Chapter II

LITERATURE REVIEW

The disposal of untreated wastes into land and water bodies from tanneries results in land and water pollution as well as emission of green house gases like methane and carbon dioxide. This problem can be mitigated through adoption of eco-friendly waste-to-energy technologies for treatment and processing of wastes before their disposal. One such technologies which has gained popularity is biomethanation, an environmentally benevolent technology, which leads to generation of energy from wastes besides rendering wastes suitable for land application as a rich source of organic manure. This not only reduces the quantity of wastes, but also improves its quality to meet the required pollution control standards.

The value of tannery biosludge and fleshing as organic wastes have been identified and recycling of such hazardous waste is possible. Hence, it is imperative to review the literature on current scenario of tannery industry, environmental impacts due to tanneries, technological options for recycling of tannery waste, biomethanation of tannery wastes, factors influencing biomethanation, pretreatment of substrates and digested slurry as an organic manure.

CURRENT SCENARIO OF TANNERY INDUSTRIES

The major leather production centers in the world are found in Mexico, Brazil, Japan, China, India, Pakistan and Bangladesh. The Indian leather sector is considered the fourth largest foreign exchange earner (Thiagarajan et al, 1994). Tanning in India has a long history and has been a traditional occupation for subdued groups of people.
The CLRI (1,987) estimated the total employees in tanneries during 1987-88 were 65,814; of which 70 percent of the employment was in the SSI (Small Scale Industries) sector and the rest in the larger industrial sector. According to Dixit (1995), the Indian tanning industry currently employs 82,299 workers and the estimated production in a year ranges between 600 and 1500 million square feet of leather. However, Thiagarajan et al. (1994) pointed out that the potential production capacity was high (2250 million square feet per year).

Geographically, the industry is concentrated in three regions of India namely the states of Tamil Nadu (mainly Vellore, Dindigul, Erode and Trichy), West Bengal (mainly Calcutta) and Uttar Pradesh (mainly Kanpur). In Kanpur, leather tanning particularly blossomed during British colonial rule, and the need for foots, saddlery and harness equipment was high (Dixit, 1995). Most of the tanneries are located in the state of Tamil Nadu, which are concentrated mainly in North Arcot Ambedkar District (Vellore), Pallavaram, Chrompet, Dindigul, Erode and Trichy as large clusters. In Tamil Nadu, 70% of the tanneries are located on the banks of river Palar in Vellore district.

Around 170 tanneries are in the Kanpur area. The vast majority of the Indian tanneries (80%) are small scale, only 20% are of medium to large scale (Thiagarajan et al. 1994; Varadarajan and Krishnamoorthy 1993).

Tamil Nadu accounts for 52.3 % of the tanneries, followed by West Bengal (21.5%) and Uttar Pradesh (13.6%). The three states accounted for seven-eighth’s of the total tanneries in India. Most tanneries are located on river basins viz., Ganga river system in U.P. and West Bengal and Palar and Cauvery river systems in Tamil Nadu (CLRI Report, 1990).
India produces annually about 21 million cattle hides and 15 million buffalo hides that can be processed to leather and leather products. According to the CLRI, survey of hides and skins in 1987, the off-take rates for cattle and buffaloes in India were only 10.8 percent and 20.7 percent respectively. The off-take rate for bovine animals’ worldwide is 20.2 percent while the figure for U.S.A. is 35.7 percent. In the United States, over 20,000 hides are tanned per day of which 23.5% are with vegetable tannins and 76.5% with chromium (Hemingway and Karchesy, 1989).

There are 270 registered tanneries in Bangladesh and 90 percent of these are located at Hazaribag on about 25 hectares of land. Most of these are semi-mechanized units, which use old processing methods. During the peak period, 15,000 labourers work daily in these tanneries, the numbers go down to 8,000 - 12,000 during the lean period. (People’s Report, 2002-2003).

Baskaran (1997) reported that the quantity of total wastewater discharged for 100 kg of skins/hides accounts to 3,000- 3,400 litres. Yadav (1988), points out that Jajman tanneries generate about 400 tonnes of solid waste per day, including hair, trimmings and fleshings, sludge, salts, shavings and vegetable tannins. According to World Bank (1999), the solid wastes can represent up to 70% of the wet weight of the original hides. Large quantities of sludge are also generated in the tannery while treating its effluent. Throughout the world, the environmental influence of tanning area is of major concern and impediment to the tanning industry (Huber and Satyendra, 1990).

In India, approximately 1,50,000 tonnes of tannery wastes in the form of raw hide trimmings, limed animal fleshings, green animal fleshings, hide splits and chrome shavings were available during leather processing which are not utilized or under
utilized, thus creating a solid waste disposal problem in tanneries (Muralidahara Rao, 1994).

Hertin et al. (2001) recognized that total changes required a reappraisal of effluent treatment. The sludge should be readily treatable and the need for sulphide oxidation should be eliminated. At present, this latter process presents a significant health and safety risk. In Tamil Nadu state where more than 60 percent of India’s economically important tanning industry is located, tannery work containing chloride and sodium compounds has, over many years, contaminated 55,000 hectares of agricultural land. Soils sampled near tanneries in the Vellore area of India had chromium concentrations of upto 7% (70,000 mg/kg) (Australian Centre for International Agricultural Research, 2003).

In Dindigul, vegetable tanning process is mostly used. Dindigul (India) is an important centre for leather processing with 63 tanneries. Around 25,000 to 30,000 tonnes of hides are processed in Dindigul and accounts to 6-7 percent of the total quantity processed in India. Minimum effluent produced is 3,000 to 4,000 liters per 100 kg of hides processed.

ENVIRONMENTAL POLLUTION DUE TO TANNERIES

Increasing industrialization, agricultural and commercial activities have brought about many changes in the environment. Although India has had relatively stringent environmental regulation for the past 10-15 years, the country continues to encounter enormous environmental problems; many of these are a result of industrial activity.

While manufacturing of leather, leather goods, fur products etc., solid wastes along with high amounts of wastewater are generated. The uncontrolled release of tannery
effluents to natural water bodies increases the risk for human beings and causes environmental pollution.

The production of finished leather from raw hides and skins can be grouped under three major operations such as beam house operations; tanning operations; and post tanning operations. All aspects of these operations were found to be potential sources of waste generation, although the beam house operations - where the raw hides are cleaned and prepared was found to contribute over 50% of the tanneries total pollution load. The generation of pollution is significantly high in the pre tanning operations compared to the post tanning operations (Puvanakrishnan et al., 1988).

The pollution load from the tanning activity has been estimated to be 50% more in weight than the weight of the hides processed (Gherdaker, 1998). Taylor (1953) reported contamination of well water supply when tannery wastes are disposed off by careless drainage. Wastewater generated is by far the most important environmental challenge being faced by Pakistan’s tanneries (Iqbal, 1998).

Tannery effluent is potentially hazardous since it contains large amounts of sodium chloride, which is used in the leather industry as preservative of hides and skins. The productivity of soil decreased when tannery wastes were applied in fields and some parts of the land became completely infertile. Germination of *Oryzxi sativa* (paddy) seeds was not satisfactory also resulting in stunted growth of *Lycopersicon esculentum* (Thabaraj et al. 1964). Obnoxious odour in the vicinity of tanneries are of constant concern and pollution caused by the extremely toxic sodium sulfide used in the dehairing process step as has been reported by Roth et al., 1995.

Rapid industrialization in India has resulted in substantial increase in the liquid waste which is traditionally discharged into open land or into nearby water bodies,
causing a number of environmental problems. Tannery effluent wastes are ranked as primary pollutants among all industrial wastes (Eye and Lawrence, 1971). Tannery effluent also contains large amount of organic and inorganic compounds, which are toxic to aquatic plants, and other organisms (Banerjea and Motwani, 1960).

Rajagopalan and Davies (1967) reported that the productivity of the soil decreased when tannery wastewater was applied and the land became infertile. The cultivable land in North Arcot District in Tamil Nadu declined from 68,000 to 22,000 hectares due to tanneries (India Today, 1986). Alexander (1961) observed that plants with reduced root system have caused less nitrogen intake when treated with tannery effluent.

Improperly treated wastewater has severe impacts on surface water, ground water and soil quality and these have recently been the main reasons for the mass closures of tanneries in the state of Tamil Nadu. The amount of wastewater varies between 30 and 50 liters per kg of processed skin (Alexander et al., 1992; Prasad, 1991).

Hariharan (1968) reviewed the effect of discharge of wastewater from a group of tanneries in the dry bed of Palar river contaminated domestic and irrigational subsurface water. Bose (1994) studied the quality of drinking water around the leather industries concentrated in Kanpur area and reported that the drinking water contains high concentration of sulphate and sulphide and that it affects plants, animals and human beings.

The sensitivity to chromium by different species of aquatic organisms varies greatly. Hexavalent chromium is a strong oxidizing agent, and therefore more toxic than trivalent chromium. Chromium deactivates cellular proteins, the lethal levels for fish range from 17 to 118 mg/l, 0.05 mg/l for invertebrates, and 0.032 to 6.4 mg/l for algae (Anonymus, 1974). The high value of chlorides in effluents pollutes the drinking water
and making it suitable for easy growth of microbes, leading to water borne diseases (Arora and Chattopadhy, 1975). Ahmed et al. (1977) studied the pollution effects of tannery water and reported that the salts in tannery effluent percolate through the soil, thereby causing severe salinity of the land.

About 90 percent of Hazaibag tannery workers die before they reach the age of 50 due to unhygienic working environment. About 58.1% workers suffer from ulcer, 31.28% have skin diseases, 16.76% suffer from malnutrition, 11.73% have high blood pressure and 10.61% suffer from rheumatic fever. This was revealed in a study conducted by the Society for Environment and Human Development (SHED, 1994); a Dhaka- based non-governmental organization. A study conducted by the Allergy, Asthma, Environment Research and Skin Care Institute, Bangladesh, found that most of the residents in the area suffer from a number of diseases. Chromium and iron levels of serum, urine and hemoglobin in tannery workers and unexposed workers were measured. The result suggested chromium adversely affects iron metabolism, possibly associated with excessive hexavalent chrome accumulation (Kornhauser et al, 2002).

Under certain environmental conditions, Cr III may be oxidized to Cr VI compounds, which are carcinogenic (Pettine, 1990; Fendrof, 1972). Such compounds become a threat to the environment as reported by Chuan, 1996. Chromium and arsenic, which are the major components of tannery effluent, affect the health of the ecosystem (De, 1990; Dara, 1998). The wide range of chromium applications has resulted in its occurrence as a common contaminant in soils (Proctor et al, 1997). Inside the tannery, chromium should be handled with care, since exposure to elevated concentrations of chromium in the air (>0.1 mg/m3) may lead to lung cancer (Anonymus, 1974).
According to a report by Blacksmith Institute, U.S.A. (2006), one of the world’s ten most polluted places is Ranipet (India) polluted by chromium and azodyes released from tannery industries. The high pollution level of the Palar river in Tamil Nadu because the tanneries are completely dependent on the river for their fresh water used in the production processes and their effluents releases (Tehelka, 2005). According to World Health Organization, about half a million residents of the Bangladesh capital, Dhaka, are at risk of serious illness due to chemical pollutants from tanneries near their homes (WHO, 2001). According to the news report in Taunton Gazette, 6,200 citizens in Southeastern Massachusetts were affected by cancer due to local dumpsite of tannery wastes (Taunton Gazette, 2006).

The increased mortality from bladder cancer is due to exposure to benzedrine based leather dye was reported by Montanaro et al. (1997). Cross sectional surveys in 1988 and 1994, revealed that tannery workers in Kanpur exposed to chemicals, dust and awkward ergonomic conditions, and health status, showed that 44% of workers have lower BMI than workers without exposure (Ory et at, 1996).

According to Dindigul District Collectorate, 4,952 acres of agriculture land in some villages of Dindigul were worst affected owing to discharge of effluents from 107 tanneries. About 6,863 farmers in these villages were affected according to press report released by THE HINDU, 2005. About 10,000 acres of agricultural land was seriously affected by the tanneries of North Arcot District and 30,000 acres of land was moderately affected in Vaniyambadi, Ambur, Vellore, Dindigul, Visharam and Timur township areas (Sastry and Prasad, 1980).

Apart from the chemical effluents, 385.6 kgs of solid waste is generated for every tonne of hide processed. No technology is in place to handle the solid wastes. Solid
wastes including sludge and animal fleshing, hair are either dumped or buried in trenches according to a report released by FRONTLINE, 2002. Van Villmen (1972) reported that the high amount of organic matter released by tanneries as solid waste inhibits the enzymatic activities of plant growth.

The Tamil Nadu Pollution Control Board (TNPCB) estimates that about 1,500,000 tonnes of solid wastes accumulated over two decades of plant operation are stacked in an open yard (three to five meters high and on 2 hectares of land) on the facility premises thus contaminating the ground water. Chandra et al. (2004) reported on the leachates form tannery solid wastes (TSW) and their possible genotoxic effects on the somatic cells of *Vicia faba*. The root tips examined for cytogenetic damage revealed that leachate of TSW significantly inhibited the mitotic index and induced significantly frequent chromosomal and mitotic aberrations in a dose-dependent manner.

If the solid waste with organic matter had been dumped in the open to decay, then the methane generated can escape into the atmosphere and contribute to global warming. By digesting the organic matter in a closed system, the methane can be tapped and used as fuel. Shallow open dumping is implicated very strongly with emissions of methane and carbon dioxide. Methane is a green house gas (GHG) with a large GHG potential about 23 times higher than that of carbon dioxide (IEA, 2004). The methane emission is seen to last many years (20-25 years) after the material has been dumped (Aitchison, 1996). Methane concentrations have risen from 700 ppb in pre-industrial times to 1,750 ppb (IPCC, 2001).

Selected metals were evaluated in surface sediments from Cadeia and Feitoria rivers of Brazil potentially affected by tanneries. Analysis of sediment data allowed the identification of critical metals and priority areas for biological monitoring, (Kolowski
et al., 2006). Mohammed Mahabubur Rahman et al. (2007) investigated the indigenous plant species, which are well adapted to the pollutant environment as effected by industrial effluent, which can accumulate higher concentration of heavy metals of dying and tannery industries around Dhaka city in Bangladesh.

Michael Aruldhas et al. (2006) reported on in vivo spermatotoxic effect of chromium as reflected in the epididymal epithelial principal cells, basal cells and intralepithelial macrophages of a non-human primate. Result of the study indicates that the abundance of basal cells and intralepithelial macrophages and the content of LF material in these cell types increased. They also proved that occupational or environmental exposure to Cr VI would occur in the tannery, soap and other industries in developing and under developed countries.

Savitha et al. (2006) evaluated that the histological studies of clitellar region were performed in control and in the individuals subjecting them to sub lethal (3.7%); LC 50 (37%) and acute (50%) concentrations of tannery sludge. The clitellar region was removed, fixed, sectioned, stained and photomicrographed to study the effect of tannery sludge. Results revealed that clitellar region of Eudrilus eugeniae showed varied degrees of damage by the effect of tannery sludge. Chandra et al. (2006) evaluated the in vivo genotoxic potential of leachates made from three different kinds of industrial waste (tannery, metal based waste, and waste containing dyes and pigments) that are disposed of in areas adjoining human habitation. Exposure to the leachates resulted in significant (P< 0.05 or P< 0.001) dose-dependent increase in chromosome and DNA damage. They also proved that the cytogenetic abnormalities and DNA damage produced by the leachates indicate that humans
consuming water contaminated with these materials are at increased risk of developing adverse health consequences.

Acute and sub acute effects of tannery waste on phosphatase enzymes have been observed. Acid and alkaline phosphatase enzymes of liver, kidney and gills of *Notopterus notopterus* (Asiatic knife fish) on exposing to tannery waste for 5, 10 and 15 days for 8, 16 and 24 hours has been studied by Ritu *et al.*, 2005. To assess the extent of groundwater deterioration due to tannery industries in and around Dindigul has been carried out. The concentration of cations such as Ca$^{2+}$, Mg$^{2+}$, Na$^+$, and K$^+$ and anions such as HCO$_3^-$, SO$_4^{2-}$, Cl$^-$ and NO$_3^-$ in the ground water have been studied by Mondal *et al.*, 2005.

Nath *et al.*, 2005 reported on the influence of different levels of tannery treated effluent and their corresponding concentration of chromium (Cr$^{6+}$) on seed germination and seedling growth in *Raphanus sativus* L. (Radish). This study showed reduction in seedling growth and related enzymatic activities with increase in concentration of Cr$^{6+}$ in treatments. The toxicity of leather tannery effluent affecting a population of *Mytilus edulis* (common mussel) in the Colligan estuary, Ireland was investigated. The growth condition and chromium concentrations were measured in transplanted, local and control mussels. At the cellular level, the degree of lipid peroxidation was measured in the tissues of field-sampled mussels (Walsh *et al.*, 1997).

**TECHNOLOGICAL OPTIONS FOR RECYCLING OF TANNERY WASTES**

Indian tanneries are generally outdated, use obsolete technology and depend excessively on traditional skills. Energy can be recovered from the solid wastes either by adopting thermo-chemical conversion such as incineration, gasification and pyrolysis
or by biological-conversion namely biomethanation or anaerobic digestion. Thermochemical conversion process involves thermal decomposition of organic matter to produce either heat energy or fuel oil or gas. For this, the wastes containing high percentage of non-biodegradable organic matter and low moisture content are more suitable. The most common way to manage tannery solid waste is by disposing of it on controlled land sites (Ferbabdez-Sempere et al, 1997).

The thermal treatment of sludge involves incineration, gasification and pyrolysis as a means of disposal, along with recovering energy from waste was reported by Hikmet et al. 1994. Pre treatment of tannery wastewaters by an ion exchange process for Cr III removal and recovery was reported by Tiravanti et al, 1997. Onur Yilmaz et al. (2007) evaluated on three different types of tannery wastes (chromium and vegetable tanned shavings and buffing dust) were pyrolyzed. Gas, oil, ammonium carbonate and carboneous residue were obtained.

Biological treatment of hazardous waste is becoming a realistic option in the treatment strategies for hazardous wastes. It is most useful process with less cost. The method is selected based on the type of pollutant, type of degradation (aerobic or anaerobic) and various other parameters (Chakraborthy, 1972).

Singaram (2001) reported on the biological treatment of tannery effluent polluted water after removal of chromium. Treatment of tannery effluents with hydrophytes viz., Chara intermedia, Typha angustifolia, Hemarthria compressa, Pistia stratiotes, Mars ilea minuta and Salvinia natans showed a reduction in the heavy metal component of the effluents after growing them in tannery effluent (Ejaz et al, 2005). Aravindan et al. (2004) reported that brown seaweed (Turbinaria ornate) pretreated with sulfuric acid
acid, calcium chloride and magnesium chloride could remove chromium from tannery wastewater. Deepak and Gupta (1991) studied the physicochemical methods such as reduction, precipitation, adsorption, ion exchange and solvent extraction processes, which are available for the removal of Cr$^{6+}$ from industrial wastes. The oldest and most frequently used method for removal of Cr$^{3+}$ from wastewater is precipitation (Joseph, 1985; Santiago, 1993).

Antonio Perez et al. (1999) reported that treatment of tannery effluent by reverse osmosis and ultra filtration. Polyamide membrane was used to perform an efficient separation of chromium III contaminated in the tanning wastewater. The government is now planning to promote reverse osmosis technology to improve the quality of effluent being discharged. A pilot plant using reverse osmosis to deal with this problem has been tested in Vellore (Tamil Nadu) (Warrier Gopikrishna, 2001). Scholz et al. (2005) investigated the use of a combined membrane bioreactors and reverse osmosis treatment process to treat tannery effluents to an acceptable level for irrigation purposes. This treatment reduced the COD, BOD and ammonia concentrations of the effluent by 90-100%. Ugurkurt et al. (2007) experimentally evaluated COD reduction potential of leather tanning industry wastewaters by Electro-Fenton (EF) oxidation.

Solidification is another process that has been considered as an alternative solution to the disposal of sludge containing heavy metals. In this process, waste materials are mixed with various binding media to obtain new product with improved physical properties. Recent papers have reported the use of solidification stabilization and vitrification processes to recycle different wastes such as low-level debris (Singh et al., 1998). The application of advanced oxidation processes (H$_2$O$_2$/UV, TiO$_2$/H$_2$O/UV and
TiO$_2$/UV to treat tannery wastewater was investigated in batch and continuous UV reactors using TiO$_2$ as a catalyst (Sauer et al., 2006). Global Environmental Facility (GEF) has supported the setting up of biomethanation plant at the Melvisharam common effluent treatment plant in Vellore district. The plant can handle two tons of CETP primary sludge and three tonnes of fleshing every day (Warrier Gopikrishna, 2001).

Dweek et al. (2000) and Filibeli et al. (2000) studied the solidification of tannery wastes with cement. They proved that the final product is resistant to aggressive environmental agents and can be safely stored in landfill sites. The effect of ultrasonic treatment on flocculating settling of tannery effluent and removal of COD was studied. The influences of irradiation method and duration of ultrasonic as well as prescription of coagulants on the efficiency of minimizing organic chemicals in wastewater were investigated by Li-Guoying et al. (2001). Swarnalatha et al. (2006) studied the solidification/stabilization of thermally treated toxic tannery sludge. This study proved that the sludge was subjected to starved-air combustion at 800°C, which prevented the conversion of Cr$_3^+$ to Cr$_6^+$. Aravindhan et al. (2007) studied on an integrated chemo- enzymatic methodology has been explored which would minimize or to some extent eradicate the unsafe chemicals involved in the process to provide a clean environment. This modified process results in decrease in COD and TS loads by 67 and 78% respectively, as compared with control process.

Tania et al. (2002) studied the environmental and technical aspects of the utilization of tannery sludge as a raw material for clay products. The environmental characterization of the product indicates that a material containing 10% tannery sludge; can be used safely. A qualitative assessment of the risk of human exposure to dust was
made throughout a commercial Kenyan tannery. An optical setup using microscopy and digital imaging techniques was used to determine dust particle numbers and size distributions (Mwinyihija et al., 2005).

Vital et al. (2004) evaluated the biodegradability and toxicity of diluted unhairing wastewater after being treated by an activated sludge system. Goltara et al. (2003) reported on carbon and nitrogen removal from tannery wastewater with a membrane bioreactor. Ganesh et al. (2006) studied on respirometry combined with sequencing batch reactor as an effective way for the removal of COD in tannery wastewater. Toxic effect due to chloride and condensable tanning on anaerobic digestion of vegetable tanning wastewater was investigated at different hydraulic retention times viz., 24, 48 and 60 hr respectively (Vijayaraghavan and Ramanujam, 1999).

Gilet (1989) described a new system, which can virtually eliminate the difficulties of disposing safety wastes. In the process waste is centrifuged, the proteins and other residues separated off as sludge. Salt is recovered from the aqueous phase by the use of vacuum evaporator and can be reused. The sludge may be further treated and sold as fertilizer. Rodrigues et al. (2008) evaluated the electrochemical treatment of tannery effluent and the results indicated a remarkable removal efficiency of more than 98.5% for all ion species present in effluents.

Ramadori et al. (2006) earned out an investigation at lab scale to assess the effectiveness of an innovative technology for treating municipal/industrial wastewater. An efficient adsorption process was developed for the decontamination of trivalent chromium form tannery effluents. Activated carbon was prepared from coconut shell fibers, which was characterized and utilized for Cr III removal from wastewater. A commercially available activated carbon fabric cloth was also studied for comparative
evaluation (Mohan et al, 2006). Swarnalatha et al. (2006) studied the starved air combustion on solidification/stabilization of primary chemical sludge from a tannery. The dried sludge was incinerated at 800° C in an incinerator under starved oxygen supply to prevent the conversion of Cr $^{3+}$ to Cr $^{6+}$. The efficiency of starved air combustion was studied under the different loading rates of the sludge.

Batch test stripping was the most effective process for sulphide removal compared to precipitation. Also, anaerobic degradation velocity was higher if sulphide was removed by stripping. In fixed bed reactors operated continuously, stripping was shown to be a reliable method for eliminating hydrogen sulphide toxicity (Schenk et al, 1999). Rivela et al. (2004) reviewed a Chilean leather tanning industry and studied in terms of input/output analysis of beam house, tanyard and retanning process. In this study, it was suggested that some streams may be re-used but it is necessary to apply anaerobic or aerobic treatment first, depending on their organic load. Swarnalatha et al. (2008) investigated the starved air incineration of chrome buffing dust from a tannery. The efficiency of starved air combustion was studied under the different loading rates of the chrome buffing dust.

Sastry et al. (2005) reported on chrome shavings is the manufacture of leather boards and related products. Two processes have been developed to offer an alternative and better solution for the disposal of chrome shavings. The first process is preparation of parchment like membrane and the second process is related to development of leather like material. The utilization of the chrome shavings in preparation of those two products not only reduces the environmental pollution by at the same time value added products can also be obtained. The potential of Aspergillus sp. for removal of chromium was evaluated in shake flask culture in different pH, temperature, inoculum
load, carbon and nitrogen sources (Srivastava et al., 2006). They also demonstrated that five morphologically different fungi were isolated from leather tanning effluent, both Aspergillus sp. and Hirsutella sp. had higher potential to remove chromium.

The degradation of Navitan Fast Blue S5R, a very important commercial diazo dye in the tannery and textile industries was investigated. Pseudomonas aeruginosa decolourized this dye at concentrations upto 1200mg l⁻¹ and the organism was also able to decolourize various other tannery dyes at different levels (Nachiyar et al., 2003). The ability of a chromate reducing Pseudomonas aeruginosa strain, isolated from tannery effluent to survive and reduce chromate in the effluent of a tannery and an electroplating unit was evaluated by Ganguli and Tripathi (1999). Sivasubramanian et al. (2008) reviewed a hair shaving process is developed for dehairing of skins and hides using a bacterial alkaline protease preparation, completely eliminating the use of lime and sulfide with environmental excellence.

BIOMETHANATION OF TANNERY WASTES

Among various options available, biomethanation is the most suitable method of recycling tannery solid wastes because it produces energy and provides biodigested slurry. The energy crisis in the 1970’s prompted American research into alternative energy strategies and anaerobic digestion was one such option. This resulted in the first digester built in America in 1970 where the methane obtained was used for heat and power (Lusk, 1999).

Biomethanation has several advantages over thermo chemical conversion. Kispert et al. (1975) reported that the cost of producing methane is economically acceptable when compared with projected costs of natural gas. Thermo chemical processes are cost efficient (Murphy, 2004) but not applicable to the developing countries like India in
account of the low calorific value and high organic content of waste (Gerlagh et al. 1999). The anaerobic digestion of tannery soak liquor was studied using a UASB. COD removal reached 78% at an OLR of 0.5 kg COD m$^{-3}$ d$^{-1}$, a HRT of five days and a TDS concentration of 71 g L$^{-1}$. The combination of the UASB with an aerobic post-treatment enhanced the performance of the overall wastewater treatment process and COD removal efficiency by 96% (Lefebvre et al., 2006). Vasudevan and Ravindran (2008) studied on recycling of biological tannery sludge through biomethanation and they reported on biogas production revealed that higher quantity of gas is produced when the biological sludge is added at an optimum level and it depends on reduction of carbon and pH of the feedstock materials. The influence of lipid concentration on hydrolysis and biomethanation of a lipid-rich (triolein) model waste was evaluated in batch by Cirne et al. (2007).

Biomethanation is a series of chemical reactions during which organic material is decomposed through the metabolic pathways of naturally occurring microorganisms in an oxygen-depleted environment. The main advantages of such a technology are to recover energy, reduce the sludge volume, reduce the pollution transformation load and limit odour. The metabolic reactions that occur during the anaerobic digestion consist of a complex series of reactions, which are catalyzed by consortia of microbes. Barnes et al. (1987) reported that the anaerobic degradation of organic compounds consists of several biological reactions and nutritional interaction involving several groups of microorganisms. The reactions in stages are termed as hydrolysis, acidogenesis, acetogenesis and methanogenesis (Cho et al., 1995). Various groups of bacteria are involved in such a transformation.
Carbon flow diagram of the biogas process adapted from Angelidaki et al. (2002)

Hydrolysis

The first stage of methanogenesis is hydrolysis, or liquefaction, hydrolytic bacteria convert the complex polymers to their respective monomers. For example, celluloses are converted to sugar or alcohols, proteins to peptide or amino acids, and lipids to fatty acids. This is carried out by several hydrolytic enzymes (cellulases, amylases, lipases etc.) secreted by microbes (Themelis and Verma 2004).
**Acidogenesis**

The acid forming phase or acidogenesis immediately follows hydrolysis. In this process, acidogenic bacteria turn the products of hydrolysis into simple organic compounds, mostly short chain (volatile) acids e.g. (acetic, propionic, formic, lactic, butyric or succinic acids) and alcohols. The specific concentrations of products formed in this stage vary with the type of bacteria as well as with culture conditions, such as temperature and pH (United Tech, 2003). The acidogenic bacteria have a higher growth rate than methanogens and acetogens (Lin et al., 1997; Bhattacharya et al., 1996). Thus, most often the conversion efficiency of the feed stock is determined by the hydrolytic and fermentative action. Typical reactions in the acid forming stages are shown in two reactions.

\[
\text{C}_6\text{H}_{12}\text{O}_6 \rightarrow 2\text{CH}_3\text{CH}_2\text{OH} + 2\text{CO}_2 \\
\text{(Glucose)} \quad \text{(Ethanol)}
\]

\[
\text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2 \rightarrow 2\text{CH}_3\text{CH}_2\text{COOH} + 2\text{H}_2\text{O} \\
\text{(Glucose)} \quad \text{(Propionic acid)}
\]

**Acetogenesis**

Acetogenesis occurs through carbohydrate fermentation, through which acetate is the main product, since about two thirds of the methane formed in a biogas reactor is derived from acetate (Gujer, 1983). Decrease in the activity of the acetate utilizing methanogens severely affects the anaerobic digestion process. The result of this process is a combination of acetate, carbon dioxide and hydrogen. The role of hydrogen as an intermediary is of critical importance to anaerobic digestion reactions. Typical reactions in the acetogenic stage involve the conversion of glucose and ethanol to acetate.

\[
\text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2\text{O} \rightarrow 2\text{CH}_3\text{COOH} + 2\text{CO}_2 + 4\text{H}_2 \\
\text{(Glucose)} \quad \text{(Ethanol)}
\]
\[
\text{CH}_3\text{CH}_2\text{OH} + 2\text{H}_2\text{O} \rightarrow \text{CH}_3\text{COO}^- + 2\text{H}_2 + \text{H}^+
\]

\[\text{(Ethanol)} \quad \text{(Acetate)}\]

**Methanogenesis**

A group of bacteria called methane producers (Methanogens) produces methane in the fourth stage called Methanogenesis. Methanogens derived the energy for growth only by methanogenesis and are the only organisms known to produce methane as a catabolic product (Thayer, 1998). Many microbiologists consider Archaea to be the “third form of life” along with prokaryotes and eukaryotes (Sowers and Schreier, 1995). Methanogenic archaea, a diverse group of obligate anaerobes, occurring in most anaerobic habitats, form the terminal electron sink during anaerobic digestion of organic matter to methane (Boone et al., 1993). Methanogenesis takes place either by means of cleavage of acetic acid molecules to generate carbon dioxide and methane, or by reduction of carbon dioxide with hydrogen. Methanogenesis is expressed by the following equations (Karki and Dixit, 1984).

\[
2\text{CH}_3\text{CH}_2\text{OH} + \text{CO}_2 \rightarrow 2\text{CH}_3\text{COOH} + \text{CH}_4
\]

\[\text{(Ethanol)} \quad \text{(Acetic acid)} \quad \text{(methane)}\]

\[
\text{CH}_3\text{COOH} \rightarrow \text{CH}_4 + \text{CO}_2
\]

\[\text{(Acetic acid)} \quad \text{(methane)}\]

\[
\text{CO}_2 + 4\text{H}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O}
\]

\[\text{(methane)}\]

According to a report by International Union of Leather Technologists and Chemists Societies, limed fleshing mixed with tannery sludge are digested to produce methane by grinding and warming to allow microbiological activity with increased fat or grease content resulting in increased methane production. The volume of gas evolved
(containing 75% methane) is estimated to be 615 liters per kg of organic material introduced into the digester, after 25 to 30 days at 35°C.

Sludge treated with 80% fleshing (20% dry matter) and 20% raw sludge (5% dry matter) with a retention time of 26 days produces 0.9 M³ biogas per kg of volatile solids.

**Types of biodigesters for biomethanation:**

The biodigester is a physical structure, commonly known as the biogas plant. Since various chemical and microbiological reactions take place in the biodigester, it is also known as bioreactor or anaerobic reactor. The main function of this structure is to provide anaerobic condition within it. It can be made of various construction materials and in different shapes and sizes. Conventional anaerobic digesters are in batch, semi-continuous or continuous operations. Semi-continuous or continuous operations are preferable as maximum growth rate can be achieved constantly at steady state by controlling the feed rate. In batch system, the steady state cannot be achieved as the concentrations of components in the digester are changing with time (Klass, 1984). Some of the commonly used designs are floating drum digester, this model was put forth in 1956 by Jashu Bhai J. Patel. Fixed dome digester, this is the Chinese model biogas plant (also called drum less digester) was built in China in 1936. Deenabandhu model, this model was proposed in 1984 by the Action for Food Production (AFPRO), New Delhi, India. Bag digester was developed in 1960’s in Taiwan. It is made of PVC or red mud plastic. Plug flow digester is similar to the bag digester and it first used in South Africa in 1957. Anaerobic filter was developed in the 1950s. This reactor also known as Fixed film or retained film digester (Bioenergy Systems Report, 1984). A batch digester produces less gas, has a lower loading rate, and carries a risk of explosion
during emptying of the reactor (Vandeviviere et al, 2002). Upflow anaerobic sludge blanket (UASB) design was developed in 1980 in Netherlands. Many full-scale UASB plants are in operation in Europe using wastewater from sugar beet processing and other dilute wastes that contain mainly soluble carbohydrates (Bionergy Systems Report, 1984).

A number of scientists using the upflow anaerobic sludge blanket reactor (Sayed et al, 1987; Ganga et al, 1995; Ramesh Sivathanu and Kasturi Bai 1998) successfully treated tannery effluent. A study report presented by Rajamani et al. (1995) revealed that a pilot plant UASB reactor with 10,000 liters capacity could treat the tannery wastewater at 12 hours detention time with BOD removal of 60% and 70% COD removal. Lopez et al. (2003) studied the feasibility of the anaerobic degradation of three natural tannin extracts in three upflow anaerobic sludge blanket reactors, which were fed with increasing concentrations of two condensed (quebracho and wattle) and one hydrolysable tannin extract (chestnut). The sulphates in the raw tannery wastewater after anaerobic treatment in the lab scale UASB reactor consuming 0.85 g COD/g of $\text{SO}_4$. Generally, the amount of COD consumed will be 0.67 g COD/g of sulphate reduced as reported by Arceivalla (1998).

According to Renewable Energy World (January, 2006), In Cambodia about 400 low-cost, plastic tube digesters have been installed by various organizations such as Cel-Agrid of the University of Tropical Agriculture by the Cambodian Rural Development Team (CRDT) and the FAO-Telefood programme. Moog et al. (1997) reviewed the promotion and utilization of polyethylene biodigester in small hold farming systems in the Philippines. According to Ministry of Non-conventional Energy Sources, India (MNES) annual report 2004-2005, there is an estimated potential of 12
million domestic biogas plants in the country, a total number of 3.67 million biogas units had been installed by the end of fiscal year 2004. A large number of biogas plants are currently found throughout the world. In Denmark, the first centralized biogas plant was built in the 1980s. In 2001, Danish biogas plants treated approximately 1.2 million tonnes of manure and approximately 300,000 tonnes of organic industrial waste (Angelidaki and Ellegaard, 2002).

A study was carried out on kinetics of biomethanation of solid tannery waste and the concept of interactive metabolic control (Lalitha et al, 1994). During the 80 days study period, the overall decrease in volatile solid level was 65%, whereas the collagen level declined by 85% with 0.45 l of methane yield/g of volatile solid degraded. The study also revealed that anaerobic digestion of calfskin collagenous waste was optimized for a batch process based on accelerated maximal methane yield per gram of input volatile solid.

Treatment of tanneries waste liquors is also an important field that should be concerned about biomethanation. Yi Yang Wang (1998), studied on biomethanation of industrial effluents for energy recovery in China.

Ravindranath (1998) carried out a pilot study to assess the feasibility of anaerobic digestion of fleshing and subsequent biogas generation. An upflow anaerobic fixed film reactor, which retains a high concentration of accumulated biomass in the form of biofilm supported by a carrier, has been developed to treat tannery wastewater. The effects of major process variables such as Hydraulic Retention Time (HRT), Organic Loading Rate (OLR), and temperature on Chemical Oxygen Demand (COD) removal, and methane yield performances of the reactor were evaluated by Song et al. (2003). Different composition of waste fleshing and primary sludge was studied for anaerobic
digestion process carried out in a laboratory scale reactor to develop appropriate
technology for the recovery of bioenergy and to dispose the residue safely (Arumugam
et al, 2006).

Untanned wastes mixed with farming domestic and fish wastes can be used for
methane production; full-scale plants are in operation in Denmark and Sweden
(IULTCS, 2004). The application of upflow anaerobic sludge blanket process was
evaluated for the treatment of vegetable tannery wastewater. The biogas production
varied from 0.3 to 23.9 m³ with the average composition of 65% methane and 35%
carbon dioxide. Routh (2000) removed tannins, which are the complex components of
vegetable tannery waste efficiently. A study was carried out on demonstration of
biomethanation plant in treatment of leather processing wastes (Dhussa, 2002)

FACTORS INFLUENCING BIOMETHANATION

Several factors within the digester effect the physical environment and therefore the
rate of digestion and biogas production. Facility managers must monitor and maintain
the following parameters within the acceptable ranges: temperature, pH, nutrient
content, volatile fatty acids, C: N ratio and retention time.

Methane and carbon dioxide are the terminal fermentation products of organic
compounds in anaerobic environments (Baresi et al 1978). Biogas conversion process
is microbiological in nature and is affected by the following factors.

Temperature

Temperature has direct effect on physical-chemical properties of all components in
the digester and affects the thermodynamic and kinetics of the biological process. The
methanogens are inactive in extreme high and low temperatures. When the ambient
temperature goes down to 10°C, gas production virtually stops. Satisfactory gas
production takes place in the mesophilic range, between 25 to 37°C. Proper insulation of digester helps to increase gas production in the cold season. When the ambient temperature is 30 degrees C or less, the average temperature within the dome remains about 4°C above the ambient temperature (Lund Andersen and Torry Smith, 1996). Generally, temperature is one of the rate limiting factors in biological reactions and in the variety of intermediate compounds that accumulate (Maly et al., 1971). Anaerobic digestion occurs under two main temperature regimes namely, Mesophilic (between 20-40°C usually 35°C) and Thermophilic (between 50-75°C usually 55°C). Some research has shown that temperature is one of the most important factors affecting methanogenesis processes in a biogas digester (Castillo et al., 1995; Chynoweth and Pullamanapallil, 1996). The optimum temperature for anaerobic digestion is either 35°C or 55°C, mostly depending on the application and its operating conditions (Hessami et al., 1996).

Aceticlastic methanogens are primarily mesophilic bacteria. Most species of the genera Methanoseta are mesophilic, having an optimum temperature range of 35-45°C. Very little methane production occurs during lengthy incubation at 10°C; however, cells can remain viable and continue to grow and produce methane during subsequent incubation at 35°C. Hill (1984), conducted an experiment on anaerobic digestion of poultry droppings at 35°C for a retention period of 30 days and he reported that the gas production was 0.36 m³/kg of volatile solids added. No growth or methane production occurs during incubation at 45°C, and cells are no longer viable after 4 weeks of incubation at this temperature (Patel, 1984). As temperature rises, enzyme activation increases while at the same time enzyme denaturation also increases. In addition, higher temperature also increases the irreversible destruction of many of these vital
proteins. Such mechanisms will cause a typical bacterium to have a range of temperature viability as well as an optimum temperature for growth and functioning (Harmon et al. 1993). Some work has been done on the methanogenesis of cattle waste and thermophilic methane production (Varel et al. 1997; Wiegant and Lettinga, 1985). Tanticharoen et al. (1985) reported that solids degradation and biogas production were higher in thermophilic digester compared to mesophilic digester.

**pH**

One of the environmental factors associated with optimizing methane production within a digester is pH (Bhadra et al. 1984; Brunmeler et al. 1985; Manson et al. 1991; Speece, 1996). In general, the anaerobic fermentation process of methanogenesis is impaired at pH values below 6.5 and above 8.2 (Anderson et al. 1982). pH is known to affect enzymatic activity owing to the fact that only a specific and a narrow pH range are often suitable for the maximum activity.

There is significant variation in the values reported in literature for the optimum pH required for methanogens. A pH range between 6.7 and 7.4 is reported to be suitable for most methanogenic bacteria to function (Verma, 2002). The rate of methanogenesis may decrease if the pH is lower than 6.3, the main reason for this is the absence of biodegradation due to the rapid acidification of the waste (Verma, 2002). This is generally due to the over production of VFA by the activity of hydrolytic acedogenic bacteria capable of degrading the waste in the first few days of incubation (Gomec, 2003).

**Ammonia**

Ammonia produced in the protein degradation may cause problems in anaerobic digestion as unionized ammonia inhibits anaerobic microorganisms, particularly
methanogens. Free ammonia (NH₃), at high enough concentrations is considered toxic to methanogenic bacteria. McCarty (1964) reported a toxicity concentration range of 1500-3000 mg/l as NH₄-N at pH above 7.4 with 3000 mg/l being toxic at any pH.

Sterling et al., (2001) determined the feasibility of monitoring hydrogen gas as an indicator of the digester being upset due to ammonia overloading. This experiment also revealed the amount of ammonia nitrogen in the digester feed impacted digester hydrogen production, methane production and volatile solid removal. Small increases in ammonia nitrogen resulted in hydrogen and methane production whereas large increases caused the inhibition of hydrogen and methane production.

**Nutrient content**

To optimize growth and methane production, methanogens require certain nutrients and trace metals. All methanogens utilize ammonia as a source of nitrogen, and all methanogens appear to require nickel, cobalt, and iron (Jarrell and Kalmokoff, 1988). Speece (1996) recommends that nitrogen in the form of NH₄ is to be provided to stimulate methane production. Sufficient nutrients are also important to microbial cell growth. Macronutrients such as carbon, hydrogen, nitrogen and oxygen are the main components in biomass cells. Sulphur, phosphorus, potassium, calcium, magnesium and iron are required for specific proteins (Kayhanian and Rich, 1995).

A few researchers suggest that additional trace metals are necessary for methanogenic growth. Selenium, molybdenum, manganese, aluminum and boron have been recommended as additional components in the media (Brummeler et al. 1985; Yang and Okos, 1987; Azbar et al. 2000). Baresi et al. (1978) conducted studies on enriched cultures of *Methanosarcina* sp. by varying the concentrations of yeast extract
from 0.01- 0.5%. Results showed a dramatic increase in the rate of methane production in medium containing 0.1-0.5% yeast extract.

**Volatile fatty acids**

The long chains fatty acids were found to be inhibitory to the several kinds of essential reactions in the anaerobic digestion because of their toxicity to the bacteria (Hanaki *et al.*, 1980). The rate of methane production from hydrogen was lowered by long chain fatty acids. Wang *et al.* (1999) investigated the efficiency of anaerobic digestion to evaluate the effect of C2-C6 VFAs on the methane fermentation and to examine the behavior of VFAs in anaerobic digestion. At shorter retention times, VFA production could exceed the utilization rates, leading to the digester failure (Ghosh, 1991). Volatile fatty acid is a core potential inhibitor and the concentration of volatile fatty acids (VFA) is one of the best indicators for process instability (Hill and Holmberg, 1988; Mechichi and Sayadi, 2005).

Increasing the load gives more biogas production but risks overloading, which results in VFA accumulation. High concentration of VFA decreases pH, makes VFA become more toxic to the methanogens. Several studies have highlighted the importance of individual VFA as an early warning indicator for process failure (Ahring *et al.*, 1995; Cobb and Hill, 1991).

**Carbon to Nitrogen** ratio (C: N)

The C: N represents the relationship between the amount of carbon and nitrogen present in organic material. Optimum C: N ratios in anaerobic digesters are between 20:1 and 30:1. A high C: N ratio will lead to a rapid consumption of nitrogen by the methanogens and results in a lower gas production. On the other hand, a lower C: N ratio causes ammonia accumulation and pH value exceeding 8.5, which is toxic to methanogenic bacteria. The process of biomethanation is optimal with a C: N ratio of
around 30:1 (Cowley and Wase, 1981). Optimum C: N ratio of the feedstock materials can be achieved by mixing waste of low and high C: N ratio, such as organic solid waste mixed with animal manure.

C: N ratio of the feedstock materials was obtained by using the following formula so as to obtain the optimum ratio for biomethanation.

\[
R = \frac{Q_1 (C_1 x (100 - M_1)) + Q_2 (C_2 x (100 - M_2)) + Q_3 (C_3 x (100 - M_3)) + ...}{Q_1 (N_1 x (100 - M_1)) + Q_2 (N_2 x (100 - M_2)) + Q_3 (N_3 x (100 - M_3)) + ...}
\]

in which,
\[
R = \text{C: N ratio}
\]
\[
Q_n = \text{mass of material n (wet weight)}
\]
\[
C_n = \text{carbon (%)}
\]
\[
N_n = \text{nitrogen (%)}
\]
\[
M_n = \text{moisture content (%) of material n}
\]

**Retention time/ Residence time:**

Retention time is the time needed to achieve the complete degradation of the organic matter. The retention time varies with process parameters such as process temperature and waste composition. The retention time is determined by the average time it takes for organic material to digest based on the COD and BOD of the effluent. Zkang and Naike (1994) reviewed that retention time had a considerable effect on the methanogens. Recent research has shown that volatile suspended solids in a digester could be reduced by 64-85% only after 10 hours, but retention time of 10 days were typical for complete digestion (Lin et al., 1997; Vlyssides and Karlis, 2003). The retention time is also dependent on the temperature; the higher the temperature, the lower the retention time (Lagrange, 1979). In a cow dung plant, the retention time is calculated by dividing the total volume of the digester by the volume of inputs added daily.
In mesophilic digestion, the digester is heated to 35° C (95°F) and the typical time of retention in the digester is 15-30 days, whereas in thermophilic digestion at 55°C (131 °F), the time of retention is typically 12-14 days (Vandevivere et al. 2002).

Pretreatment of substrates for biomethanation

To increase the biodegradability of the wastes, a number of pretreatment processes and methods have been put into practice over the years. Pretreatment processes may be broadly classified as mechanical, thermal, chemical and biological. Several attempts have been made by modify (pretreated) water hyacinth to make it more suitable for biogasification. Among these are mechanical crushing, pulverization of dried water hyacinth and chopping of water hyacinth into fine bits (Chanakya, 1985).

Three types of pretreatment are reported in literature, viz., enzymatic hydrolysis, acid hydrolysis and alkali pretreatment for improved digestibility of lignin rich biomass waste. Organic materials, when subjected to high temperature with or without variation in pressure, breaking up make them more susceptible to hydrolytic activity during anaerobic digestion. Thermal pretreatment before digestion seemed to be the better alternative (Hang et al. 1998).

Alkali treatment of lingocellulosic materials has been used extensively to increase the biodegradation of municipal solid waste. In a study conducted with 0.2% NaOH, there was no noticeable change beyond 4 hours although the experiment which was conducted for 20 hours (Bellamy, 1974). Several researchers have increased volatile acid and gas production in anaerobic digesters by adopting alkali pretreatment methods. Gunaseelan (1974) reported that the high lignin content of parthenium was one of the
factors accounting for the low methane yield. Alkali pretreatment of parthenium significantly enhanced the methane production and cellulose reduction.

It has been demonstrated in a number of studies that a combination of chemical and thermal pretreatments has yielded increased bioconversion rates of MSW. Sawayama et al. (1995) reported that thermo chemical treatment of a mixture of primary and waste activated sludge at 175°C for one hour led to a 60% increase in biogas generation.

Pratap Chandran et al. (2006) reported that pretreatment processes for enhanced biomethanation of municipal solid wastes. Their studies also revealed that the future is in a combination of pretreatment methods, which will be more effective in ensuring maximum conversion and improving overall plant efficiency. Ling Chen et al. (2007) constructed a cascade process of rotational drum fermentation system with leachate recirculation from a methanogenic to the acetogenic reactor to enhance the hydrolysis and acidification of solid organic waste.

Higher biodegradability of organic matter can be achieved in pretreated plant residue supplement slurries, to get higher gas production than untreated substrates. Hassandar and Tandon (1987) found that the addition of pretreated (1% NaOH) to 7 days of lantana residue, wheat straw, and apple leaf litter to cattle dung improved microbial digestibility and biodegradability during anaerobic fermentation. There was an almost two fold increases in biogas and methane production. Muller and Trosch (1986) demonstrated screening of white-rot fungi for biological treatment of wheat straw for biogas production.

Thermal, mechanic and chemical treatment have been investigated as a possible pretreatment step to accelerate sludge hydrolysis. Different combinations of these treatments have been also studied (Chiu et al. 1997). Several works in the field of
bioengineering were earned out on enzymatic hydrolysis of collagen, which is the most representative protein of leather wastes (Kawahara et al, 1998; Matsushita et al, 1994; Asdormithee et al, 1994).

The main advantages of the use of enzymes are specificity, stereo specificity, and activity under mild conditions, possibility of producing ‘natural’ products, non-pollutants, and biodegradability (Vie, 1991). The microbial proteases are commercial enzymes, which have found wide applications in various industrial, biotechnological and other basic research fields (Mala et al, 1998; Weisman, 1993; Gupta et al., 2002). Proteases represent one of the most important groups of industrial enzymes and account for at least a quarter of the total global enzyme production (Herbert et al, 1992). Alkaline proteases have considerable application in leather tanning industry (Godfrey and Reichelt 1985; Dayanandan et al, 2003).

Zerdani et al. (2004) have studied digestion of solid tannery wastes by strains of Bacillus sp. isolated from compost. In their studies, they also proved that tannery wastes could be treated biologically by collagenase producing microorganisms. This enzymatic hydrolysis could be a safe method of recycling these organic materials. Isolation of a keratinolytic- producing Bacillus subtilis strain and the characterization of the exceptional dehairing properties of its subtilisin-like keratinase. This enzyme can be an alternative to sodium sulphide, the major pollutant from tanneries, and may completely replace it (Macedo et al, 2005). Bacillus subtilis, isolated from tannery waste, produced an alkaline protease at optimal activity when grown in a casein/gelatin medium in a stirred tank fermentor at 37 C with the dissolved oxygen tension at 40% air saturation. Optimum protease activity (223 U ml *) was at pH. 8.5 and was stable for 1 h upto 45°C but at 60°C lost 80% activity (Hameed et al, 1996).
Taylor et al. (1998) studies on the detoxification of tannery wastes have focused on the use of enzymes or the combination of alkaline treatment and enzymatic hydrolysis. A solid-state fermentation experiment was conducted on study of the biodegradation of chromium shavings and *Aspergillus carbonarius* (Katsitas, 2004). In this study, approximately 97% liquefaction of the tannery waste was achieved. *Streptomyces* spp. G-157 exhibited the highest potency for the production of extracellular proteases. After preliminary fractionation by differential precipitation two proteolytically active fractions, fraction 1 and 2 were obtained by Sampath *et al.* (1997). For solid organic wastes, the common pre-treatment method is to use hydrolysis reactor to liquefy the substrate before feeding into methanogenic reactor (Scherer *et al.*, 2000; Pavan *et al.*, 2000).

A pilot study was carried out to assess the feasibility of anaerobic digestion of fleshings and subsequent biogas production generation. Based on the laboratory results obtained, liquefaction of fleshings using UASB treated tannery effluent was found to be suitable (Ravindranath, 1998). Raju *et al.* (1997) have investigated chicken intestine as a source of proteolytic enzymes for the hydrolysis of tannery fleshings. A combination of tannery fleshings and chicken intestines at acidic pH, when incubated at 37°C, leading to optimum hydrolysis of tannery fleshings as evidence by tyrosine release. Altered amounts of protein content in the supernatant indicated an increased rate of hydrolysis of fleshings in the presence of enzyme.

Shahanara Begum *et al.* (2007) studied an intracellular protease, which was extracted and purified from *Pseudomonas aeruginosa* by ion exchange chromatography on DEAE-cellulose followed by CM-cellulose and rechromatography on DEAE-cellulose.

Vasudevan and Ravindran (2007) studied on biotechnological process for the treatment of tannery fleshing for methane generation. They evaluated the proteolytic bacteria can treat tannery solid waste (fleshing) biologically, thus enhancing the rate of biodegradability used for higher rate of biomethanition. A novel process for liquefaction of solid organic matter by biological method. This process is envisaged to have enormous application in tanning industry for effective disposal of various solid wastes, which otherwise add to the pollution load (Rengasamy Sunthararajan et al., 2007).

Proteases execute a large variety of functions and have important biotechnological applications. Proteases represent one of the three largest groups of industrial enzymes and find application in leather industry, food industry and bioremediation processes (Anwar and Sateemuddin, 1988; Gupta et al., 2002). A protease isolated from *Pseudomonas aeruginosa* PD 100 could act in the presence of SDS and Tween 80. The immobilized protease showed 15-20% increase in temperature stability and the entrapped enzyme retained 83% of its initial activity after six cycles. This protease finds potential application for waste treatment, used in detergents and leather industry (Mohsen Fathi Najafi et al., 2005).

Production of alkaline protease by *Pseudomonas aeruginosa* grown on protenacious solid waste (fleshing) generated during processing of leather. The isolated strain salt laden wastewater (soak liquor) of tannery was selected by its ability to greater protease production. When cultivated in minimal medium containing only fleshing as substrate (Ganesh Kumar and Sekaran, 2006).
Biological hydrolysis is identified as the rate-limiting step in anaerobic digestion (Tiehm et al, 2001; Li et al, 1992). Pfleiderer et al (1986) recognized the economic reasons, why enzymes from microorganisms have come to play a significant role in recent years and enzyme products of microbial origin are already being produced on a wide scale.

Apart from the chemical treatment of leather waste, treatment with enzymes at low temperature for short period, give products with commercial value. Enzymatic treatment of solid tannery waste gained importance in recent past due to environmental restriction imposed on disposal of wastes (Chakraborthy, 2003). Hoq (1985) described lipolysis as the “constructive” consequences of the ability of lipase to hydrolyse lipids so as to obtain fatty acids and glycerol, both of which have important industrial applications.

Lipolytic enzymes are currently attracting enormous attention because of their biotechnological potential (Jaeger et al, 1998). Among bacterial lipases, attention has been usually focussed on particular classes of enzymes such as the lipases form the genus *Pseudomonas*, which are especially interesting for biotechnology (Svendsen et al, 1995). Castell and Garrard (1941) reviewed that 85% of unpolluted well waters from rural areas were contaminated with proteolytic and lipolytic psychrophilic bacteria, many of these numbers of the genus *Pseudomonas*.

Kennelly et al. (1997) have studied a modified pork fat based agar method was specially developed to determine the lipolytic activity of starter strains used meat fermentations. Bean and Griffin (1990) described *Bacillus cereus* as an aerobic rod shaped bacterium that is widely distributed in the environment and that produces spores, lecithinase and enterotoxins.
Lipolysis was monitored based on determining the concentration of free fatty acids in milk, on the model case of Ultra High Temperature (UHT) treated milk. Contamination with spores of 15 *Bacillus subtilis*, *Bacillus cereus* and *Bacillus licheniformis* isolated from farm environment and raw milk. This study made known that significant lipolytic activity was detected in association with *Bacillus licheniformis* and *Bacillus cereus* (Janstová et al., 2006).

Akiosugihara et al. (2002) studied on purification and characterization of a novel cholesterol esterase form *Pseudomonas aeruginosa* isolated from soil with its application to cleaning lipid stained contact lenses. A novel, oil degrading bacterium strain (TO was isolated from a hot spring in Hokkaido, Japan. It efficiently degrades different types of fats and oils, including edible oil waste. When grown in a mineral salt medium containing 1% triacylglycerol (as salad oil), hydrolysis products were 1, 3 and 1,2 — diacylglycerols, monoacylglycerol and free fatty acids (Briones et al., 2004).

A novel, moderately lipolytic halophilic bacterium (strain SM 19) that displays lipolytic activity has been isolated and characterized (Martin et al. 2003). Heung chaejung et al. (2006) studied on the display of a thermostable lipase on the surface of a solvent resistant bacterium. *Pseudomonas putida* GM 730 and its applications in whole-cell biocatalysis.

Kumar et al. (2005) reviewed the interest in microbial lipase production has increased in the last decades, because of its large potential in industrial applications particularly leather (removal of lipids from animal skins) and medical (blood triglyceride assay). Bacterial lipases are involved in solution of such environmental problems as the breakdown of fats in domestic sewage and anaerobic digesters (Godfrey
et al. 1983). Several Bacillus sp. were reported to be the main source of lipolytic enzymes (Kim et al. 1994).

An extreme thermophilic bacterium, isolated from mangrove detritus, produced an extra cellular alkaline thermostable lipase. The bacterium was identified based on cell morphology, growth characteristics, as a strain of Bacillus alkalophilus (Essam et al. 2004).

**Digested Slurry - an organic manure**

Biomethanation results in the generation of three products namely biogas, digested slurry and spent residuals. Hons et al. (1993) utilized the sludge generated by the anaerobic fermentation of biomass to methane for land application. This study was done on the effects of loading rate of methane generator sludge on nutrient mineralization, plant growth and nutrients uptake and potential water pollution. Result indicated that the sludge can serve as a source of nutrients, especially nitrogen and phosphorous and may be used to help in neutralizing soil acidity.

One of the principal objectives of anaerobic treatment of solid waste is to generate significant quantity of biogas i.e., about 250-350 m³/ton of waste (NEERI report, 1996) and generated digested material was used as a high-grade soil conditioner (CPHEEO, 2000). A study was carried out on the interaction of metals, present in tannery waste and their tolerance in sunflower (Helianthus annus L.). The oil was extracted from the seeds of the plant and the level of oil content was increased upto 35 % due to tannery sludge application as compared to control followed by decrease at higher tannery sludge ratio (Singh et al., 2004).

Shraddha Singh et al. (2004) studied the scanning electron microscopic studies and growth response of Helianthus annum L. grown on tannery sludge amended soil. The
analysis of scanning electron micrographs of the leaf surface of *H. annum* grown on 50% and 100% tannery sludge after 90 days showed an increase in the frequency of stomata and trichomes. The root length of the plant increased up to 35% tannery sludge followed by significant (P<0.01) decrease at higher amendments, whereas the shoot length of the plant increased with increase in sludge amendment ratio at all the exposure periods, compared to their respective controls.

Thorstensen *et al.* (1979) reported on a technical and economic evaluation indicates that tannery sludge has definite value as a fertilizer based on its nutrient content. According to Biogas and Natural Resources Management (1998), the slurry has been proved high quality organic materials for plant nutrition. The technology is economically viable, technically feasible, environmentally sound and socially being increasingly more acceptable. Tannery effluent composted with cow manure and wheat straw for 90 days to reduce pathogens and toxic organic compounds was monitored by (Contreras *et al.*, 2004).

Application of tannery sludge increased C and N mineralization and could thus provide valuable nutrients to pioneer vegetation and no inhibitory effects on the biological functioning of the soil were found (Barajas *et al.*, 2001). Chawla (1984) reported that the addition of organic manures increases productivity of crops, since biologically originated organic manures contain all plant nutrients needed for the metabolism. Effect of different concentrations (1, 2.5, 5, 10, 25 and 50%) of tannery effluent on seed germination and early seedling growth of *Cajanus cajan* L. and *Oryza saliva* L. were studied by Bera *et al.* (1998).

Characterizations of effluents of various industries and their effects on soil and crops and management strategies to be adopted have been discussed by Yadav *et al.* (2006).
Gupta and Sinha (2006) reviewed the metal accumulation potential of *Chenopodium album* L. grown on various amendments of tannery sludge for a period of 60 days. This study also proved that shoot length of the plant increased by the addition of sludge, whereas, no marked change was observed in root.

An experiment was conducted to evaluate the tree species for areas affected with tannery effluent in Vellore district. The tree crops including neem, pungam, bamboo, cassia, eucalyptus and casuarina were planted at the normal spacing. Among the tree species, eucalyptus recorded higher percentage of establishment and plant height (Kumaran *et al.* 2003). Singh *et al.* (2004) conducted an experiment to study the effect of different amendments of tannery sludge on physiological and biochemical parameters of tomato plant (*Lycopersicon esculentum* L.). The statistical analysis of the results showed on increase in chlorophyll and protein contents in lower sludge amendment ratio at all exposures however, a decrease was at 100% sludge amendment.