Preliminary Evaluation of Novel Plant Mediated Green Intention for the Treatment of Biomedical Waste Obtained From Salem City, Tamil Nadu

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Abstract: ***The living systems of world population is facing lot of health care issues with the unpredictable diseases. The modern trend would also lead to the various biomedical infectious which is highly resistant to the available drugs. The proper disposable and management of infectious and hazardous biomedical waste is needed. The research in the field of medical industry would address the issues of disposable and management of infections of Biomedical waste. At this juncture, this paper attempts to describe an alternative novelapproach for handling and management of biomedical waste. Plant species mediated biomedical waste management will be the key concept to minimize the treatment strategy of Biomedical Waste. Biomedical wastes comprises of category 3 & 6 were collected from the hospital and subjected to treatment with plant species like Ocimum tenuiflorum, Curcuma longa, Seaweeds like Padina tetrastomatica, Steochospermum marginatum, home-made Panchakavya and mixed plant consortium. The preliminary treatment was done for about 48-72hr with pH of 8.5. Then the treated sample was subjected to alkaline hydrolysis with the presence of ethanolic extract of Eichhornia crassipes at 45ºC under the pressure of 1 bar. The result indicated that the plant Curcuma longa showed significant reduction in microbial count, Alkalinity, Electrical conductivity, TDS, BOD, COD and pH followed by thulasi in the preliminary treatment whereas seaweed extract showed less number of reduction in microbial populations. Panchakavya revealed the considerable reduction in alkalinity as well as electrical conductivity but in case of microbial count has been increased due to the resistivity. The overall result concluded that plant mediated green approach will be the next level of management of biomedical waste.***

Index terms: TDS, Alkalinity, Panchakavya, Seaweed.

I. INTRODUCTION

There is an emergency in the trend of biomedical waste management which includes the generation, accumulation, storage and disposal. These wastes were generated from healthcare facilities which not only includes hospitals but also clinics, doctor's offices, Veterinary hospitals and Clinical laboratories. The improper management of these wastes may results in serious impact among healthcare workers and on environment .This in turn makes the researchers to make a high note on biomedical waste disposal. In 1999, a report by World Health organization (WHO) stated that about 85% of these wastes are meant to be hazardous and the remaining 15% were non-hazardous. Overall, the range of infectious wastes were 15 to 35% percentage of total waste generated . The improper and unscientific implementation of biomedical waste would resul in very serious diseases to patients and also to healthcare workers in wide range (CEET, 2008). Although, there are wide range of disposal methods most of them are harmful. This changed the current scenario of waste management systems in health care facilities to prompts the administration to shift towards the new and innovative scientific ideas with safe and cost effective because waste management is considered to be the important component of quality assurance in health care systems. As per the defined rules for biomedical waste management, 1998 the wastes were categorised under ten categories such as Human anatomical waste, animal waste, microbiological and biotechnology waste, waste sharps, discarded medicine and cytotoxic drugs, soiled waste, solid waste, Liquid waste, Incineration and chemical wastes. Further some revisions have been made in this regulations in the year of 2016. According to that amends, states pollution control board is responsible for implementing revised legislations. In recent years, world is moving towards the green environment where plants are considered to be the promising source. This in turn makes the researchers to have a belief that implementation of plant in the waste management system may provide cheap, safe and efficient system rather than conventional system of incineration and autoclaving[1].

The present study aims at investigating a novel strategy of using plants as a target material to use in biomedical waste management system. Plant species mediated biomedical waste management would be the key idea of this current research work with cost effective strategy when compared to conventional method. The biomedical wastes of category 3 and 6 were collected from hospitals around Salem region and were treated with plant species like *Ocimum tenuiflorum*, *Curcuma longa* and seaweeds like *Padina tetrastomatica*, *Steochosperum marginatum* home-made panchakavya and plant consortium.

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The microbial count, alkalinity, electrical conductivity, TDS, BOD, COD and pH were analysed to study the significance of plant species in the waste management system[2].

The seaweeds have good antimicrobial activity. *Ocimum tenuiflorum* used in this study were commonly known as tulasi which can be valued as elixir of life. The researchers had found that this plant may originate from north central India . Dried leaves of this plant when mixed with stored grains can be used as insect repellents (Biswas and Biswas, 2005). The chemical constituents present in the plant are rosmarinic acid, carvacrol, ursolic acid, oleanolic acid, β-caryophyllene, eugenol, linalool, germacrene D and β-elemene that can have good antioxidant, antimicrobial and radical scavenging activity. A study reveals that the extracts of tulasi have good antibacterial activity against *E. coli, S. aureus* and *P. aeruginosa*. *Curcuma longa* commonly named as Turmeric, another plant species used in this studies has a large oval rhizome with sessile cylindrical tubers containing curcumin content. They are commonly used as a flavouring agent, preservatives and colouring agent including biomedical applications. In India, Tamil Nadu is the largest producer of turmeric. The rhizome isolates of turmeric possess a wide range of biological activities like antibacterial activity, anti-inflammatory , anticancerous , and wound healing[3],[4]

II. MATERIALS AND METHODS

All the chemicals and Glasswares were purchased from Himedia and Chemico, Erode. The biomedical waste was required from the hospital with proper pack. Pathogenic strains were obtained from National Centre for Industrial Microorganism, Pune for comparison purpose. Plant species such as *Ocimum tenuiflorum, Curcuma longa,* Seaweeds like *Padina tetrastomatica, Steochospermum marginatum*, home-made Panchakavya and mixed plant consortium were required for the present study.

a. Sampling Site

The biomedical waste (Category 3 & 6) was collected from various hospitals at Salem district, Tamil Nadu, with prior concern. The collected waste was segregated with the based on the different category as per Biomedical Waste Management. The two different plant samples such as *Curcuma Longa, Eichhornia* were collected from in and around Salem districts of Tamil Nadu. Two seaweeds such as *Padina tetrastromatica* and *Steochospermum marginatum* were collected from Rameshwaram coastal areas of Tamil Nadu. The plant samples were collected and authenticated by Botanical Survey of India, Coimbatore, for the present study. The *Eichorrnia* Species were obtained from the Cauvery River, Erode.

Fig. 1: Collection of plant sample used in the present study

b. Preparation of powdered extract

Collected plant specimens were shade dried without any moisture. A powder of 150g was taken into 500mL of methanol using soxhalet apparatus under room temperature for three days. The solution were filtered through the filter paper. The colourless solution in the supernatant which indicates the complete extraction process. Then concentrate of bioactive compounds from plant species were obtained by using rotatory evaporator ..

c. Preliminary Treatment schedule for Biomedical Waste

Biomedical waste was segregated based on the colour code. A weight of around 1 Kg was taken and 10g of crude methanolic plant extract was added on it. The treatment process was carried out for 2-3 days with constant stirring speed (Uday *et al.*, 2004)

Control - without any treatment

Group 1 – Treated with methanolic extract of *Ocimum tenuiflorum* (10g)

Group 2 – Treated with methanoloic extract of *Curcuma longa* (1
0g)

Group 3 – Treated with methanolic extract of *Padina tetratomatica* (10g)

Group 4 – Treated with Methanolic extract of *Stechospermum marginatum* (10g)

Group 5 – Treated with Panchakavya (100mL)

Group 6 – Treated with mixed plant consortia (10g)

d. Secondary treatment of alkaline digestion using Eichhornia crassipes

The preliminary treated biomedical waste was further subjected to alkaline digestion process using potassium hydroxide (10%) with the presence of *Eichhornia crassipes* (5g) (Anne *et al.*, 2006). The system was operated at room temperature for 6-8hours with the pressure of 1bar. Then the digested fraction was monitored and analysed for the various chemical and biological parameters.

e. Quantitative analysis of Treated and untreated biomedical waste

The sample was taken from both untreated and treated from preliminary and secondary treatment.

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The sample has been subjected to measure the bacterial count using spread plate techniques with the sample volume of 0.1mL. It subjected to analyse the alkalinity, pH, TDS, BOD and COD level in both treated and untreated sample. The identification was done based on the morphology and appearance in microscope (Baveja *et al.*, 2000).

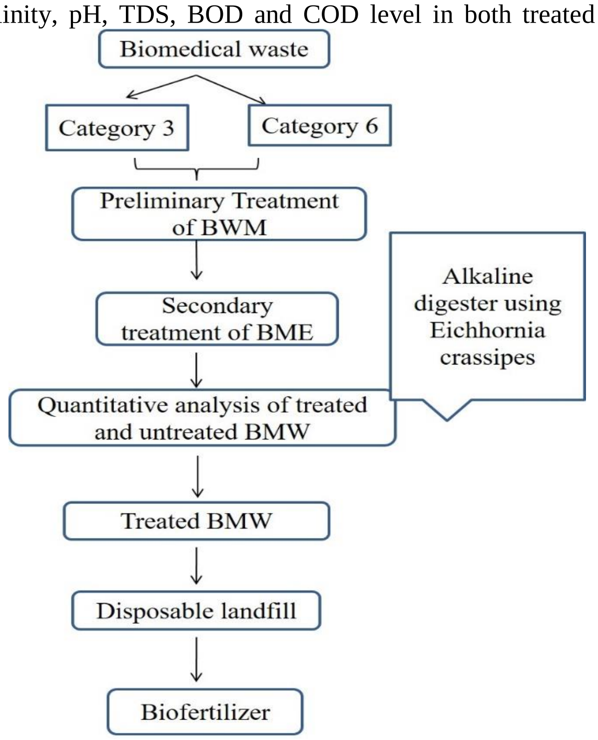

Fig. 2: Overall flow diagram of Biomedical waste Management

i) Toxicity test

Toxicity test was studied for the final treated waste by using Fish toxicity test (Zebra fish). Zebra fish was collected and maintained at laboratory for 15 days as acclimatization process. Fish tank was circulated with different concentration of biomedical treated waste (1%, 2%, 3%, 4% and 5%) for the assessment of growth. The mortality rate was measured for individual group with respect to the viability.

III. RESULTS AND DISCUSSION

The study area has been selected based on the number of hospital, surgical operations, number of patients and waste generation as per the data of Approved nursing homes by Government of Tamil Nadu (Table 1). The study indicated that Chennai holds the highest number of BMW (5680.08 Kg/day) followed by Coimbatore of having 1883.7Kg/day. The Salem city showed the generation of BMW was around 753.48 Kg/day. The city has conventional BMW facility with the provision of incinerator. Thus, the alternative approach of green mediated treatment facility is necessary for both cost wise and safety of environment. Wayne (2003) demonstrated that survival probability of bacillus spores in the generated waste was very high. The report indicated that thermophilic spores were present in the highest survival probability. As per the WHO (2004), 85% of hospital wastes were categorized as non-hazardous, 10% are infectious substances and remaining 5% are consider as non infectious but it is hazardous. Accordingly our result were considered as the similar category of BMW.

TABLE 1. GENERATION OF BIOMEDICAL WASTE PER HOSPITAL AT CORPORATION LEVEL

S. No.	Name of the corporation	No. of Hospitals	No. of beds	Total Quantity of BMW generated (Kg/ day) (Approximately)	Total Quantity of BMW generated per hospital (Kg/ day/Hospital) (Approximately)
1	Chennai	139	19600	5680.08	40.864
2	Coimbatore	148	6500	1883.7	12.728
3	Salem	72	2600	753.48	10.465
4	Madurai	54	3875	1122.975	20.796
5	Trichy	37	2800	811.44	21.931
6	Tirunelveli	41	2000	579.6	14.137

TABLE 2: CHARACTERISTIC OF BIOMEDICAL WASTE OBTAINED FROM SALEM DISTRICT, TAMIL NADU

S.No.	Parameters	Value
1	Moisture content (%)	8-25
2	Total solids (mg1^{-1})	20000-60000
3	Volatile solids (mg1^{-1})	10000-70000
4	COD (mg1^{-1})	3000-14000
5	Alkalinity (mg1^{-1})	1000-6000

The collected BMW waste was observed in the range of 8-25% moisture level. Total dissolved solids were present in the average of 40,000 mg1^{-1}. It has the highest solid content in it which has to be treated. The chemical oxygen demand and alkalinity also showed highest level of toxic substances at Salem BMW waste. The appropriate reduction **is** the range of normal level.

a. Total microbial count of treated BMW by standard plate technique

Fig. 3: Collection and segregation site of various biomedical waste

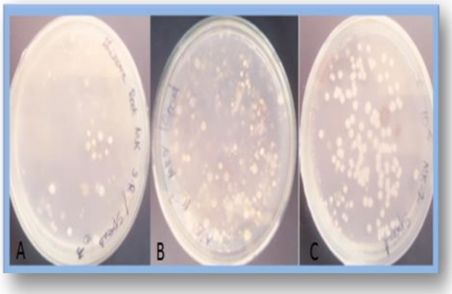

Fig. 4: Bacterial count of biomedical waste using standard plate technique
A. Curcuma longa B. Ocimum tenuiflorum C. Padina tetratoma

TABLE 3. REDUCTION OF PATHOGEN COUNT IN PRELIMINARY TREATMENT OF BMW

S. No.	**Time (hr)**	***Curcuma longa* Treated**	***Ocimum tenuiflorum,***	***Padina tetrastomatica***	***Steochosperum marginatum***
1	0	2.3×10^7	1.3×10^8	2.3×10^7	1.5×10^6
2	5	1.8×10^5	1.9×10^7	1.7×10^6	1.3×10^6
3	10	2.6×10^4	2.5×10^5	1.3×10^5	0.9×10^5
4	15	12000	19×10^4	2.3×10^4	70000
5	20	9000	16000	18000	66000
6	25	8000	14000	12000	60000
7	30	5000	9000	9000	55700
8	35	3267	7467	7000	50000
9	40	1231	4888	5000	45000
10	45	780	2000	2780	34000
11	50	450	1038	1021	30000
12	55	210	800	678	20000

The reduction percentage of pathogenic microbes was studied with respect to different time interval. *Curcuma longa* showed highest inhibitory activity against the bacterial population at the end of the 55th hour as 210 colonies of bacteria followed by *Padina tetrastomatica* revealed around 678 colonies of pathogens. The *Ocimum tenuiflorum* exhibited a tune of 800 colonies at 55th hour whereas *Steochosperum marginatum* indicated less level of reduction in microbial pathogens. Kanemitsu *et al.* (2005) confirmed that the killing of bacillus spores required incineration process of having above 300°C. The similar level of death rate was observed in the present technology by turmeric species.

b. *Secondary treatment of BMW by Alkaline digestion process*

The secondary treatment revealed 10% *Eichornia* species with high alkalinity conditions using NaOH. The optimization process of pH was done which revealed increase in pH increase in microbial death rate. The exposure time was optimized as 3-4hour with the pH range of above 8.0. The result indicated that there was a significant reduction in microbial level in final treatment. The 100% mortality of pathogenic microbes were observed in the BMW treated with *Curcuma longa* followed by *Padina tetrastomatica.* The complete reduction without any toxic microbial load which is further used for agricultural purpose. The result of present study is on par with of the result of Edinburg City council (2002), that to break / disintegrate the bone, tissue require high temperature with high pressure alkaline hydrolysis around 3 hours for treating sheep waste.

c. *Assessment of Bioassay of treated BMW by Zebra fish model*

The safety aspects of treated BMW was studied using live zebra fish model to monitor the mortality at *in vivo* conditions by both primary and secondary effluent. The result of primary treated BMW showed highest mortality rate while increase in the concentration. The percentage viability of fish model was calculated nearly 90% at 1% of treated BMW. The secondary treated BMW was having highest number of viability more than 98%. The result

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suggested that secondary treated waste can be one of the safest and can be easily disposed in landfill or can be used for agricultural plant growth promoters.

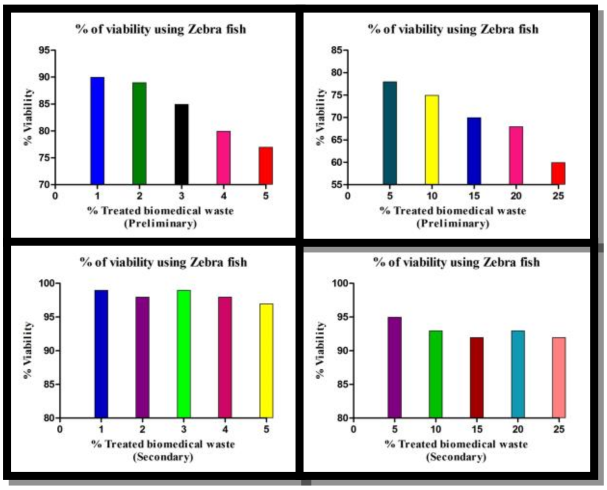

Fig. 5: Bioassay based assessment of treated BMW using Zebra fish model

IV. CONCLUSION

The biomedical waste is considered as a biggest problem in the world. There are numerous technology was adapted to reduce the biological toxin present in the biomedical waste. In developing countries like India follows the major process of Incineration in order to reduce the complete toxic level. But in major developed countries, tissue digestion process was more crucial to treat the BMW. In this context, the present study is to utilize the plant species as a green resources in reducing the microbial count. The turmeric (*Curcuma longa*), thulasi (*Ocimum tenuiflorum*) and seaweeds were used. The result indicated that turmeric variety showed highest level of reduction in microbial load in category 3 & 6. Thus, plant species mediated treatment might be fruitful technology for the effective management of BMW. The Alkaline digestion process are also much important in reducing the biological waste especially pathogenic bacteria, fungi and yeast. This technology of BMW is an effective tool for reducing toxic and microbial load. Further, residual effluent can be used for sustainable nutrition in agricultural practices in India because of enriched major and minor elements in bone, tissue, blood and organs.

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