

EVALUATION OF METAL NUTRIENT FROM SEWAGE SLUDGE ON
GROWTH OF MEDICINAL HERBS

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*To my beloved family members who show 100% support for everything that I have
done for the completion of this project.*

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ABSTRACT

Generally municipal sewage sludge can be used as fertiliser as it contains a lot of nutrients. However the level of each particular nutrient has not yet been established locally. By focusing on copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) contents in municipal sewage sludge from primary oxidation pond of Taman Sri Pulai, Johor and in plant tissue sections, this study attempts to explain the relationship between plant yield and metal concentrations and suggest alternative evaluation for future in-depth studies. Raw sludge samples were collected, then air dried and ground to powder form in the beginning of a field work. The metal concentrations of sewage sludge were copper 6.9 mg/kg; iron 330.2 mg/kg; manganese 6.7 mg/kg; and zinc 9.1 mg/kg. *Curcuma longa* (turmeric) and *Orthosiphon stamineus* (cat's whiskers) were selected and the plants received different quantity of sludge application ranging from 1 g per plant to 4 g per plant weekly for six consecutive weeks while observations were done for eleven weeks inclusive of the first six weeks of sludge application and five weeks of post application period. The field experiments spanning 12 rows of vegetation and 65 pH grid points were carried out on iron rich soil. Physical parameters such as leaf length, plant height, rhizome yield, shrub width and shrub height were monitored parameters used to determine plant growth together with the chemical contents of both types of plants. All samples were acid digested using EPA Method 3050B and analysed using HACH DR5000 spectrophotometer. Soil pH readings during and after sewage sludge surface application at plant base section throughout the entire experiment did not display any statistically significant changes using one way ANOVA at $\alpha=0.05$. Turmeric displayed 99% confidence level of negative correlation in the iron-manganese interaction in the plant using one way ANOVA at $\alpha=0.01$ while cat's whiskers displayed no definite pattern of metal relationships within the shrubs. Metal ratios between copper, iron, manganese and zinc were analysed to determine the occurrence of potential metal induced stress. Cu:Mn, Fe:Mn, Cu:Zn, Fe:Zn and Mn:Zn ratios suggested that turmeric suffered from manganese and zinc deficiency under high iron environment while Fe:Mn, Cu:Mn and Mn:Zn ratios suggested that cat's whiskers suffered from manganese deficiency only. Through this study, the determined optimum sewage sludge dosage for turmeric was 3 g per plant while the optimum dosage for cat's whiskers was between 2 to 3 g per plant.

ABSTRAK

Secara amnya, enapcemar kumbahan boleh digunakan sebagai baja kerana ia mengandungi banyak bahan nutrien. Namun kandungan setiap nutrien masih belum dikenalpasti secara terperinci. Dengan memberi fokus kepada kandungan kuprum (Cu), ferum (Fe), mangan (Mn) dan zink (Zn) dalam enapcemar kumbahan dari Taman Sri Pulai, Johor dan juga dalam komponen-komponen tisu tumbuhan, kajian ini cuba mengaitkan hubungan antara tumbesaran tumbuhan dengan kandungan logam dan mencadangkan kaedah alternatif untuk kajian yang selanjutnya. Sampel enapcemar dikutip secara mentah, kemudian dikeringkan secara udara dan dikisar menjadi serbuk halus sebelum digunakan dalam kajian. Kandungan logam dalam enapcemar adalah seperti berikut: kuprum 6.9 mg/kg; ferum 330.2 mg/kg; mangan 6.7 mg/kg dan zink 9.1 mg/kg. *Curcuma longa* yang dikenali sebagai pokok kunyit dan *Orthosiphon stamineus* yang dikenali sebagai pokok misai kucing, telah dipilih sebagai subjek dalam kajian ini diberi dos aplikasi enapcemar kumbahan yang berlainan bermula daripada 1 g per tumbuhan kepada 4 g per tumbuhan untuk enam minggu yang pertama berturut-turut manakala pemerhatian dilakukan sepanjang sebelas minggu, termasuk enam minggu dengan aplikasi enapcemar dan lima minggu lagi yang tidak diberi sebarang nutrien. Eksperimen yang merangkumi 12 batas dan 65 titik grid pH ini dijalankan pada tanah yang mengandungi kandungan ferum yang tinggi. Tumbesaran diukur dalam parameter seperti kepanjangan daun, ketinggian tumbuhan, berat rizom, kelebaran dan ketinggian semak selain mendapatkan kandungan kimia dalam tumbuhan. Semua sampel dalam kajian ini dicerna menggunakan asid mengikut kaedah EPA 3050B sebelum dianalisis dengan menggunakan spektrofotometer HACH DR5000. Dalam eksperimen ini, nilai pH tanah didapati tidak berubah secara ketara semasa dan selepas tempoh aplikasi enapcemar pada permukaan tanah di sekeliling batang tumbuhan melalui ANOVA pada $\alpha=0.05$. Interaksi ferum-mangan dalam kunyit didapati menepati 99% korelasi negatif melalui ANOVA pada $\alpha=0.01$ manakala tiada terdapat corak yang tetap dalam interaksi logam pada misai kucing. Nisbah logam antara kuprum, ferum, mangan dan zink dianalisis untuk mengesan tekanan yang disebabkan oleh logam-logam tertentu pada tumbuhan. Nisbah-nisbah Cu:Mn, Fe:Mn, Cu:Zn, Fe:Zn dan Mn:Zn mencadangkan bahawa kunyit mengalami kekurangan mangan dan zink dalam keadaan persekitaran yang kaya dengan ferum manakala nisbah-nisbah Fe:Mn, Cu:Mn dan Mn:Zn mencadangkan bahawa misai kucing mengalami kekurangan mangan sahaja. Secara keseluruhan, kandungan optimum enapcemar bagi kunyit adalah 3 g per tumbuhan manakala kandungan optimum enapcemar bagi misai kucing adalah antara 2 hingga 3 g per tumbuhan.

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LIST OF SYMBOLS AND ABBREVIATIONS

Ag	-	Silver
ANOVA	-	Analysis of Variance
As	-	Arsenic
B	-	Boron
BOD	-	Biochemical Oxygen Demand
C	-	Carbon
Ca	-	Calcium
CaCl ₂	-	Calcium chloride
CaCO ₃	-	Calcium Carbonate
Cd	-	Cadmium
CDM	-	Chemically Defined Medium
Cl	-	Chlorine
cm	-	Centimeter
Co	-	Cobalt
CO ₂	-	Carbon Dioxide
CO(NH ₂) ₂	-	Urea
Cr	-	Chromium
Cu	-	Copper
COD	-	Chemical Oxygen Demand
DDT	-	Dichloro-Diphenyl-Trichloroethane
DO	-	Dissolved Oxygen
EPA	-	Environmental Protection Agency
EPB	-	Environmental Protection Branch
Fe	-	Iron
ffu	-	Focus Forming Units
g	-	Gram
H	-	Hydrogen

H ₂ O	-	Water
H ₂ O ₂	-	Hydrogen Peroxide
H ₂ PO ₄ ⁻	-	Dihydrogen Phosphate
H ₂ S	-	Hydrogen Sulphide
HAc	-	Hydrogen Acetate
Hg	-	Mercury
HNO ₃	-	Nitric Acid
HPO ₄ ²⁻	-	Hydrogen Phosphate
K	-	Potassium
K ₂ O	-	Potassium Oxide
kg	-	Kilogram
lb	-	Pound
LSD	-	Least Significant Difference
m	-	Meter
m ³	-	Meter Cube
Mg	-	Magnesium
mg/kg	-	Miligram per Kilogram
mg/L	-	Miligram per Liter
ml	-	Mililiter
Mn	-	Manganese
Mo	-	Molybdenum
N	-	Nitrogen
Na	-	Sodium
NA	-	Nutrient Agar
NADH/NAD ⁺	-	Nicotinamide Adenine Dinucleotide
NADPH/NADP ⁺	-	Nicotinamide Adenine Dinucleotide Phosphate
ng/kg	-	Nanogram per Kilogram
N:P	-	Nitrogen to Phosphorus
N:S	-	Nitrogen to Sulphur
NF	-	Nitrogen Fixing
NH ₃	-	Ammonia
NH ₄ ⁺	-	Ammonium
NH ₄ -N	-	Ammonia Nitrogen
Ni	-	Nickel

NO_2^-	-	Nitrite
NO_3^-	-	Nitrate
$\text{NO}_3\text{-N}$	-	Nitrate Nitrogen
NPK	-	Nitrogen, Phosphorus and Potassium
NPKS	-	Nitrogen, Phosphorus, Potassium and Sulphur
O	-	Oxygen
P	-	Phosphorus
P_2O_5	-	Phosphorus Pentoxide
PAH	-	Polycyclic Aromatic Hydrocarbons
Pb	-	Lead
PCB	-	Polychlorinated Biphenyls
PCDD	-	Polychlorinated Dibenzodioxins
PCDF	-	Polychlorinated Dibenzofurans
PE	-	Population equivalent
PFU	-	Plague Forming Units
PO_4^{2-}	-	Orthophosphate
ppm	-	Parts per Million
PS	-	Phosphate Solubilising
S	-	Sulphur
Sb	-	Antimony
Se	-	Selenium
SiO_2	-	Silica
Sn	-	Tin
SO_2	-	Sulphur Dioxide
SO_4^{2-}	-	Sulphate
TCID_{50}	-	Tissue Culture Infection Dose for 50% response
TS	-	Total Dry Solids
UV	-	Ultra-Violet
V	-	Vanadium
v/v	-	Volume / Volume
wt	-	Weight Percent
Zn	-	Zinc
%	-	Percent
$^{\circ}\text{C}$	-	Degree Celsius

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CHAPTER 1

INTRODUCTION

1.1 Historical Overview

Historically, human waste disposal became problematic when communities first formed. The earliest covered sewers uncovered by archaeologists were in regularly planned cities of the Indus Valley Civilization while the Cloaca Maxima found in ancient Rome disgorged into the Tiber. Since population densities were still low enough at that time, human waste was handled by surrounding land or waterways as the carrying and absorption capacities were fairly high. As populations grew, the nearby land could not handle all the wastes, so waste was dumped into streams and rivers. In medieval European cities, small natural waterways used for carrying off wastewater were eventually covered over and functioned as sewers (Reid, 1993).

Chinese society reused sewage, called “night soil,” on surrounding farmland for thousands of years. This practice helped maintain soil fertility by closing the nutrient cycle. Nutrients from farms were exported to the cities in crops, and the nutrients in the municipal wastes were returned to the farms. This type of system was ideal because two problems were solved at once: maintaining soil fertility and treating a source of pollution. Chinese society was unique in its development of an ecologically sound system for recycling waste where most other early urban civilizations focused on improving ways to dispose of wastes from cities. The wastewater was discharged without any treatment, and receiving watercourses

became heavily polluted, often causing severe pollution and mass contraction of diseases (Frost, 2001).

Sewage management saw certain level of progress when large-scale cropland application of municipal wastewater was first practiced about 150 years ago after flush toilets and sewer systems were introduced into cities in Western Europe and North America. In 1869, the practice was implemented in Berlin, Germany, which bought large areas of cropland to be irrigated with raw sludge. The city of Paris, France also purchased farmland to be irrigated with sewage. In 1897, Melbourne, Australia went to land treatment of raw wastewater at Weribee. The concept eventually gained wide acceptance throughout the region both in Europe and the United States (Cecil *et al.*, 1998).

1.2 Current Practices

The main components of sewage sludge are human waste, food waste and animal waste but throughout the years the matrix become more and more complex as various toxic chemicals were discharged intentionally and unintentionally into the sewers. The systematic application of treated or untreated sewage sludge remains a problem to the global community though direct application of these wastes onto crops were still widely practiced by world wide suburban populations, There were several risks involved, including the risk from contaminated food by *E. coli*, which has resulted in numerous deaths and contamination of food by *Salmonella sp.* and other bacteria; the risk from contaminated water, which has resulted in many persons being ill, as well as numerous deaths; and the risk of ingesting fish contaminated with heavy metal such as mercury, as compared with ingesting heavy metal from bio-solids-contaminated food. Nevertheless over the years, land application of sewage sludge has been increasingly managed to protect human health and the environment from harmful micro-organisms, heavy metals and toxic organic chemicals (Environment Protection Agency, 1997).

Although there is still a lot of debate regarding the safety of treated sludge application on land. Discontinuation of marine disposal has put more pressure on land-base method of utilisation and disposal. Epstein (2003) stated that in 1997 alone the United States oversaw a 54% of sludge and biosolids in land application and distribution while in 1999 on average in Europe, 36.4% of the biosolids was used in agriculture. Many relevant publications have already stated some beneficial effects of the application of composted sewage sludge from wastewater to several kinds of soils, including agricultural soils, under different weather conditions and using compost from different sources highly variable in nutritional composition (Johannesson, 1999; Fernandes *et al.*, 2005). A review of several literature commonly available showed previous experiments using other economically relevant species as experimental models, such as barley (Epstein, 2001), lettuce (Frost, 2001), petunia (L'hermite, 1991), corn (Warman and Termeer, 2004; Chen *et al.*, 2007), sweet pepper (Casado-Vela *et al.*, 2007) and wheat (Sutapa and Bhattacharyya, 2008; Chandra *et al.*, 2009).

Recent development in China saw a rise of research in sewage sludge as fertiliser even though such practices had existed for thousands of years. China having a huge population and landmass was looking into the potential benefits and ways to minimise the environmental impact of sewage sludge land application.

In Malaysia, several researches regarding the potential of sewage sludge had been carried out under the banner of Indah Water Konsortium Sdn. Bhd. The research projects can be found on Indah Water Konsortium website. Over the years sewage sludge were being used for commercial plantation and urban landscape plants but the use of sewage sludge on agriculture especially daily consumption plants were still restricted due to poor understanding and manipulation techniques of local authorities and farm operators. To date there was limited guideline to regulate the land application of sewage sludge on agriculture sector.

1.3 Problem Statement

Domestic wastewater is treated separately from industrial wastewater in Malaysia, which in turn generates safer sewage sludge in the sense of environmental and human health aspects. As of December 2008, Indah Water Konsortium (2008) reported that there are a total of 9,525 sewage treatment plants serving a population equivalent (PE) of 21,472,975 in Malaysia. Unfortunately the treated sludge is disposed either at landfills or incinerators. Many countries such as the United States, United Kingdom, Australia and Singapore are practicing land application of sludge due to its economical and environmental advantages. However in Malaysia, according to Standards & Industrial Research Institute of Malaysia (1991), land application of sludge is not allowed for land which is used for growing food crops to be eaten raw. Although it is a fact that sewage sludge could be recycled as soil conditioner and fertiliser (Indah Water Konsortium, 2006), much of the interaction of organic and inorganic compounds between sewage sludge, soil, plant and microbial activity remain ambiguous. Furthermore, different plants have different level of tolerance in terms environmental stress due to sludge nutrient loadings.

Under the 9th Malaysia Plan 2006-2010 published by the Economic Planning Unit (2006), the agriculture sector involving large-scale commercial farming, the wider application of modern technology, production of high quality and value-added products are revitalised to become the third engine of growth so there will be a steady rise on the demand of fertilizer. Utilising the high potential of nutrient content in treated sewage sludge would provide significant cost reduction in fertilizer acquisition while minimising overall disposal of solid waste to landfills and incinerators. Therefore it is crucial to identify the optimum sludge dosage with reference to specific plant types by studying the nutrient availability, metal transport and soil-sludge interaction which might arise as a result of land application of sludge in any economic scale to maximize the potential usage of sludge while minimising the environmental impact caused by it thus ultimately benefit the country's agro-based industries and economy.

1.4 Objective

The objectives of this study were:

- i) To determine metal nutrient availability in sewage sludge to be applied on plants at different rates.
- ii) To study the plant growth of two types of important medicinal herb on subsurface and surface plant sections respectively under sewage sludge application.
- iii) To determine the influence of sewage sludge on soil pH and plant metal distribution.

1.5 Scope of Study

The primary concern of the study was to determine the content of micronutrients namely copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) which were used by the plants as catalytic agents in the metabolic processes concerned with growth and synthesis (Sauchelli, 1969; Prasad, 2004; Sharma, 2006).

Sludge samples were taken from the primary oxidation pond at Taman Sri Pulai, Skudai, Johor. Samples were air dried, ground to fine powder form and analysed at the Environmental Laboratory of Faculty of Civil Engineering in UTM. Sludge powder was then applied onto the plants weekly starting from week 1 to week 6.

Two types of medicinal herbs with different edible parts, *Curcuma longa* (turmeric) with the subsurface edible part of rhizome and *Orthosiphon stamineus* (cat's whiskers) with the surface edible part of leaf were grown as part of the experiment in measuring the efficiency of sewage sludge.

The experimental plot was a piece of open land with fairly high concentration of iron ranging between 300 mg/kg and 400 mg/kg, and initial pH values ranging between 4.6 and 5.8 with an average of 5.1. Atmospheric conditions were assumed to have uniform effects throughout the plot therefore would not be taken into consideration during result analysis.

1.6 Significance of Study

This study would promote a better understanding of the sludge-plant behaviour in the context of topical sludge application. Armed with the research findings, systematic and effective utilisation of sewage sludge in large scale commercial farming would be more sustainable, cost saving mainly due to the usage reduction of synthetic fertiliser and achieve higher yield through the implementation of precision farming or sustainable farming management, in which application of sludge as fertiliser would only take place based on individual nutrient requirement scenarios. Effective utilisation of large amount of sludge would also translate into minimisation of landfill, incinerator usages and ultimately the reduction of overall environmental impact, besides enhancing the national agro-based industries and economy.

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