



Faculty of Resource Science and Technology

**BACTERIAL INDICATOR CONCENTRATIONS IN A LOCAL ECOLOGICAL  
SANITATION ( EcoSan) SYSTEM**

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**BACTERIAL INDICATOR CONCENTRATIONS IN A LOCAL ECOLOGICAL  
SANITATION (EcoSan) SYSTEM**

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This project is submitted in partial fulfillment of the requirements for the degree of  
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## IV

### LIST OF ABBREVIATIONS

<b>EcoSan</b>	Ecological Sanitation
<b>FCC</b>	Faecal Coliform Count
<b>GTF</b>	Greywater Treatment Facilities
<b>IMViC</b>	Indole, Methy Red, Voges-Proskauer, Citrate
<b>TBC</b>	Total Bacterial Count
<b>TCC</b>	Total Coliform Count

# **Bacterial indicator concentrations in a local Ecological Sanitation(EcoSan) system**

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## **ABSTRACT**

Greywater Treatment Facilities (GTF) located at Kenanga College, Universiti Malaysia Sarawak, is based upon the Ecological Sanitation (EcoSan) system where it recognizes domestic wastewater not as waste but as a resource that can be treated when necessary to be safely used again in many purposes. The present study is a study on the efficiency of the GTF to reduce the level of microorganisms. Bacteriological parameters such as total coliform, total bacterial, heterotrophic bacterial and faecal coliform counts as well as a series of biochemical tests namely IMViC test and Kligler Iron Agar test were carried out in this study. It was found that the GTF's efficiency varied depending on the treatment stage. Based on the bacterial indicator concentrations, the oil and grease trap at the early stage of the treatment before heading to the reservoir demonstrated efficiency within the range of 22% to 100% in reducing the microbial population compared to the biofilter tanks utilizing different filter media which has shown increment in the bacterial indicator concentrations within the range of 3.7% to 97% for both outlets. There are several contributing factors affecting the efficiency such as structure and maintenance of the system and nutrient availability. Further research can be carried out in order to aid in the improvement of the established treatment system

Key words: Ecological sanitation, greywater

## **ABSTRAK**

*Fasiliti rawatan air kumbahan domestik yang terletak di Kolej Kenanga, Universiti Malaysia Sarawak, merupakan fasiliti yang berdasarkan prinsip Ecological Sanitation (EcoSan) . Prinsip ini mengenali air kumbahan domestik bukan sebagai bahan buangan tetapi sesuatu yang boleh diguna semula untuk pelbagai tujuan sekiranya diberi rawatan yang sepatutnya. Kajian ini dilakukan untuk melihat keberkesanan fasiliti rawatan tersebut dalam mengurangkan populasi mikroorganisma. Parameter bakteriologi seperti kiraan jumlah koliform, jumlah bakteria, bakteria heterotrofik dan koliform najis bersertakan dengan ujian biokimia seperti ujian IMViC dan Agar Kligler telah dilakukan dalam kajian ini. Berdasarkan keputusan yang diperolehi, fasiliti ini mempunyai tahap keberkesanan yang berbeza mengikut fasa rawatan. Perangkap minyak dan gris pada awal peringkat sebelum menuju ke reservoir telah menunjukkan pengurangan mikroorganisma yang ketara dengan julat 22% hingga 100% jika dibandingkan dengan tangki biopenapis yang menggunakan media penapis yang berbeza telah menunjukkan peningkatan populasi mikroorganisma dengan julat 3.7% hingga 97% untuk kedua-dua outlet. Faktor yang boleh mempengaruhi keberkesanan fasiliti ini adalah seperti struktur dan penyelenggaraan sistem dan kehadiran nutrien. Kajian lanjutan boleh dilakukan untuk meningkatkan tahap keberkesanan sistem yang sedia ada.*

*Kata Kunci: Ecological Sanitation (EcoSan), Air kumbahan domestik*

## 1.0 INTRODUCTION

The rise in human population will definitely lead to the increase in the consumption of water and use of household chemicals. With the increase number of houses per surface area, this limit the space for proper managing of domestic water and consequently leading to widespread community health problems, environmental problems and restraining economic and agricultural growth (Okoh *et al.*, 2007). This calls out for the need of appropriate measures to be taken to treat domestic water which is also known as greywater, to ensure the discharges of safe quality greywater effluents into receiving bodies that promises a healthier environment. The solution to this matter can be seen from the notion behind “ecological sanitation” or better known as EcoSan. This is where sanitation problems could be solved more feasibly and efficiently based on recycling principles. The resources contained in excreta and wastewater is treated when necessary, to be recovered and safely used again.

With that, the existing Greywater Treatment Facilities (GTF) are of great importance. The GTF at Kenanga College, Universiti Malaysia Sarawak is based upon the EcoSan system which aims to prevent environmental contamination by using appropriate techniques for purifying water with the earnest attempt to reuse or return it to nature in an accountable way. The choice of greywater management system depends on a few factors such as type of habitation, land-use patterns, existing drainage system, degree of pollution, social and cultural (Ridderstolpe, 2004). Greywater has to be managed carefully to avoid waterlogging, foul smell, and the uncontrolled release of chemicals and anthropogenic elements including microorganisms into the environment. The Greywater Treatment Facilities at Kenanga College involve a few steps before the water is

being discharged into a retention pond and lastly to the external drain heading to Samarahan river.

Firstly, the greywater will pass through the oil and grease trap where all large particles, fibres, grease and oil will be removed to prevent clogging of the pipe system. Then, the water will be discharged into a holding tank where it is constructed for gravimetric separation of particles from the water. The next target is to remove the biodegradable compounds and to reduce the levels of microorganisms with the help of the biofilter tanks. The biofilter constructed uses four different types of filter media namely granulated activated carbon, crushed red bricks, coarse sand and light weight aggregate which are capable of improving water quality.

The greywater treatment system has to be monitored to ensure a constant performance and that maintenance can be carried out if the system is found to be not functioning. Monitoring steps often involve a large number of water quality parameters such as Chemical Oxygen Demand (COD), Total Organic Carbon (TOC), Suspended Solids (SS), Dissolve Oxygen (DO), Biological Oxygen Demand (BOD), pH, specific conductance and temperature (Plummer & Long, 2007; Okoh *et al.*, 2007). In addition, indicator organisms such as faecal and total coliform are often used to measure microbiological status of water.

In spite of this, the problem centers on the ability of the system to reduce the microbiological contaminants. The efficient elimination of pathogens from greywater treatment is a crucial task and varies according to the treatment process type, pH, retention time, oxygen concentration, and temperature. Typically, an optimal microbial reduction of 90 to 99 % may be achieved in a biological-chemical treatment process, but in some cases, reductions may be poor.

Pathogenic bacteria have been proven to even multiply in the treatment process system (Koivunen *et al.*, 2002). From the study carried out by Koivunen *et al.*, (2002), microbial pollution to the receiving natural waters was not strongly reliant on the influent water quality. Instead, it was related to the efficacy of the treatment process in removing the organic load, suspended solids and other pollutants of concern. Result from the study has shown microbial reduction and improvement of water quality during the tertiary treatment process which involves contact filtration, compared to the result obtained in the secondary treatment where significant numbers of enteric bacteria were detected.

Another study as cited in Wery *et al.*, (2007) reported a significant diminution of indicator and pathogenic bacteria in the wastewater treatment plant due to the concentration of bacteria in sludge after settling. Settling is a part of the primary treatment which uses only physical separation methods to separate solid and particulate organic and inorganic materials from wastewater. This treatment permits sanitization as most microorganisms are associated with solids and in consequence of biological treatment such as pH, temperature, aeration and competition with indigenous organisms, causes the reduction of the bacterial concentration.

Other than that, another study by Decamp & Warren (2001) has shown a decreased by 50% of *E. coli* in the activated sludge process with the reason unknown, but thought that die-off could be the main cause involved. The die-off could be due to competition with indigenous microorganism which has better adaptability to the environment in the treatment system. Another interesting discovery made known in the same study as well is that *E. coli* from wastewater that is undergoing secondary treatment could be removed as a result of predation by ciliated protozoa.

In this wastewater treatment study, indicator organisms such as enteric bacteria, coliform, faecal coliform and heterotrophic bacteria are of great interest due to the presence of biodegradable materials in greywater which favours their growth (Ottoson & Stenström, 2002). Besides, diseases caused by these organisms such as diarrhoea, urinary tract infection, dysentery and typhoid fever is a disturbing affair which imparts the need of this study to be carried out so as to ascertain whether the greywater treatment process is efficient and functioning properly.

The rationale of this study is that the greywater treatment system can be improved in order to reduce the number of pathogen to a level where human as well other living organism are not at risk of being infected due to poor water quality.

### **1.1 Objectives:**

With the existing greywater treatment facilities (GTF), the water quality should not be a disturbing issue anymore. However, the efficiency of the GTF in removing contaminants is still of a critical concern. Therefore, the objectives of this study are as follows:

- To determine the associations between the removal efficiency of microorganism and other greywater pollutants, and the effect of the treatment process, in this case, the efficiency of the different treatment stages namely oil and grease trap as well as the biofilter tanks.
- To compare the changes of the indicator organisms' concentrations at the influent, reservoir and effluent of the investigated system.

## **2.0 LITERATURE REVIEW**

### **2.1 Ecological Sanitation (EcoSan)**

A more holistic approach towards ecologically and economically sound sanitation is offered by the concepts referred to as "ecological sanitation". According to Ridderstolpe (2004), Ecological Sanitation (EcoSan) takes the principle of environmental sanitation a step ahead. Environmental sanitation means keeping our environment clean and out of harm's way where as EcoSan is premised on recycling principles. EcoSan is non-piped sanitation which involves only a small locally piped set-up.

There are a few driving factors that call out for the need of recycling such as water scarcity due to little rainfall and excessive demand as well as environmental and economic drivers (Jefferson *et al.*, 2004). The goal of this approach is not to promote a certain technology, but rather a new idea of dealing with what has been considered as waste in the past. Ideally, ecological sanitation systems make possible the complete recovery of all nutrients from faeces, urine and greywater for the benefit of agricultural productivity, water supply and sanitation, resource conservation, environmental protection, urban planning, irrigation and food security.

This concept has been known to be put into practice with ecological sanitation pilot projects in diverse regions around the world such as in Germany and Sweden. This sensible solution has been implemented in Kuching, Sarawak to solve the problem of organic wastewater with a pilot project of EcoSan involving nine houses at Hui Sing Garden, Sarawak. This is to document the environmental and economical efficiency of urban ecological sanitation which may therefore prove vital for the entire region.

## **2.2 Greywater**

According to Ridderstolpe (2004), 'greywater' is the term given to all used water discharged from a house, except for toilet water. Currently, the separation and reuse of greywater which hails from kitchen sink, hand basin, laundries and showers are increasing due to ecological, economical and structural importance (Gunther, 2000). Greywater has to be managed carefully to avoid waterlogging, bad odour, and the uncontrolled release of chemicals and other elements produced by man including microorganisms into the environment. In developing areas, the consumption of water and the use of household chemicals start to increase. As a result, the risk of environmental dilemma and human contact with non-healthy water conditions is rising. Therefore, proper design and maintenance of technical systems for collecting, treating and discharging greywater is needed.

### **2.2.1 Characteristics of Greywater**

This water is called 'grey' water because it turns grey and might produce foul smell if stored for a day or so. Toilet water is generally called 'blackwater'. Grey water contains suspended solids, biodegradable compounds, pathogens, nutrients, metal and heavy pollutants. Greywater is made up of wastewater from bathroom sinks, and showers ('light grey') and may also consist of more contaminated waste from dishwashers, laundry facilities, and, in some instances, kitchen sinks ('dark grey'). The content of greywater varies greatly and reflects the lifestyle of the residents, customs, installations, washing habits and the preference of household chemicals for washing-up and laundry. If not handled properly, greywater can be breeding sites for mosquitoes and contaminate groundwater as well as drinking water reservoirs.

### **2.2.1.1 Suspended solids and Biodegradable compounds**

Among the greywater characteristics is that it often contains high concentrations of suspended solids and easily degradable organic material, for example, fat, oil, and other organic substances from soap, shampoo, laundry detergents, toothpaste, food scraps, shaving cream, hair and lint. Recent research has proved that growth of enteric bacteria is favoured in greywater due to its content of easily degradable organic compounds (Ottoson & Stenström, 2002).

### **2.2.1.2 Microorganisms in greywater**

The fraction of pathogens in greywater is generally at a low level. As greywater does not contain faeces, it is normally regarded as rather risk-free. However, they may be minimal faecal contamination due to faecally contaminated laundry, childcare and showering. Based on previous studies (eg. Ottoson & Stenström, 2002; Koivunen *et al.*, 2002), numerous pathogenic organisms and microbial indicators have been found in greywater. These include coliform bacteria, faecal coliform including *E. coli*, fecal streptococci including enterococci, *Salmonella* sp., *Shigella* sp., *Vibrio cholerae*, *Campylobacter jejuni*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and *Clostridium perfringens*. Some of these organisms, such as enteric bacteria, are commonly found on the skin or in the oral cavity of humans and are shed routinely during bathing.

### **2.2.1.3 Nutrients, metals and other toxic pollutants**

Nutrients also occupy a small fraction in greywater. Low levels of nitrogen and other plant nutrients can be found in greywater but in certain circumstances. However, a high concentration of phosphorous which originates from washing and dish-washing powder can also

be obtained. Data from Sweden pointed out that greywater contains less than half the concentration of nitrogen and phosphorus compared with farm run-off (Ottoson & Stenström, 2002). According to Ridderstolpe (2004), the content of metals and organic pollutants in greywater is generally low, but can increase due to the toting up of environmentally hazardous substances such as preservatives, detergents, furnishing fabrics and floor coatings.

Nutrients such as calcium, manganese, zinc and a few others are known to serve important functions for the survival of bacteria in greywater. Table 2.0 represents the role of trace metals present in greywater (Jefferson *et al.*, 2004).

**Table 2.0: Role of trace metal present in greywater**

<b>Nutrient</b>	<b>Role of trace metal</b>
Ca	Cell transport systems and osmotic balance in bacteria
K	Cell transport systems and osmotic balance in bacteria
Fe	Growth factor in bacteria, fungi and algae
Mg	Enzyme activator for a number of kinases and phosphotransferase in heterotrophic bacteria
Mn	Activates bacterial enzymes. Often interchangeable with magnesium in kinase reactions
Cu	Bacterial enzyme activator required in trace quantities. Can inhibit metabolism.
Zn	Bacterial metallic enzyme activator of carbonic anhydrase and carboxypeptidase A. Dissociable on active site of enzymes. Stimulates cell growth.

The various components and physical parameters of greywater that may have negative impact on human health, environment, and other living organisms, can be summarized as in Table 2.1.

**Table 1.1: Physical parameters and components of greywater**

<b>Component</b>	<b>Examples</b>	<b>Reference</b>
Pathogens	<i>Shigella</i> sp., <i>Salmonella</i> sp., <i>Vibrio cholerae</i> , <i>E. coli</i> , <i>Campylobacter jejuni</i> , <i>Pseudomonas aeruginosa</i> , <i>Klebsiella pneumoniae</i> , and <i>Clostridium perfringens</i> .	Ottoson & Stenström (2002), Koivunen <i>et al.</i> (2002)
Chemicals and Organic pollutants	Shampoo, perfume, preservatives, hair dyes, dish washer, detergent, and floor cleaners,	Ridderstolpe (2004)
Metal and other pollutants	Dust, cutlery, dyes, corrosion of pipes, zinc.	Ridderstolpe (2004)
Physical parameters	Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Organic Carbon (TOC), Suspended Solids (SS), Dissolved Oxygen, Total Dissolved Solids (TDS), Total phosphorus, Total Nitrogen.	Koivunen <i>et al.</i> (2002)

### 2.3 Greywater treatment

At present, there are several technologies that have been used and some are in progress for greywater treatment. These technologies include basic coarse filtration, natural treatment systems, chemical processes, physiochemical processes, physical treatment such as settlement of solids and biological processes such as would be produced in an activated sludge plant or a domestic septic tank. The right choice of technology for greywater treatment depends on many factors such as the scale of operation, end use of water (Jefferson *et al.*, 2004), type of habitation, land-use patterns, existing drainage system, degree of pollution, social and cultural customs and practices (Ridderstolpe, 2004). The stages commonly involved in greywater treatment are as follows:

### **a. Pre-treatment**

As greywater is collected or stored for longer duration, particular technical components are required for pre-treatment and treatment of greywater. Problems such as clogging of system from fats and biodegradable organic compounds as well as occurrence of bad odour will arise if pre-treatment approach is not taken. Oil and grease trap are implemented into the system to separate suspended solids (SS) and grease from greywater. Other mechanical methods involved in removing SS are filters, screens and by gravity.

- **Holding tank**

This unit requires gravity and no pumping is needed. It is constructed for gravimetric separation of particles from the water. Floating particles are collected as scum in the top of the tank and sinking particles as sludge at the bottom.

### **b. Treatment**

The most suitable measure usually taken to remove easily degradable and to reduce levels of pathogens and microorganisms in greywater is to use aerobic attached biofilm techniques. These techniques can range from extensive land application systems to intensive applications such as trickling filter and biorotor.

- **Trickling filter**

This unit is composed of a bed (filter media) of porous material such as bricks, rocks, sand or other media with a high surface area and permeability. Greywater is first distributed over the surface of the media

by spraying technique, where it flows downward as a thin film over the media surface for aerobic treatment. The microorganisms will attach themselves to the bed. Then, the discharged water will be collected at the bottom through an under drain system (Okoh *et al.*, 2007).

- **Biorotor**

In biorotor, the media is mechanically rolled up and down in the water. Thus, the biofilm is fed intermittently by water, organic matter and air.

**c. Post-treatment**

After going through treatment, the water can be used for many possible purposes such as irrigation, discharged to surface water for creation of wetlands and percolation to groundwater.

### **2.3.1 Efficiency of Greywater treatment system**

A study which has been carried out by Jefferson *et al.* (2004), revealed that biological treatments may face performance and operational problems due the high COD/BOD ratio coupled with a nutrient and micro metal imbalance. Another suggested reason by Laine (2001) (as cited in Jefferson *et al.*, 2004) is that the inefficiency of greywater operational may be affected due to solids processing issue. Jefferson *et al.* (2004) stated that more stable effluent quality is often related to biological approach that contain tough barrier to solids namely membrane bioreactor and biologically aerated filter.

Nevertheless, this system has proved to be working well in a few studies such as one conducted by Godfree and Farrell (2005) (as cited in Wery *et al.*, 2007) has shown a significant diminution of indicator and pathogenic bacteria in the wastewater treatment plant due to the

concentration of bacteria in sludge after settling. This treatment permits sanitization as most microorganisms are associated with solids and in consequence of biological treatment such as pH, temperature, aeration and competition with indigenous organisms, causes the reduction of the bacterial concentration.

### **2.2.3 Applications of Greywater**

With the advancement of greywater treatment, the treated greywater serves many purposes. These treated sources are most commonly used for irrigation of golf courses and parks, vehicle washing, preservation of wet lands, agriculture, toilet flushing and groundwater recharge. These applications require water that are of geospecific quality but usually contain criteria based on solids, organic and microbiological content of the water (Ridderstolpe, 2004; Jefferson *et al.*, 2004).

### **2.4 Indicator Organism**

The quality of greywater can be measured, in microbiological terms, by measuring indicator organisms. In spite of this, concerns have been raised regarding the suitability of these indicators for evaluating the removal of other pathogens such as viruses and protozoa, leading to a general agreement on the need for alternative indicators (Lucena *et al.*, 2004). To function as effective indicators, such microorganisms should be detectable in an easy, rapid and inexpensive method. These microorganisms should also be a member of the intestinal microflora of warm-blooded animals and present when pathogens are present. However, there are possible reasons for pathogen to be absent but indicator to be present or the other way round. In short, there is no direct association between the presence of any indicator with enteric pathogens (Grabow, 1996 as

cited in Ashbolt *et al.*, 2001). Microbial indicators can be grouped into three categories (Ashbolt *et al.*, 2001):

- a) **Process indicators** such as total heterothrophic bacteria or total coliform demonstrate the effectiveness of a process such as water treatment management.
- b) **Index and model organisms** represent a group or species indicative of pathogen presence and behavior respectively.
- c) **Faecal indicator** implies the presence of organisms that indicates faecal contamination. Thus, they only infer that pathogens may be present.

Table 2.2 below is adapted from Curtis (2003) (as cited in Mara & Horan, 2003), regarding the concentrations of bacterial pathogens in untreated domestic wastewater.

**Table 2.2: The reported concentrations of bacterial pathogens in untreated domestic wastewater.**

<b>Organism</b>	<b>Disease</b>	<b>Reported concentrations per 100 ml</b>
Genus <i>Campylobacter</i>	Campylobacter enteritis	70-1630
Enterohaemorrhagic <i>E. coli</i>	Haemorrhagic colitis	Ubiquitous but no quantitative reports
Other enterovirulent <i>E. coli</i>	Diarrhoea and bacillary dysentery	Detective in wastewater but no quantitative reports
<i>Helicobacter pylori</i>	Gastritis	No quantitative reports
<i>Salmonella typhi</i>	Thyphoid	No quantitative reports
<i>Salmonella enteritidis</i>	Salmonellosis	20- >1800

#### 2.4.1 Coliform

The bacteria to be found in the coliform group can be divided into “faecal” and “nonfaecal” subgroups even though both can be found in polluted water or sewage. Coliform