial and aquatic environment as caused by the sector. This process will develop the appropriate diagnostic and remediation strategies to be deployed subsequently. Various systems of characterization have been used recently including x-ray diffraction, photo acoustic spectroscopy and electron paramagnetic resonance (Wararatananurak, 2000).

It has been reported by investigations undertaken by Ritchie et al. (1972), that the humus–clay complexes which are formed with smectite clays form bonding not easily destroyed by even microorganism. Therefore with high amounts of organic matter the shrink/swell propensity is reduced (Ritchie et al., 1972). Many organism such as fungi's, diatoms and earthworms are found in such soils irrespective of discomforts due to the shrinking (due to dry conditions) and swelling(due to wet conditions). According to Killham (2001), he denotes that soil being heterogeneous in nature it is imperative during its study to investigate and eventually conceptualise all the soil parameters to aquire an integrated understanding of the role of microsites in soil ecology. These parameters include minerals, organic matter, living biomass, structure, water, atmosphere, redox, pH, temperature and light. Thus in our current research proposal forming the basis of the proposed thesis, the study of microsites impacted by the effluent form the curing sites will form the main basis of investigation, enveloping the understanding of interaction between different components of the soil biota (Killham, 2001). In view of that, during the field studies, modern molecular techniques involving the marking and *in situ* detection of components of the soil biota coupled with the traditional soil micromophological methods would be considered to better characterise the soil at the microsite level.

To provide the best Ecotoxicological diagnosis the investigator intends to obtained effluents that are raw; this will be for the purpose of identifying the predominant toxicants and establishing their levels of toxicity when spewed out to the environment. The traditional method of soil and effluent analysis were undertaken following *in situ* sample collection. This included (a). sample preparation, moisture content determination, particle size analysis, and measurement of soil pH, organic carbon, cation exchange capacity, and total nitrogen and (b) determination of major constituents dissolved in riverwater within the effluent discharge areas and outwith the discharge areas.

Conclusion

Ecotoxicology as an emergent science has all the advantages of finding the causes and effects to pollutants and their associated impact to various ecosystems. The current study leading to the proposed thesis will investigate the influence of effluents emanating from the curing premises at a selected site in Kenya is opportune and appropriate so as to provide a cost effective and easy to apply technique in the proposed area. Indeed previous study by the author has had a fundamental influence and scientific backing to the proposed area of investigation. Thus the focus on areas such as the study of complexity of effluents form the curing of hides and skins will benefit the zeal to expand the knowledge horizon in ecotoxicology as an emerging science. This will not only provide the essentials in ecotoxicology but provide a an in-depth scope to this field of stud as it has never been attempted before therefore depicting the efforts as a potential to very interesting results to be shared with the whole world.

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