WET COFFEE PROCESSING WASTEWATER CHARACHTERIZATION AND TREATMENT USING EFFECTIVE MICROORGANISM ACTIVATED SOLUTION

M.Sc. Thesis

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November 2012

Jimma University

School of Graduate Studies

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As thesis research advisor, I hereby certify that I have read and evaluated this thesis prepared, under my guidance, by Fuad Oumer, entitled Wet Coffee Processing Wastewater Treatment using Effective Microorganism Activated Solution (EMAS). I recommend that it can be submitted as fulfilling *thesis* requirement.

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WET COFFEE PROCESSING WASTEWATER CHARACHTERIZATION AND TREATMENT USING EFFECTIVE MICROORGANISM ACTIVATED SOLUTION

Thesis

Submitted to the School of Graduate Studies

Jimma University College of Agriculture and Veterinary Medicine

In Partial Fulfillment of the Requirements for the Degree of

Master of Science in

Horticulture (Coffee Tea Spices)

BY

Fuad Oumer Ali

November 2012

Jimma, University

DEDICATION

I would like to dedicate this M.Sc. thesis work to my mom and my son.

STATEMENT OF THE AUTHOR

First, I declare that this thesis is my work and that all sources of materials used for this thesis have been duly acknowledged. This thesis has been submitted in partial fulfillment of the requirements for M.Sc. degree in Horticulture (Coffee, Tea and Spices) at Jimma University College of Agriculture and Veterinary Medicine and is deposited at the university library. I solemnly declare that this thesis is not submitted to any other institutions anywhere for award of any academic degree, or certificate.

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BIOGRAPHICAL SKETCH

Fuad Oumer was born on May 10, 1983 in Chiro (Asebe Teferi) Town. He completed his elementary school at Chiro number three and Chiro Number one elementary schools from 1991 to 1998 and secondary school at Chercher Comprehensive secondary school from 1999 to 2002.

In 2002, he joined Jimma University College of Agriculture and Veterinary Medicine and graduated in 2006 with BSc. degree in Horticulture. After graduation he joined Werabe town Administration as environmental protection and community development expert and worked there from September 2006 to December 2007 as project planning, monitoring and evaluation expert from January 2007 to August 2008 and as town agriculture officer from September 2008 to October 2008. From November 2010 to September 2010 he was working as town Mayor special Advisor on public organization. Finally, in October 2010 he rejoined Jimma University College of Agriculture and Veterinary Medicine to pursue his M.Sc. degree in Horticulture (Coffee Tea Spice).

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LIST OF ABBREVIATIONS

APHA	American Public Health Association
ARDO	Agricultural and Rural Development Office
BOD	Biological Oxygen Demand
BU	Bulbulo
DFID	Department for International Development
DO	Dissolved Oxygen
EC	Electric Conductivity
EEPA	Ethiopian Environmental Protection Authority
EMAS	Effective Microorganisms Activated Solution
FDRE	Federal Democratic Republic of Ethiopia
GM	Gembe
GR	Geruke
IMF	International Monetary Fund
IPMS	Improving Production and Market Success
ICO	international Coffee Organisation
ITC	International Trade Center
LMC	Lutheran Mission Commission
MCL	Maximum Contaminant Level
EMoARD	Ethiopia Ministry of Agriculture and Rural Development
NRI	Natural Resource Institute
OB	Omobeko
OFCCU	Oromia Farmer's Coffee Cooperative Union
ORG	Oromia Regional Government
PSDC	PSDC, Penang Skills Development Centre
TDS	Total Dissolved Solids
TSS	Total Suspended Solids
USEPA	United States Environmental Protection Agency
WHO	World Health Organization

ABSTRACT

Wet coffee processing, if not conducted properly, can harm the environment. Hence, a study conducted in Ethiopia, Jimma Zone, Gomma and Mana wereda at Geruke, Gembe, Omobeko and Bulbulo sites from four points (upper stream of the river, pulping, fermentation tank, downstream). In order to characterize the wet coffee processing wastewater on physicochemical qualities and the laboratory treatment of pulping wastewater using effective microorganism activated solution at Jimma University environmental health laboratory. Physicochemical characterization of water samples taken from four points of all locations was done with three replication. The laboratory analysis was in factorial arrangement of CRD designed with two factors and three replications. The factors were pulping wastewater from four locations (Omobeko, Bulbulo, Gembe and Geruke) and the effective microorganism with two types activation and five levels. $EMAS_1$ with 0ml, 2ml, 4ml, 6ml, and 8ml and $EMAS_2$ with 0ml, 3ml, 6ml, 9ml and 12ml. As a result, the wastewater showed very high BOD (12150 mg/l), TSS (1805.8mg/l), TDS (1702), turbidity (647.5 NTU), EC (568.5 µS/cm), NO_3 (24.87 mg/l), NH_3 (11.06 mg/l), PO_4 (4.85 mg/l) and low pH (2.95) and DO (0.02 mg/l) values. The $EMAS_1$ treated pulping wastewater show a reduction of 49.5 - 60.5% BOD, 23.05 - 26.88% NO_3, 41-46% NH_3 36 - 52% PO_4, 24 - 33% EC, 35 - 47% turbidity, 37.5 - 52.5% TSS and 41.7 - 49.1% TDS and increment of pH by 1.27 - 1.69 and DO by 1.4 - 2.5mg/l amount for effluent treated with 4ml $EMAS_1$. Whereas $EMAS_2$ pulping wastewater was show reduction of 40 - 45% NO_3, 72 - 82% NH_3, 30 - 52% PO_4 when treated with 9ml $EMAS_2$ and 49.5 - 60.5% BOD_5, 29 - 40% EC, 40 - 53% turbidity ,39.9% - 52.5% TSS and 47.8% - 56.6% TDS and increment of pH 1.1 - 1.59 and DO by 1.99 - 2.8 mg/l amount when treated with 6ml $EMAS_2$. The quality of wastewater improved by using EMAS; the relatively good improvement of wastewater was on the use of $EMAS_2$ 6ml due to its economical benefit and better wastewater quality improvement than $EMAS_1$ of 4ml. So using 6ml $EMAS_2$ advantageous to use for the treatment of wet coffee processing wastewater as far as this study was concerned. The wastewaters from four locations were highly polluted with the high pollution load on pulping and fermentation wastewater. Hence, there should be strict environmental regulation imposed on coffee processing stations not only during establishment but also during subsequent operations. However, further multidisciplinary researches are imperative on the impact of wet coffee processing wastewater and its treatment. It is advisable to do confirmation of this research both on in-situ and ex-situ condition.

Index Terms: wet coffee processing wastewater, physicochemical characteristics, pulping wastewater, characterization, treatment, and effective microorganism activated solution

1. INTRODUCTION

Coffea arabica L., as it has been written and rewritten, finds its birthplace in southwestern Ethiopian forests (Laurent 2009). The main coffee of commerce, Arabica coffee (*Coffea arabica* L.), is indigenous to the high lands of Ethiopia and the Boma plateau in the Sudan (Wrigley, 1988). It is one of the stimulating crops widely drunk in the world next to tea (Ali, 1999). The relationship between Ethiopians and coffee is deep-rooted and coffee production and consumption closely intertwined with Ethiopian history, culture and economy. Coffee has been cultivated, traded and consumed over centuries and still play a significant role in the daily life of most Ethiopians and for the state of Ethiopia as a whole (Stellmacher, 2007).

Coffee processed by two widely known methods - dry and wet coffee processing. From exported coffee of Ethiopia 70% is sun-dried while the rest 30% is wet processed coffee (washed and semi-washed) (FDRE, 2006). In contrast to the dry method, wet processing requires a higher degree of processing know how and is applied mainly for Arabica coffee (Vincent, 1987) the sole species grown in Ethiopia. Wet processing is producing a higher quality product, so called “mild coffees” (Rothfos, 1979). Coffee processing in Ethiopia is carried out by small and medium sized plantations for whom there are inadequate compulsions to carry out efficient resource recovery and wastewater treatment (Solomon *et al.*, 2008).

In Ethiopia 965 wet coffee processing stations 580 in SNNPR and 385 in Oromia regions have been found (Simayehu, 2008). The wet coffee processing system is characterized with: the excessive consumption of water (at each stage of the process), which generally varied greatly from 59-105m^3 and 24-114m^3 of water per tons of dry parchment (Mburu *et al.*, 1994); 50m^3 per ton for the washing and demucilaging (IPMS, 2007). The process of separating the beans from coffee cherries during wet processing generates enormous volumes of waste material (solid and liquid). The wastewater discharged during the operations constitutes high organic matter either dissolved in wastewater or associated with suspended material. The significant characteristic of such a pollutant is its bio-degradability (Anon, 1991), and the reduction of the level of oxygen in water. This results in objectionable colour, turbidity, smell

and sometimes death of aquatic organisms, which according to Adams and Dougan (1987), implies a reduction in the rivers' resource value. In addition, the composition of coffee pulp is organic and mainly contains carbohydrates, proteins, fibers, fat, caffeine, polyphenols, and pectins (Gathua *et al*., 1991). According to Alemayehu and Devi (2008) this wastewater which was discharged to the nearby water bodies and thus causing many severe health problems on like dizziness, eye, ear and skin irritation, stomach pain, nausea and breathing problem among the residents of nearby areas when the people exposed to highly polluted river for extended period of time.

Despite the factors mentioned the presence of cheap labour market in Ethiopia coupled with the high demand for wet processed coffee has increased the number of coffee processing stations in the country. Therefore, the government has encouraged cooperatives and traders to invest in machinery to raise the output of washed coffee (LMC, 2003). In 1980/81 washed coffee was only 9.1 per cent of total coffee exports but, its amount was raised to 32.7 percent by the year 2004/5 (FDRE, 2006). However, the rise in the number of coffee processing stations has not been coupled with environmental impact assessment that contaminates surface water bodies with their waste products (DFID, 1999; IPMS, 2007).

However, the lack of the hitherto enforcement of environmental assessment before issuing the permit to the newly established processing stations and failure of monitoring and evaluation activities of the existing processing stations have resulted in the generation of huge amounts of processing by products which have the potential of polluting the environment. In general, water pollution is a pressing problem in developing countries, particularly, where there is high population growth, great development demands, high waste production without well developed waste treatment technologies, and lack of comprehensive environmental policies and water quality monitoring system and standards (James, 2005). As a result, the Environmental Policy of Ethiopia (Proclamation 300/2002) recently formulated to foster monitoring of pollution and management of hazardous waste in the country (IPMS, 2007). The policy emphasized the need and importance of environmental impact assessment for industrial activities in the country.

Numerous processes were to clean up wastewaters depending on the type and extent of contamination. Most wastewater treated in industrial-scale wastewater treatment plants, which may include physical, chemical and biological treatment processes. However, this method required a high cost to manage. The practice of re-circulation recommends 22.5m^3 of water per ton of parchment processed, non-returnable to watercourse (Anon, 1991). However, a recent survey found that the recommended volume exceeded in all cases of operation (Mburu *et al.,* 1994). Some of the key issues that environmental assessment has to address are the social and economic impacts of proposed projects (Abaza *et al*., 2004).

Introduction of EM in treating wastewater seems to overcome this problem due to the fact that it can be obtained easily and does with minimum cost even it can be made in home from kitchen garbage. Moreover, EM technology does not use highly sophisticated machine compared to wastewater treatment plants. Besides reducing cost, EM technologies also help in protecting the environment by reducing garbage. Hence, these study done by selecting the four wet coffee processing sites. Three from the high coffee production weredas of Jimma Zone three sites (Omobeko, Bulbulo and Gembe) from Gomma wereda and one (Geruke) from Mena wereda and assessing the physicochemical characteristics of four points of wet coffee processing wastewater and the evaluation of different EMAS concentration for wet coffee processing wastewater treatment with the following general and specific objectives.

General Objective

To characterize wet coffee processing wastewater and to evaluate the level of different EMAS concentration the wastewater to propose possible solution for the coffee wastewater treatment

Specific objective

1. To characterize the physicochemical properties of wet coffee processing wastewaters

2. To evaluate different concentration of EMAS for the wet coffee processing wastewater treatment

2. LITERATURE REVIEW

2.1. Coffee Plant

The coffee tree is a shrub with a straight trunk which can survive for about 70 years (Mutua, 2000). Botanists classify Coffee as a member of the Rubiaceous family. The genus *Coffea* L. consists of approximately 100 species (Bridson and Vercourt, 1988). However, the commercial coffee production relies on two species of coffee Arabica (*Coffea Arabica*) and Robusta (*Coffea Canephora*). Coffee Arabica is considered as a superior quality coffee, and contributes over 65 percent of the world's coffee production, while Robusta shares around 35 percent (Scholer, 2004).

More than 50 developing countries, 25 of them in Africa, depend on coffee as an export, out of these 17 countries earning 25 percent of their foreign exchange from coffee. Coffee is providing income for approximately 25 million smallholder producers (DFID 2004), and employing an estimated number of 100 million people. Coffee has been cultivated, traded and consumed over centuries and still play a significant role in the daily life of most Ethiopians and for the state of Ethiopia as a whole (Stellmacher, 2007).

2.2. Coffee Production in Ethiopia

Coffea arabica L., as it has been written and rewritten, finds its birthplace in southwestern Ethiopian forests even if Linnaeus gave its scientific name in 1753 paying tribute to his future country. Ethiopia also has the best inherent potential for coffee production (FDRE, 2006). The total area covered by coffee in Ethiopia is about 600,000 hectares, with a total of annual coffee production ranges from 300,000-350,000 tons, which is about 600kg ha^{-1} on average. Of which, the coffee produced by small-scale subsistent farmers covers more than 90% while the remaining comes from private and government owned large-scale farms (MoARD, 2008).

Coffee farming systems in Ethiopia conventionally divided into four categories: forest coffee, semi-forest coffee, garden coffee and semi-modern plantation. Yields are considered to be

very low compared to other countries, with estimates of less than 200 kg per ha for forest coffee and around 450–750 kg per ha for semi-modern coffee plantations (FDRE, 2003a). Most or all except for state farm which use some fertilizer coffee farmers do not use fertilizers, pesticides or herbicides (LMC, 2000).

The latest report by the International Coffee Organisation (ICO, 2012) puts Ethiopia as the world's 5th largest coffee producer next to Brazil, Vietnam, Indonesia and Colombia and 9th in consumption for the 2011/2012 crop year. According to ICO (2012), Ethiopia has export 6,500,000bages (391477.273 tons) in 2011/12 but the export was decrease by 13% from the previous year which was 7,500,000bages (451704.545 tons) in 2010/11.

2.3. Coffee Processing

Coffee is processed in two ways; wet processing by which parchment coffee is prepared and dry processing by which cherry coffee is prepared. Parchment coffee prepared by the wet method is popular in the market. For preparation of both parchment and cherry coffee, coffee fruits are picked as and when they become red ripe. The under – ripe and over - ripe fruits cause deterioration in quality and is rejected. The collecting bags for harvested fresh cherries washed and dried frequently. Bags in which fertilizers, pesticides and fungicides are stored cannot use for this purpose (Sengupta, 2006).

2.3.1. The dry method

Coffee bean is drying in the cherry, part of the drying sometimes-taking place before harvesting. Picking and drying are often the only operations undertaken by the small-scale grower, especially for agro-forest coffee. When these carried out, dried beans (*janfal*) addressed to central market sent to local processing plants where the dried pulp and the parchment removed in one operation. The dried bean then sent to Addis Abeba, where it processed again in export case. Dry coffee still constitutes the bulk of Ethiopian coffee produced but it considered that is impossible to produce a first class coffee by this method.

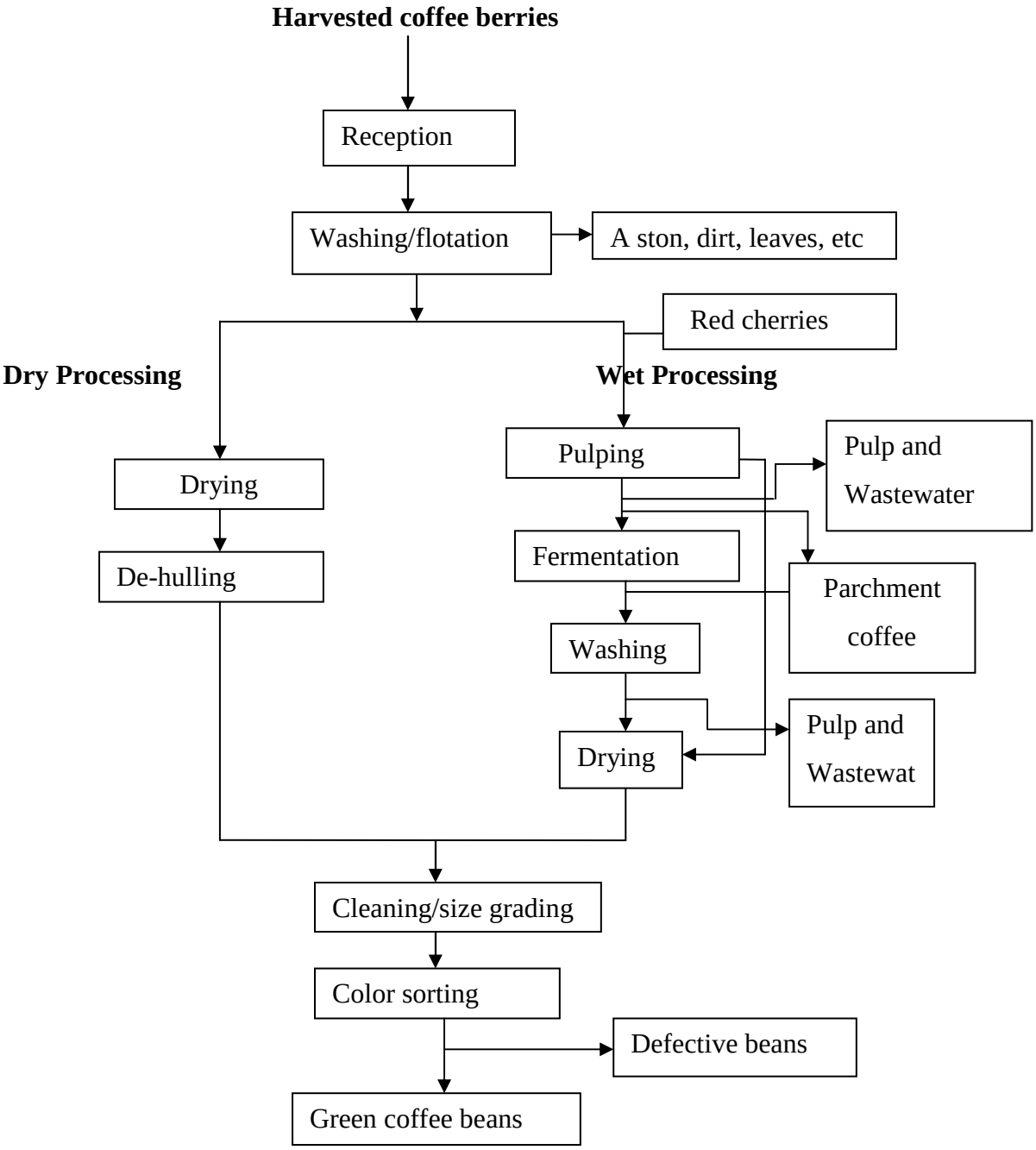

Figure 1 schematic representation of dry and wet coffee processing (Jan and Calvert 2002)

2.3.2. The wet method

This new procedure introduced to Ethiopia during the 1970's socialist regime and is now preferred to reach quality goals and prices. The sitting of the pulping is near a river or stream, which can to provide with good supply of water, and evacuation channel, at picking time since the whole process carried out in presence of clean water. The pulpier were laid out so that the coffee is moving through each stage of the process by gravity flow (Bantee, 1995). The main constraint of this method is the water contamination inherent to rubbish then evacuated in streams feeding neighboring households. Moreover ripe cherries start to ferment very soon after it is picked and become brown. Therefore, it is important to keep it in the shade and to process it as soon as possible, on the day it picked. The final product is coffee with parchment. This latter will be removed in Addis Abeba, once coffee with parchment has been bought to the Ethiopia Commodity Exchange, before getting exported (Bossolasco, 2009).

The purpose of fermentation of coffee is to ease the removal of a layer of mucilage from the seed/inner integument to which it adheres. After washing, mucilage must be completely removed from parch before drying. Given that pulping always takes place in the late afternoon through the evening and requires everyone's attention, fermentation periods will tend to be about 18h, 40h or 64h. Robusta usually requires at least one day more than Arabica (FAO, 2005). During wet coffee processing $50M^3$ of water used per ton of coffee in Gomma wereda as the IPMS (2007) report indicate.

2.3.2.1. De-pulping

Wet coffee processing procedure requires the mechanical removal of the pulp with the help of water, which produces considerable amount of wastewater. The water used for de-pulping of the cherries referred to as pulping water. It accounts for just over half of the water used in the process (Enden and Calvert, 2002). Pulp and mucilage consist of complex carbohydrates, such as pectin, and high content of proteins. Consequently, pulp water is rich in sugars and other substrates because of the fermentation activities of bacteria on the cherries. It also contains

acids and other toxic chemicals like polyphenolics (tannins and caffeine) (Solomon *et al.*, 2008).

2.3.2.2. Washing

According to Chanakaya and Alwis (2004), washing of the fermented beans leads to the release of fermentable sugars and proteins, which are amenable to rapid biodegradation. The fermentation of the sugars creates acidic conditions in the washing water lowering the pH to levels around four. This considered detrimental for aquatic life when such wastewater discharged directly into surface waters (Enden and Calvert, 2002).

These by-products of coffee processing (coffee pulp and processing wastewater) result in bad odour in the surrounding areas, breeding of disease vectors, when dumped around the processing plants and pollution of ground water and surface water bodies through leaching and run-offs, respectively (Mburu *et al.*, 1994).

2.4. Water Pollution

Water is used for navigation; as a coolant, cleanser, and diluting; for recreational purposes; as a food resource; as a means of power; as a source of tranquil aesthetic enjoyment; as a transporter of disease; as a container for nuisances; and finally, as the once unlimited area for disposal of society's wastewater products. It is indeed a wonderful chemical medium, which has unique properties of dissolving and carrying in suspension huge varieties of chemicals because of it can easily contaminated. Much of water pollution is due to anthropogenic activities (Santra, 2001).

Generally, water pollution caused by the presence of some organic, inorganic, biological, radiological or physical foreign substances in the water that tend to degrade its quality. The presence of undesirable and hazardous material and pathogens beyond certain limit will also cause water pollution (Narayanan, 2007). Commercial, industries, agricultural and domestic activities are the main causes for water pollution (Santra, 2001).

2.5. Characteristics of Wastewater

Wastewater quality can be defined by physical, chemical, and biological characteristics. Physical parameters include color, odor, temperature, solids, turbidity, oil, and grease. Solids further classified into suspended and dissolved solids as well as organic (volatile) and inorganic (fixed) fractions. Chemical parameters associated with the organic content of wastewater include the Biochemical oxygen demand (BOD) (Manahan, 2001). Inorganic chemical parameters include pH, acidity, nutrients and the like (Carl *et al.,* 1999).

2.6. Characteristics of Coffee Wastewater (Effluent)

The main pollution in coffee wastewater stems from the organic matter set free during pulping particularly the difficult to degrade mucilage layer surrounding the beans. The mucilage contains mainly proteins, sugars and pectin. The pectin make up the gel like constitution of the mucilage by polymerizing galacturonic acid made from sugars. The sugars contained in the mucilage will quickly ferment to alcohol and CO_2. However, in this situation the alcohol quickly converted to vinegar or acetic acid in the fermented pulping water. Other substances in coffee wastewater are toxic chemicals like tannins, alkaloids (caffeine) and polyphenolics. These components make the environment for biological degradation of organic material in the wastewater more difficult (Solomon *et al.*,2008).

According to IPMS the wet coffee produced per year in Gomma wereda is 3000tons and $50m^3$ of water used per ton of wet processed bean and for 3000tons $150,000m^3$ of water used. This wastewater released to the nearby river or vegetation. A study in Central America in 1998 showed that processing 550,000 metric tons of coffee generated 1.1 million metric tons of pulp and polluted 110,000 cubic meters of water per day. This was equated with a city of 4 million populations dumping raw sewage into the region's waterways. In that period, Costa Rica estimated that coffee processing was responsible for two-thirds of the pollution, as measured by total Biochemical oxygen demand, in its rivers. As freshwater supplies become scarce and demand for fresh water increases, this issue is become even more important (Manion *et al.,* 1999 in Jacobs 2009).

2.7. Physicochemical Characteristics of Wastewater

2.7.1. Dissolved oxygen

Dissolved oxygen refers to the amount of oxygen dissolved in water. Oxygen is one of several dissolved gases important for aquatic life and it is the single most important component of surface water for self-purification processes. Dissolved oxygen is usually expressed as a concentration of oxygen in a volume of water (milligrams of oxygen per litre of water, or mg/l) (Vousta *et al*., 2000). Primary sources of oxygen in surface water are photosynthesis by aquatic plants, algae and diffusion of atmospheric oxygen across the air water. Typically, the concentration of dissolved oxygen in natural surface water is less than 10 mg/l (Canada EPA, 1994). Dissolved Oxygen concentrations below 5mg/l may adversely affect the functioning and survival of biological communities (Chapman and Kimstach, 1996).

2.7.2. Biochemical oxygen demand

Wastewater quality can be defined by physical, chemical, and biological characteristics. Chemical parameters associated with the organic content of wastewater include the Biochemical oxygen demand (Jan, 2008). Biochemical oxygen demand (BOD) is amount of oxygen required to decompose a given amount of organic material. Water ordinarily contains some natural BOD such as plant debris and wildlife feces. The increased organic loading stimulated microbial decomposition that utilized dissolved oxygen (DO) in the surface water. This consumption of DO and attendant DO depletion in many cases led to the development of anaerobic conditions that could not support desired aquatic life, such as fish, and also caused aesthetic water quality problems. High BOD means the requirement of dissolved oxygen by microorganism present in the wastewater thus reduces or depletes the DO in water. (Marquita, 2010). BOD is a measurement that allows comparing the relative polluting strength of different organic substances. This method attempts to replicate the oxidation condition found in the natural environment (Roger, 1994).

According to Ronaldo *et al.,* (2006), oxygen demand is the measurement of amount of dissolved oxygen consumed in five days by biological processes breaking down organic matter. That part of oxygen demand associated with biochemical oxidation of carbonaceous, as distinct from nitrogenous, material. Biochemical oxygen demand could be determined by allowing biochemical oxidation to proceed, under conditions specified in standard methods, for 5 days (BOD_5) (DAUSCE, 1999).

2.7.3. Temperature

Temperature is a one of the physical factor which determine the integrity of ecosystem (Canada EPA, 1994). Water temperature tells many things about the health of a river. Cold water holds more oxygen than warm water. As temperature increase the amount of oxygen needed by aquatic organisms increased. Warm water enters the river, raises the temperature of the downstream area and changes oxygen levels (Abdullah *et al.,* 2011). Each microorganism is able to grow within a specific temperature range. While single species can grow only over a 40°C range, others can grow below 0°C to above 90°C (Nicholas 1996). The temperature affects the biological wastewater purification due to the dependent nature of microorganisms. Based on optimum growth temperatures, microorganisms can be classified as: psychrophiles (less than 20°C), mesophiles (20-45°C), and thermophiles (greater than 45°C).

2.7.4. pH and water

According to WHO (1984; 2006) pH is a measure of the hydrogen ion concentration in solution. pH values reflect the solvent power of water, thereby indicating its possible chemical reactions on rocks, minerals, and soils. Most microorganism exhibit optimal growth at pH values between 6.0 and 8.0 and most cannot tolerate pH levels above 9.5 or below 4.0 (Danalewich *et al.,* 1998). The pH reading of a solution is usually obtained by comparing unknown solutions to those of known pH (Firdaus, 2007).

2.7.5. Turbidity

Turbidity is the presence of suspended materials such as clay, silt, finely divided organic material, plankton, and other inorganic material. Turbidity inhibits light transmissions in the water. In the sense that light transmission is inhibited, is known as turbid. Turbidity is undesirable for many reasons some are: aesthetic considerations, solids may contain, pathogens or other contaminants. Turbidity, although not a hazard itself, may be an indication that pollution introduced into the water bodies (Nicholas, 2002). In wastewater turbidity is used as an indicator of the reduction of light due to haze, smoke and other particles (James and Edward, 2006).

2.7.6. Nitrogen

Nitrogen occurs in five major forms in aquatic environments: organic nitrogen, ammonia, nitrite, nitrate, and dissolved nitrogen gas. Ammonia is one of the intermediate compounds formed during biological metabolism and, together with organic nitrogen, is considered an indicator of advanced pollution. Aerobic decomposition of organic nitrogen and ammonia eventually produces nitrite and finally nitrate (Joanne, 2001). High nitrate concentrations, therefore, may indicate that organic nitrogen pollution occurred upstream that the organics have had time to oxidize completely. Ammonia converts to Nitrate under certain condition of excess oxygen the process is Nitrification. Denitrification is the conversion of nitrate to nitrogen gas, which requires the presence of denitrifying bacteria (Marcel, 2006). Both nitrification and denitrification require ideal conditions for the most favorable results, and may occur in the same tank, but at different times and in different environments. The principal ingredients required for nitrification and denitrification are sufficient oxygen levels and adequate bacterial concentrations (DAUSCE, 1999).

Excess nitrates also induce health risks, specifically methemoglobinemia or blue-baby syndrome, especially in children. Accordingly, the WHO (1984) sets a maximum contaminant level (MCL) for nitrate-nitrogen concentrations at 10 mg/l for safe drinking water. Ammonia levels in excess of the recommended limits may harm aquatic life. Although the ammonia

molecule is a nutrient required for life, excess ammonia may accumulate in the organism and cause alteration of metabolism or increases in body pH (McGraw, 2006).

2.7.7. Phosphorus

The nutrient phosphorous mainly occurs in solution as particles or waste elements in microorganism, the most common forms are: orthophosphates (PO_4^{3-}, HPO_4^{2-}, $H_2PO_4^-$, and $H3PO_4^-$); polyphosphates (P_2O_7) (DAUSCE, 1999). As in the case of the nitrogen forms ammonia, nitrite and nitrate, orthophosphates can also cause eutrophication in receiving streams (Arcadio and Gregoria, 2003). Much of the excess phosphorus available to plants in the environment comes from farm and lawn fertilizers, runoff containing soil-bound phosphate, yard waste, and runoff from animal feedlots, storm water and certain industrial wastewaters. Though phosphorus is essential for life, it may become toxic, when found in high level (Marquita, 2010). High phosphorus concentration, together with nitrate and carbon dioxide is often associated with heavy aquatic plant growth. Small amount of phosphate (to the level of 0.01mg/l) can have measurable effect on aquatic communities (USEPA, 2006).

2.7.8. Total suspended solid (TSS)

Total suspended solids (TSS) include all particles suspended in water which will not pass through a filter. Suspended solids are present in sanitary wastewater and many types of industrial wastewater. Suspended solids are visible in suspension in water (Ruth and Robin, 2003). As levels of TSS increase, a water body begins to lose its ability to support a diversity of aquatic life. Suspended solids absorb heat from sunlight, which increases water temperature and subsequently decreases levels of dissolved oxygen (warmer water holds less oxygen than cooler water). As plants and algae produce less oxygen, there is a further drop in dissolved oxygen levels. TSS can also destroy fish habitat because suspended solids settle to the bottom and can eventually blanket the riverbed (Joanne, 2001).

2.7.9. Total dissolved solids (TDS)

Total Dissolved Solids (TDS) is an index of the amount of dissolved substances in water (Canada EPA, 1994). Solids in the water that remain after filtration and evaporation as residue are called total dissolved solids (TDS) and used as indicator of water quality. TDS may be organic or inorganic and may cause physiological effects, as well as color, taste, and odor problems (Joanne, 2001). The presence of such solutes alters the physical and chemical properties of water (USEPA. 2007). The palatability of water with a TDS level of less than 600 mg/l is generally considered to be good. However, drinking-water becomes significantly and increasingly unpalatable at TDS levels greater than about 1000 mg/l (WHO, 2006). If the TDS concentration exceeds 2000 mg/l, laxative effects have been observed in humans and livestock (Canada EPA, 1994).

2.7.10. Electrical conductivity (EC)

Conductivity is a numerical expression of water's ability to conduct an electrical current. The conductivity of water is dependent on the concentration of dissolved salts and temperature. Specific conductance provides a good indication of the changes in water's composition, especially in its mineral concentration but provides no indication of the relative quantities of the various components (Canada EPA, 1994). Conductivity in fresh water range between 10 and 1000μS/cm, but it may exceed the maximum value of the range in polluted waters (Chapman and Kimstach, 1996).

2.8. Effect of Wet Coffee Processing Wastewater

The wastewaters from wet coffee processing divided into two parts. Firstly, the pulping water with a high content of quickly fermenting sugars using enzymes from the bacteria on the coffee cherries. Secondly, depending on the processing method applied the water from fermentation/washing or the thick effluents from the mechanical mucilage removers (Wayan, 2005).

The environmental impact of wet and semi-wet processing is considerable. Problems occur through large amounts of effluents disposed into watercourses heavily loaded with organic matter rather it is than inherent toxicity. Providing the self-purification of the watercourse exceeded, the microbial degradation reduces the level of oxygen to anaerobic conditions under which no higher aquatic life is possible (Solomon *et al.,* 2008).

According to IPMS (2007) one of the exciting environmental issues is Pollution of rivers and streams by wet coffee processing plants, especially during the beginning of the dry season when rivers and streams carry lower water volumes but when coffee processing is peak. Pollution of rivers and streams as a result of wet coffee processing plants has been raised by the community as affecting both human and livestock health. Jan (2008) reported that the pollution to produce one ton of coffee equivalent to the daily pollution load of domestic swage output about 2000 people.

Considering that about 3,000 tons of washed coffee produced in Gomma and 50 M^3 of water used per ton of coffee bean, about 150,000 M^3 of wastewater per year discharged into the rivers in the wereda. However, if all wet coffee processing plants in Gomma are working at full capacity (130 tons/season/plant), they have a collective capacity of processing 6,500 tons of clean dry coffee beans. This will generate 325,000 M^3 polluted water each year. At the same time, a similar amount of pulp and hull dumped around the rivers. This generates pungent smell. As this takes place in only four months each year, the level of concentration of the organic waste in the rivers is very high and hence impacts are also high (IPMS, 2007). In wet and semi wet coffee processing to produce one ton dry green bean 6.25 ton of red cherries feed in to the pulping mill; this process release 2.5 tons of pulp, 25000 liters of wastewater and 375kg BOD to the environment (Jan, 2008).

2.9. Studies on Physicochemical Characteristics of Coffee Wastewater

The physicochemical analysis of the wastewater generated from coffee processing plant that all the parameters like pH, BOD_5, total suspended solids, phosphate and nitrate were much more than the prescribed limits by WHO (Alemayehu and Devi 2007; Solomon *et al.*,2008)

and zero DO values (Solomon *et al.,* 2008). In Kenya, the coffee processing plant effluent BOD ranged from 1,800 to 9,000 mg/l for pulping waters and 1,200 to 3,000 mg/l for fermentation and washing water depending on the volumes of water used (Mburu *et al.,* 1994).

According to Yared *et al.* (2008) around Mena and Gomma wereda the physico-chemical parameters of coffee effluent consists of very high amount of BOD (2200 mg/l), TDS (1810 mg/l), NO_3 (26.4 mg/l), NH_4^+ (12.6 mg/l), low pH (4.3) and zero DO values. The study on wet coffee processing wastewater done by Abebe *et al.* (2011) indicates that during coffee-processing (wet) season, the highest BOD at the downstream of the river was 1,900 mg/l, the dissolved oxygen (DO) depleted to a level less than 0.01 mg/l, and thus curtailed nitrification. Temperature, pH, BOD, suspended solids, phosphate and nitrate for the pulping effluent were 25ºC, 3.57, 14,200 mg/l, 5870 mg/l , 7.3 mg/l and 23.0 mg/l, respectively, while for downstream these values were 22ºC, 4.45, 10,800 mg/l , 2080 mg/l , 4.6 mg/l and 10.5 mg/l, respectively (Alemayehu and Devi, 2008). When these values compared with WHO permissible limits for discharging of treated effluent for irrigation purpose it indicate that concentration of all parameters were very high (Alemayehu and Devi, 2008). The other study done on the coffee wastewater depict that generation high BOD to the environment compared to other processing industries for instance Distillery waste-water (100 g/liter), Meat processing waste-water (10 g/liter) and Paper mill waste-water (2 g/liter) where as the coffee waste-water generate 150 g/liter of BOD (Enden and Calver, 2002).

2.10. Coffee Wastewater Treatment Approaches

Coffee wastewaters are high in organic loadings and exhibit a high acidity. When washed or semi washed coffee is processed in large quantities, untreated effluents greatly exceed the self-purification capacity of natural waterways. In order to overcome the pollution potential of processing wastewaters, a clear understanding of wastewater constitution in inevitable to design a feasible treatment system. Especially when expanding wet coffee processing or setting up new large scale processing operations, treatment of wastewaters needs to be considered (Enden and Calvert 2002). The wastewater also consists of mucilage, pectin’s and

lignin, which contribute to higher pollution load (Mohana *et al.,* 2011). The different ameliorative techniques improved the quality of wastewater by reducing the pollutants present in it (Rodger *et al.,* 2005).

Research efforts are going on for development of such type of wastewater treatment technologies (Mohammed *et al.,* 1998; and Wang *et al.,* 2005). There are many methods developed for coffee wastewater treatment ranging from water recycling to chemical treatment. In order to protect the environment many countries implement policies to reduce water use and water recycling is one approach. Equipment innovations such as dry pulping and the systems also address this need. According to this study, water from pre-fermentation stage can possibly be recycled for few hours but some fresh water must be added in the process. Water from post-fermentation can be used cautiously and only once (FAO, 2000).

The environmental impact assessment should also take into consideration the effect of water pollution on the local residents, on health and their perceptions and attitude towards the industrial activities around them. Some of the key issues that environmental assessment has to address are the social and economic impacts of proposed projects (Abaza *et al*., 2004). As a result, the Environmental Policy of Ethiopia (Proclamation 300/2002) formulated to foster monitoring of pollution and management of hazardous waste in the country (IPMS, 2007). The policy emphasized the need and importance of environmental impact assessment for industrial activities in the country.

The which study done in Indonesia on minimization of water indicate that water minimization treatment from pulping stage has significant effect for wastewater volume, solid waste and number of beans. Minimum water volume can inhibit separation between pulp and beans then cause uncompleted peeling. Wastewater from minimization treatment has higher concentration of organic pollution while water volume is lower. Organic material in wastewater mainly comes from pulping process of fruit and mucilage. Low pH values (4.0) derived from fermentation process of sugar component from pulp and mucilage into acetic acid. Acidity and high organic content in wastewater require specific handling in order to environmental safety (Rizal *et al.,* 2012).

The research done by Zayas *et al.,* (2006) on the removal of the natural organic matter present in coffee processing wastewater through chemical coagulation flocculation and advanced oxidation processes (AOP). The reduction by using UV/H_2O_2, UV/O_3 treatment though it show a reduction in COD by 67% (Zayas *et al.*, 2006) it is difficult to treat large amount of effluent but it is uneconomical (Devi *et al.*, 2007).

The other method COD and BOD reduction from coffee processing wastewater using Avocado peel carbon by Rani Devi *et al.,* (2007) this also have a good result in reduction of COD and BOD by 98% and 99%. Avocado peel could be a good alternative than expensive activated carbon and hence wastewater treatment process can become very economical (Davi *et al.,* 2007) the limitation availability of avocado peel treat the huge amount of coffee wastewater. These costs can be prohibitive - particularly when world coffee prices are low (Mencia *et al.,* 1994). The anaerobic co-digestion also the alternative coffee waste management method which revealed TS and VS reductions in the ranges of 50-73% and 75-80%, respectively and the methane yield attained 75-89% of the theoretical methane potential (Neves et al., 2005).

The ameliorative techniques like sand, clay, sand clay, filtration and chemical coagulations as reported by Mohana (2011) are the mitigation methods to reduce the wastewater pollution load. The amelioration of Coffee Wastewater (CWW) the study revealed that coffee wastewater successfully used for irrigation after suitable treatments and proper dilutions (Mohana *et al.,* 2011).

Alternative wastewater treatment from coffee process and bio-fuel utilization as an effort to improve the energy efficiency and to reduce the greenhouse effect is determined by appropriate level on farmer level. It needs to do financial feasibility study as a base for implementation small coffee process with water minimization. This implementation affected by various factor such as technology, environment, social cultural, institutional and government policies. Therefore, it needs in-depth study that related to various factors to support small coffee processing with quality and environmentally friendly oriented (Rizal Syarief *et al.,* 2012).

The documented studies on the impacts of coffee waste in the coffee-producing regions such as Ethiopia are few in number. These studies based merely on the physicochemical parameters measured at one point in time at few coffee-processing plants during one season (Mwaura and Mburu 1998; Alemeyehu and Devi 2008). Therefore, there is a need to treat this problem through innovative and eco-friendly techniques. The other wastewater treatment is using effective microorganism using it many polluted industrial had been treated.

2.11. Effective Microorganism (EM) Technology

2.11.1. Discovery of EM

Dr. Teruo Higa developed the technology of Effective Microorganisms (EM) during the 1970's at the University of Ryukyus, Okinawa, Japan (Sangakkara, 2002). There are three types of microorganisms, which are categorized into decomposing or degenerative, opportunistic or neutral and constructive or regenerative. EM belongs to the regenerative category whereby they can prevent decomposition in any type of substances and thus maintain the health of both living organisms and the environment (PSDC, 2009).

EM is a natural pro-biotic technology developed based on beneficial and effective micro-organisms (EM). The microbes in EM are not-genetically-engineered or modified (GMO), non-pathogenic, non-harmful, and not-chemically-synthesized. The basic groups of these microorganisms in EM are lactic acid bacteria (commonly found in yogurt, cheeses), purple bacteria (photosynthetic bacteria) and yeast (Higa, 1991). Effective Microorganisms is a mixture of groups of organisms that has a reviving action on humans, animals, and the natural environment (Higa 1995) and has also been described as a multi-culture of coexisting anaerobic and aerobic beneficial microorganisms (EM Trading 2000).

2.11.2. Application of EM technology

Studies have suggested that EM may have a number of applications, including agriculture, livestock, gardening and landscaping, composting, bioremediation, cleaning septic tanks,

algal control and household uses (EM Technology, 1998). The biological treatment, especially the use of microorganisms to improve polluted water quality is effective and widespread due to low capital and cost compared to chemical treatments (Sangakkara, 2002). The effective microorganism (EM) technology for wastewater treatment appreciated comparative to other conventional methods because of its eco-friendly nature, and requires less inputs, cost and capital (PSDC, 2009).

The basic purpose of EM is the restoration of healthy ecosystem in both soil and water by using mixed cultures of beneficial and naturally occurring microorganism. The sustainability of the freshwater supply for domestic, agriculture and industrial use need to analyze as it would be a critical aspect of sustainable water management (Zakaria *et al.,* 2010). Therefore, EM applications include sustainable agricultural, industrial, health (livestock, pets and human), odour control, waste management and recycling, environmental remediation and eco-friendly cleaning (EM Technology, 1998). EM application in wastewater decreases the BOD, COD, TSS (Kanit *et al.,* 1998).

EM technology involves growing, applying, managing and re-establishing high populations of the beneficial microorganisms in an environment or system. A natural and organic technology found to be useful in numerous ways to benefit humankind. It discovered that EM exhibits very thorough effects, and its use now spreading into applications various fields ambitiously promoted as a means of solving many of the world's problems. Recently, EM has become a successful weapon in the cleaning of water in nature, especially in regions of Asia (Zakaria *et al*., 2010) like Malaysia, where the EM technology is gaining considerable attention for its potential to reduce nutrient levels of polluted water and restoring water quality (Zuraini *et al.,* 2010).

EM applied to polluted and putrefactive water, holds a dominant position in the layer of microorganisms and help ecosystems revive and reduce sludge and foul odours. The purpose of EM application is not to create apparently-clear water by chemical means but to revive the native function of aquatic ecosystem (PSDC, 2009).

2.11.3. EMAS (Effective Microorganism Activated Solution)

EM1 is the original solution required for the production of EMAS (effective microorganism activated solution). EM1 is a liquid concentrate of fermented EM whereby the microorganisms are alive but dormant. To activate the microorganisms, the concentrated solution needed to dilute with water, and further activation performed by addition of a certain amount of molasses, acting as a food source (Firdaus, 2007 and Zuraini *et al.,* 2010). Fundamentally, the activated EM suspension (EMAS) is a mixture of molasses (sugar cane) and EM in non-chlorinated water or rice rinse water (which provides the minerals for the multiplication of the microorganisms). The product kept in a warm place of 20º to 35º. Fermentation process occurs after the second day and EMAS is ready for use 7-10 days of incubation. At this point of time, the suspension has a pH from 3.5 to 4.0, releases a pleasant sweet to sour test, appears yellowish brown in colour, and needs to utilize within two weeks (Zuraini *et al.,* 2010).

2.11.3. Studies of EM application on treatment of wastewater

The study done in Canada on dairy wastewater the application of EM on treatment showed significantly reduced ammonium nitrogen, total phosphorus, total suspended solids and Biochemical oxygen demand after three months and was found to be a very efficient way of dairy wastewater treatment (Rashid and West, 2007). Effective microbes successfully used in wastewater treatment in Japan and are becoming popular in wastewater treatment in many countries. The chemical oxygen demand (COD) and BOD of domestic wastewater significantly reduced when treated with EM (Higa and Kanal, 1998). The application of EM in Canada show significant reductions of physicochemical characteristics of dairy wastewater observed. Treatment with EM, as done in many institutions in Japan overcomes this problem. Application of EM to wastewater reduced its toxic effects. Hence, this water could be used for crop production (Higa and Okuda, 2008).

Although there are reports pertaining to the physicochemical properties and environmental impacts of wet coffee processing to date there is no reported effort on the use of EM to treat

wet coffee processing wastewater. This definitely calls for a concerted effort to evaluate efficacy of different formulations of EM so that it can serve as an alternative mitigation strategy in the fight against environmental pollution.

3. MATERIAL AND METHOD

3.1. Study Area

The study was conducted at four processing plants, which are found in Mana and Gomma weredas of Jimma zone. Jimma zone is situated in south-western Ethiopia, lying between Latitude 6° and 9° north and Longitude 34° and 38° east. The altitude ranges from 880m to 3340m above sea level. It is one of the twelve zones in Oromiya (ORG, 2003). The experiment for wastewater treatment and wastewater characterization conducted at Jimma University environmental health laboratory. The four wet coffee processing plants used as effluent sources, of which three were from Gomma wereda (Omobeko, Bulbolo and Gembe/Bore) and one from Manna wereda (Geruke) which were purposively selected. The details about these Woredas is given as follows

3.1.1. Mana wereda

Mana woreda is one of the 18 weredas in Jimma zone known for predominantly growing coffee. It is located 370 km south west of Addis Ababa and about 20 km west of the Jimma Zone capital (Jimma). The mean minimum and maximum temperatures are 13ºC and 24.8ºC, respectively (ARDO, 2010). Based on long term (15 years) weather data obtained from the nearby JARC meteorological station, the average annual rainfall is 1523 mm. Coffee and 'Chat' are important cash crops (ORG, 2003) and over 5,000 hectares is planted with coffee (OCFCU, 2007). To produce one ton of green coffee bean 50 m^3 of water used (IPMS, 2007).

3.1.2. Gomma wereda

Gomma wereda is one of the 18 Woredas in Jimma zone known for predominantly growing coffee. It is located 390 km south west of Addis Ababa and about 50 km west of Jimma town. The annual rainfall varies between 800-2000 mm, while the mean minimum and maximum annual temperatures of the woreda vary between 7ºC-12ºC and 25ºC-30ºC, respectively (ARDO, 2010). Based on 15 years weather data obtained from Gomma woreda, the average

annual rainfall is 1524 mm. One of the ex-situ coffee conservation centre (Choche) in Ethiopia is found in this woreda (ORG, 2003). Altitudinal range of the woreda is between 1387-2870 m a.s.l (IPMS, 2007). The rivers drain to Ghibe/Omo to the east and Dedesa River to the north. It estimated that 22,561.82 hectar of land covered with coffee (IPMS, 2007) and government state farms (Gomma I and Gomma II) cover additional 2704 ha coffee. To produce one ton of green coffee bean 50 m^3 of water used (IPMS, 2007).

3.2. Sample Collection

The sample were collected from four points (upper stream of the river, downstream of the river, the wastewater from the pulping and the fermentation tank) using composite sampling techniques (Bartram, Balance 2001). The locations of the sampling points for the downstream of the studied rivers placed sufficiently from 100m below wastewater entry points to ensure dispersion of the wastewater over a wide cross section of the river. Water samples collected inserting the bottles to a 25cm depth to the opposite direction of the course of the river flow. The collected wastewater samples kept in iceboxes and immediately to Environmental Health Laboratory of Jimma University for chemical analysis following the standard methods (APHA, 1998).

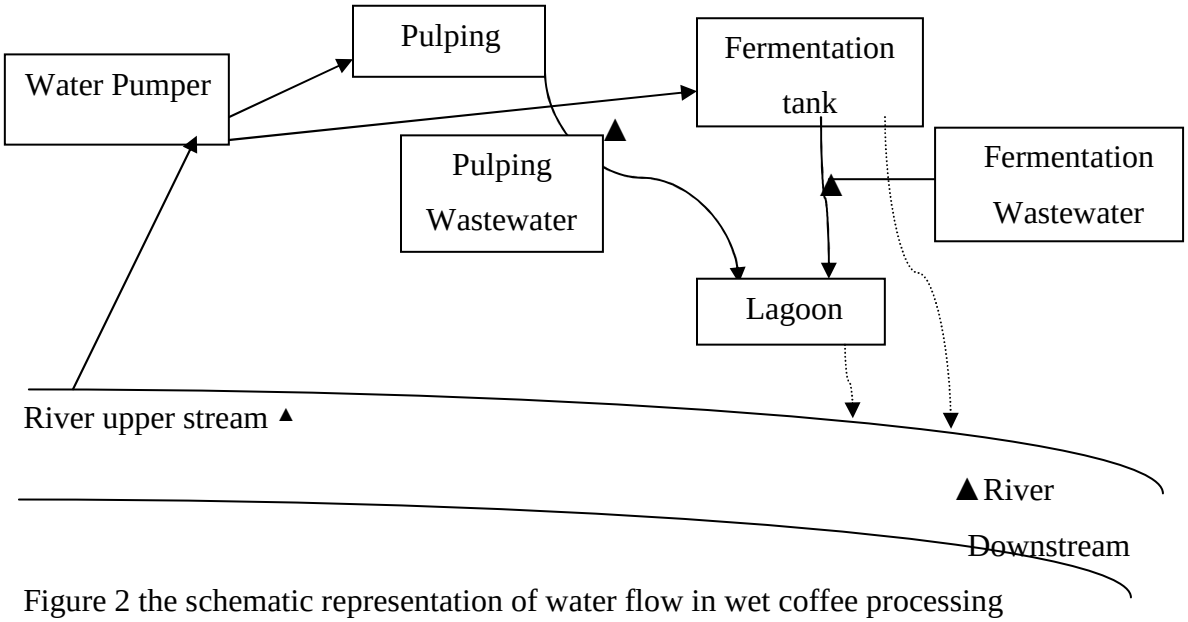

Figure 2 the schematic representation of water flow in wet coffee processing

Key ▲ = the point where the sample wastewater taken.

3.3. Experimental Design and Treatments

This study conducted in two parts. The first part was evaluation of the pollution load of the wastewater generated from wet coffee processing through physicochemical characteristic analysis. The physicochemical characteristics of wastewater taken from four locations (Omobeko, Bulbulo, Gembe and Geruke) at four different points from; upper stream of the river water, downstream of the river water, wastewater from the pulping and wastewater from the fermentation tank with three replications.

The second part was treatment of wastewater with effective microorganism activated solution executed in two phase. The first were treatment of pulping wastewater from four locations with five different concentration levels of activated effective microorganism solution with $EMAS_1$ (5% EM1, 5% sugar molasses and 90% distilled water). The second phase were treating the pulping wastewater with effective $EMAS_2$ (2.5% EM1, 2.5% molasses, 5% pulping wastewater and 90% of distilled water) both experiments were laid out in 4x5 factorial arrangement using completely randomized design (CRD) with three replication. Solution activated with effective microorganism applied on 200ml of wastewater, which taken from four different locations (wet coffee processing plants). The first five $EMAS_1$ concentration levels were 0, 2, 4, 6 and 8 ml and the next five applied with $EMAS_2$ of 0, 3, 6, 9 and 12 ml (Table 1).

The EM1 was at dormant state and must be activated using sugar molasses as a food source for microorganisms and chlorine free (distilled water) with 5% EM1, 5% molasses and 90% distilled water (Higa, 1991). As reported by Horticultural Research and Biological Husbandry Unit (2003) in New Zealand depict that effluent can also be used as an activation purpose with molasses and distilled water in different concentration level for activation with the ratio EM-1: molasses: dairy effluent: water = 2%: 2%: 5%: 91% and for the activation without effluent EM-1: molasses: water = 5%: 5%: 90%.

EM1 activated at Jimma University Environmental health laboratory. The substrates used for activation were EM_1 (commercially available EM), sugar molasses, distilled water and wet

coffee processing wastewater. Two types of effective microorganism activated solutions were prepared. The substrates in the first solution ($EMAS_1$) were 5% EM_1, 5% sugar molasses and 90% distilled water. The substrates of the second solution ($EMAS_2$) were 2.5% EM_1, 2.5% sugar molasses, 5% wet coffee processing pulping wastewater and 90% distilled water. After the substrates mixed in a plastic-bottle and well shake then put into an oven at 22ºC for 7days. On the seventh day, the activated EM solutions ready for treatment of the effluent (Higa, 1991; Firduas, 2007).

Table 1 Wet coffee processing effluent treatments (type and concentration of extended EM) for the two laboratory experiments.

Treatment (amount of applied activated EMAS)	Effluent amount to be treated (ml)	EM extended used1	Locations
Nil (control)	200	Nil	OB,BU,GM &GR
$2mlL^{-1}$	200	EMAS1	OB,BU,GM andGR
$4mlL^{-1}$	200	EMAS1	OB,BU,GM andGR
$6mlL^{-1}$	200	EMAS1	OB,BU,GM andGR
$8mlL^{-1}$	200	EMAS1	OB,BU,GM andGR
Nil (control)	200	Nil	BO,BU,GM andGR
$3mlL^{-1}$	200	EMAS2	BO,BU,GM andGR
$6mlL^{-1}$	200	EMAS2	BO,BU,GM andGR
$9mlL^{-1}$	200	EMAS2	BO,BU,GM andGR
$12mlL^{-1}$	200	EMAS2	BO,BU,GM andGR

OB=Omobeko; BU=Bulbulo; GM=Gembe; GR= Geruke; $EMAS_1$=effective microorganism activated solution one; $EMAS_2$ =effective microorganism activated solution two.

3.4. Physicochemical Property Measurements

The physicochemical data for both before and after treatment of wastewater measured according to American public health association manual (1998). Before the treatment, the physicochemical properties of the wastewater taken from four different places (upper stream, downstream, fermentation tank and pulping effluent) were determined. Then, for treated pulping effluent with effective microorganism activated with and without the pulping effluent measured after four days of treatment at Jimma University environmental health laboratory. The physicochemical characteristics: temperature, pH, DO, turbidity and EC were measured onsite. TSS, TDS, BOD, NO_3, NH_3 and PO_4 analyzed in laboratory. Methods used for the measurement of all parameters as prescribed by American Public Health Association handbook (APHA, 1998).

3.4.1. Temperature, pH, DO, Conductivity and Turbidity

The data for temperature, pH, DO, conductivity and turbidity were measured using portable probe ("HQ40D multi-Versatile pH-Oxygen-Conductivity meter" made in USA model) for thermometer, pH meter, DO meter and Conductivity and Hach portable turbidity meter ("TBIR 1000 series Bench turbidity meters" made in USA) for turbidity during sampling and after EMAS treatment of pulping wastewater in the laboratory.

3.4.2. BOD_5

The [BOD] of a water or wastewater sample is measured using a bioassay--a test in which organisms (biota) are used to determine (assess) the amount of a target substance. The BOD_5 test is a standardized test that provides information regarding the organic strength of wastewater. The amount of oxygen consumed in a sample within a five-day period measured under carefully controlled and standardized conditions (Michael *et al.,* 2008).

BOD_5 measured using the Azide Modification of the Winkler Method measuring the DO initial and DO final (after five days kept at 20ºC). Preparation of dilution water was done with

two liters volume of water in a suitable bottle and 1ml each of these reagents (phosphate buffer, $MgSO_4$, $CaCl_2$, and $FeCl_3$ solutions/l) were added on water. From the prepared solution, 299 ml of samples sampled with 1ml sample added in incubation bottles having capacity of 300 ml and initial dissolved oxygen was measured using dissolved oxygen meter. Incubation for five days at 20ºC done whose initial dissolved oxygen was measured. After five days, final dissolved oxygen measured. Then, BOD_5 was determined using the following formula.

Calculation

$$BOD_5 \text{ (in mg/l)} = (DOi - DOf)\, Df$$

Where;

DOi = initial dissolved oxygen

DOf = final dissolved oxygen

Df = dilution factor

3.4.3. TSS

TSS was measured using gravimetric method and described as follows; first the inserted disk with wrinkled side up in filtration apparatus and applied vacuum and the disk was washed with three successive 20 ml portions of distilled water continue suction to remove all traces of water and discarded washing. Then filter paper removed from filtration apparatus along with the Gooch crucible, and dried in an oven at 103 to 105ºC for 1 hour. Finally, the paper was mass of filter paper measured and the initial weight subtracted.

Calculation:

$$\text{Mg suspended solids/liter} = \frac{(A - B) \times 1000}{\text{ml sample}}$$

Where;

A= Weight of filter + dried residue, mg

B= Weight of filter, mg

3.4.4. TDS

The weight of filter paper measured. Well-mixed sample of was 20 ml volume was filtered through glass-fiber-filter, the washed with three successive 10 ml volumes of distilled water. Complete drainage allowed between washings; and continues suction for about three minutes after filtration is completed. The filter paper transferred and weighed with evaporating dish and evaporated to dry on a steam bath. The evaporating dish dried for 1 hour in an oven at 103-105ºC, then cooled in desiccators to balance temperature, and weighed.

Calculation:

$$\text{Mg dissolved solids/liter} = \frac{(A - B) \times 1000}{\text{ml sample}}$$

Where:

A= weight of dried residue + dish, mg, and

B= weight of dish, mg

3.4.5. Nitrate

Nitrate measured using the Phenoldisulfonic Acid Method by measuring the nitrate nitrogen. The value of NO_3–N changed to NO_3. The value read from the spectrophotometer after calibration curve done.

Calculation:

$$\text{Mg/liter NO3} = \frac{\mu\text{g NO3} - \text{N} \times 4.427}{\text{ml sample}}$$

3.4.6. Ammonia

Ammonia analyzed through the Direct Nesslerization Method by measuring the ammonia nitrogen. The values of NH_3-N changed to NH_3. The value read from the spectrophotometer after calibration curve done.

Calculation:

$$\text{Mg/liter NH3} = \frac{\mu\text{g NH3} - \text{N X 1.214}}{\text{ml sample}}$$

3.4.7. Phosphate

Phosphate analyzed by the Stannous Chloride Method. The values PO_4 were read using the spectrophotometer. The value read from the spectrophotometer after calibration curve done.

Calculation:

$$\text{Mg/liter PO4} = \frac{\mu\text{g PO4}}{\text{ml sample}}$$

3.5. Statistical Analysis

Data pertaining to physicochemical characteristics of wastewater from four points of processing plant and the EMAS treated wastewater were analyzed using SAS 9.2 computer software (SAS Institute Inc., 1999) after the data were checked for meeting the various ANOVA assumptions (Montgomery, 2005). Whenever, the variation among means happened to be significant, mean separation made using the Least Significant Difference (LSD). The model used for laboratory experimental analysis where, two factor analysis of variance was:

$\gamma_{ij}=\mu+\alpha_i+\beta_j+(\alpha\beta)_{ij}+\varepsilon_{ijk}$

Where,

i= 0ml, 2ml, 4ml, 6ml and 8ml for $EMAS_1$ and 0ml, 3ml, 6ml, 9ml and 12ml for $EMSA_2$

j= Omobeko, Bulbulo, Gembe and Geruke

μ = is the overall mean effect,

α_i = is the effect of the i^{th} level of factor A (concentration level),

β_j = is the effect of the j^{th} of factor B (processing sites or location)

$(\tau\beta)_{ij}$ = is the effect of the interaction between τ_i and β_j

ε_{ijk} = is a random error component.

LSD procedures at 0.05 probability level of significance used to determine differences between treatment means whenever the treatment effects found to be significant. The mean separation did using SAS 9.2 and MSTAT computer software for mean separation. The correlation among the physicochemical characteristics did using SAS 9.2.

RESULTS AND DISCUSSION

The present study tried to address issues of coffee processing wastewater from three perspectives. The physicochemical properties of wastewater from different processing stations and efficacy of different formulations of effective microorganisms as a means to treat processing wastewater did. Therefore, the results of the study interpreted and discussed under this particular chapter.

4.1. Physicochemical Characteristics

The pulping wastewater was highly polluted for all locations and the value was greater than the permissible limit by the WHO (1995) and EEPA (2003) (Table 2). This makes difficult for the discharge of wastewater to the nearby river. The comparison of water from the four points revealed a highly significant difference among points.

As per the maximum permissible limit for discharge, the wastewater from the fermentation tank and pulping water were by far higher than for the discharge and makes it impossible to use it as irrigation water. In agreement with this study, Amelayehu and Devi (2008) indicated that the water down streams of the river was highly polluted due to the discharge of wastewater without treatment especially during the peak processing season. Table 3 depicts the range of physicochemical property of four points analyzed from the four locations. The upper stream water for almost all parameters was within the acceptable limit however, after processing the downstream river water showed an increment in pollution load exceeded the maximum permissible limit.

The mean square values of physicochemical parameters of the water from the four points of processing plant varied highly and significantly (Table 4, 5, 6 and 7). Most physicochemical values of wastewater were observed to be much highest than the WHO permissible limit for irrigation water. Highest mean values of BOD_5 (12150 mg/l) and NO_3 (25.89 mg/l) were observed in Geruke while the highest mean TSS (1811 mg/l), TDS (1602 mg/l), turbidity (647.5NTU) and electrical conductivity (568.5 µS/cm) were recorded in Omobeko. The high

temperature 24.5ºC and lowest pH 2.95 recorded at Bulbulo (Table 2). The present study was in agreement with the findings of Mahana *et al.* (2011).

Table 2 Mean physicochemical properties of pulping wastewater of four wet coffee processing plant along with permissible limits for the discharge of treated effluent WHO (1995) to irrigation channel and EEPA (2003) from industrial effluent.

Parameters	WHO (MPL)	EEPA (MPL)	OB	BU	GM	GR
Temperature (◦C)	20	-	23.45	24.5	22.6	22.9
pH	6.5–8.5	6-9	3.37	2.95	3.99	3.21
BOD_5 (mg/l)	100	80	8220	11633	9413	12150
TDS (mg/l)	450	300	1602	1442.5	1128	1244
TSS (mg/l)	200	100	1811	1240	1330	1737
PO_4 (mg/l)	5	5	4.85	3.99	4.56	4.51
NO_3 (mg/l)	5	5	21.49	24.87	24.04	25.89
NH_3 (mg/l)	5	5	11.1	11.33	9.11	9.84
Turbidity (NTU)	5–10	-	647.5	539.25	381	422.75
EC (µS/cm)	100	-	568.5	483.75	445.5	398.5
DO mg/l	2	-	0.16	0.14.	0.33	0.02

OB=Omobeko; BU=Bulbulo; GM=Gembe; GR = Geruke; WHO = World Health Organization; EEPA = Ethiopian Environmental Protection Authority; MPL = Maximum Permissible Limit; NTU = nephlometric turbidity unit; TSS=Total suspended solid; TDS=Total Dissolved Solid; BOD=Biochemical oxygen demand; DO = Dissolved Oxygen; EC = Electrical Conductivity; NO_3 = nitrate; NH_3 = ammonia and PO_4 = phosphate.

The result pertaining to BOD_5 of four sampling points indicates that there was a highly significant variation among different sampling points in the same location ($P<0.01$). The mean BOD_5 levels of wastewater varied from 48 mg/l in Gembe upper stream respectively to 12150 mg/l Geruke followed by Bulbulo 11633.5 mg/l pulping effluent. The occurrence of high BOD_5 value is due to the availability of degradable organic material in the wet coffee processing wastewater. This finding is in agreement with that of Alemayehu and Devi (2008) who reported the highest BOD as 14,200mg/l. BOD_5 value recorded in the polluted river downstream from conventional wet coffee processing industries was 10,604 mg/l (Tamrat *et al.*, 2006). There was high load of pollution in the wastewater from the fermentation tank,

which was 5433 mg/l in Bulbulo to 7613.7 mg/l in Geruke. These BOD_5 values were greater than the permissible limits set for discharge water by EEPA (2003) and irrigation water by WHO (1995). Jan (2008) also reported that effluents from wet coffee processing have a BOD of 8,000 to 20,000 mg/l.

Table 3 Four location mean minimum and maximum physicochemical property values of upper stream river, pulping water, fermentation tank and downstream river with WHO and EEPA maximum permissible limit

Parameters	WHO MPL	EEPA MPL	Upper stream		Pulping		Fermentation		Downstream	
			Min	Max	Min	Max	Min	Max	Min	Max
Temp. (ºC)	20	-	18	20.5	22.6	24.5	22.3	23.3	19	22
pH	6.5-8.5	6-9	7.14	7.75	2.95	4.3	4.46	5.36	5.22	6.62
BOD_5 (mg/l)	100	80	48.2	167	8220	12150	5433	7613.7	450	1200
TDS (mg/l)	450	300	89	199	1244	1690	996	1280	262	644
TSS (mg/l)	200	100	125	170	1240	1811	1063	1293	335	720
PO_4 (mg/l)	5	5	0.17	1.02	3.98	4.87	2.07	3.82	1.03	1.49
NO_3 (mg/l)	5	5	2.78	5.48	21.6	25.89	17.96	20	9.43	12.2
NH_3 (mg/l)	5	5	1.57	3.22	9.11	11.33	6.05	9.43	4.08	6.54
Turb. (NTU)	5–10	-	11.6	27.4	378	653.3	146.7	186	48.2	122
EC (µS/cm)	100	-	85	120	392.3	548	341	445	201	280
DO mg/l	2	-	4.07	6.96	0.02	0.15	0.12	0.62	1.38	3.99

WHO = World Health Organization; EEPA = Ethiopian Environmental Protection Authority. MPL = Maximum Permissible Limit; TSS = Total suspended solid; TDS = Total Dissolved Solid; BOD = Biochemical oxygen demand; DO = Dissolved Oxygen; EC = Electrical Conductivity and NTU = Nephelometric turbidity unit

The BOD_5 for values were not the same for all location of the same sampling points. As indicate among upper streams ranged from 48.2 mg/l at Gembe to 167 mg/l at Geruke; among downstream it varied between 450 mg/l at Gembe and 1200 mg/l at Geruke. The wastewater from fermentation tanks were 5100 mg/l from Bulbulo to 7613.67 mg/l from Geruke and among pulping effluent 8220mg/l from Bulbulo to 12150 mg/l from Geruke (Table 3).

The mean upper stream BOD_5 values for the water taken from each location (processing plant) were 71.13, 48.2, 167 and 127 mg/l for Omobeko, Gembe, Bulbulo Geruke upper streams, respectively. The mean downstream values of BOD_5 were higher than the upper streams 751, 1200, 450 and 458.67 mg/l for Geruke, Bulbulo, Gembe and Omobeko down streams, respectively). Yared *et al.,* (2010) reported that the BOD_5 value for downstream of some rivers was 1770 mg/l. Though the processing station had constructed the lagoon for wastewater retention, high BOD_5 value indicates that there was leakage from the lagoon. The high BOD_5 registered for fermentation tank and pulping effluents indicate that the water from both places was highly polluted than the river upper and down streams water due to the organic matter. There was a difference in respect of BOD_5 values among the processing sites of the same place.

Alemayehu and Devi (2008) reported BOD values up to 7800 mg/l for river water and from 14, 200 to 10,800mg/l for effluent at Bilida area of Jimma zone. Moreover, BOD values as high as 20,000 mg/l from Vietnam (Enden and Calvert, 2002), 8000 to 11,500 mg/l from Hawaii, USA (Hue *et al*., 2006), and 2000-9000 mg/l from Kenya (Mburu *et al*., 1994) were reported. Since organic matter is the main pollution source in coffee wastewater, it is common to find high biodegradable pollutants from coffee wastewater.

In general, the values of BOD_5 observed at all sites, except that of the upper stream and downstream, were higher which according to Enden and Calvert (2002) reduced to less than 200 mg/l before its entrance to natural waterways. The very high BOD content mainly related to organic enrichment from the coffee processing effluent (Adams and Dougan, 1987; Annon, 1991; Enden and Calvert, 2002; Alemayehu and Devi, 2007; Solomon *et al.*, 2008; Yared *et al.*, 2010) and its effect much more pronounced due to the pulping wastewater from Geruke.

A significant variation also observed among the sampling points for DO values. The DO content of all samples from upper streams was greater than minimum permissible limit (2 mg/l) by WHO and EEPA (Table 3). Whereas there was a lower, DO at downstream 1.38 mg/l from Geruke and 1.67 mg/l from Bulbulo. Much more low DO values observed on raw effluent from the fermentation tank and pulping 0.02 mg/l from pulping and 0.12 mg/l from

fermentation of Geruke. There was a highly significant variation of DO value among different locations ($P<0.01$). There was a drastic change in DO at the downstream (0 mg/l) (Yared *et al.*, 2010). The depletion of oxygen in wastewater was due to the high amount of BOD_5. At Geruke the lowest DO (0.02mg/l) was recorded with the high BOD_5 (12150mg/l) measurement. Solomon *et al.*, (2008), JARC (2008) and Tsigereda (2011) reported similar DO values for raw wastewater.

High DO values recorded at the upper and downstream of all locations. These high DO values might relate to the discharge of the rivers that have high potential to dilute the organic wastewater entered into the river and a continuous aeration along its way to downstream points. According to Giller and Malmqvist (1998), flow velocity directly affects the amount of dissolved oxygen in river water. Therefore, fast flowing waters favor diffusion of atmospheric oxygen to the river water, thereby raises the amount of dissolved oxygen than slow backwaters. However, the very high BOD content was mainly related to organic enrichment from the coffee processing effluent (Enden and Calvert, 2002) and its effect was much more pronounced at the effluents from the pulping and fermentation tank of Geruke and Bulbulo.

The DO recorded for the wastewater from pulping and fermentation tank was very low. This is probably due to the depletion of DO. In alignment with findings of the present study, Abebe *et al*. (2012) stated that during peak coffee-processing season, the disposed untreated coffee waste consumed DO as result of high decomposition, which created anoxic condition and curtailed nitrification. During off-season, oxygen started to recuperate and augmented nitrification.

The highly significant difference of pH value observed among four points of the same site each location (Table 3). The pH values varied from 7.75 at the upper stream of Gembe to 2.95 at Bulbulo pulping effluent. The pH value for pulping and fermentation tank wastewater was continue to be acidic in all location with the lowest pH 2.95 on Bulbulo pulping to 5.36 at Gembe fermentation tank (Table 4, 5, 6 and 7). The downstream of the river were also acidic ranging 5.22 at Geruke to and slightly acidic 6.62 at Omobeko. In agreement with result of the present study, Alemayehu and Devi (2008) reported a pH of 3.57. The lowest pH observed

from pulping and fermentation tank wastewater was due to the biodegradable organic matter into organic acid and other chemicals. The low pH or acidic nature of wet coffee processing wastewater (Enden and Calvert, 2002) disturbs the chemical ecology of receiving water (Tomar, 1999).

Regarding temperature, variations observed among the four sampling points with slow increments. This was due to wastewater from the pulping and fermentation tank. Temperature of the water had been significantly different among the sampling points though the temperature between the upper and downstream did not significantly differ for all location. However, there was a significant variation between fermentation tank and pulping wastewater. The study by Yared *et al.* (2010), Solomon *et al.* (2008), and Alemayehu and Devi (2008). The highest temperature (24.5ºC) was registered from Bulbulo pulping wastewater and followed by pulping effluent of Omobeko (23.3 ºC) wastewater which might be due to the biochemical reaction as a result of degradation of freely available organic matter.

The measured EC (electric conductivity) values had a highly significant variation among four points with the highest EC values being from pulping and fermentation tank effluent. The values ranged from 548.5µS/cm for Omobeko pulping effluent to lowest 85.5 µS/cm and 86.53 µS/cm for Gembe and Bulbulo upper stream water, respectively (Table 4, 5, and 6). Similar studies (JARC and EIARC, 2007) also revealed that mean concentration of conductivity 800 µS/cm and mean concentration of DO 4.49 mg/l as common (Salvamurugan *et al*., 2010).

There was a significant variation in turbidity of the samples from different places with the highest (653.5 NTU) being from Omobeko pulping effluent and lowest (11.65 NTU) also from Omobeko (Table 3). However, the whole value of turbidity was greater than five NTU (which is higher than permissible limit for irrigation water). The turbidity was higher due to organic particles, and dissolved oxygen depletion, which may occur in the water body. The excess nutrients available will encourage microbial breakdown, a process that requires dissolved oxygen.

TSS and TDS values indicated the variation among four points with the high mean values of 1811 mg/l TSS and 1694 mg/l TDS from Omobeko pulping effluent and lower at 125 mg/l TSS at Geruke upper and TDS 89 mg/l at Omobeko upper (Table 4 and 7). The high TSS and TDS in pulping and fermentation tank were due to availability of high organic matter and nutrients from the pulping and washing process of wet coffee processing. The TSS 5870-2080 mg/l reported by Alemayehu and Devi was higher than this study however, Tsigereda (2011) reported the similar result TSS of 1564 –2310mg/l and TDS of 1580 –2133mg/l.

The inorganic ions NO_3, NH_3 and PO_4 physicochemical parameters also depicted highly significant variations among the points where the wastewater taken. Higher values observed at the wastewater from the pulping and fermentation tank. Though the values at down streams were lower than the pulping and fermentation tank, they were clearly higher than the upper streams. The highest values for NO_3 (25.89 mg/l), NH_3 (11.33 mg/l) were from Geruke pulping wastewater and for PO_4 (4.54 mg/l) from Gembe pulping effluent (Table 7 and 4). Whereas the lower value of NO_3 4.08 mg/l from Omobeko and 2.78 mg/l from Gembe of upper stream recorded (Table 4 and 5). The high amount of nutrients (NO_3, NH_3 and PO_4) observed in the wastewater from pulping and fermentation was due to the high amount of nutrients set free and high organic matter load in wastewaters. A decrease in nitrogenous pollutants in upper and downstream rivers may be associated with flow rate and dilution effects of the river water.

Table 4 Mean comparison for the physicochemical characteristics of Gembe sampling points

Points of wastewater sampling	TSS (mg/l)	TDS (mg/l)	DO (mg/l)	Temp (ºC)	pH	Turb. (NTU)	EC (µS/cm)	NO_3 (mg/l)	NH_3 (mg/l)	PO_4 mg/l)	BOD_5 (mg/l)
Upper	125^{c}	199^{d}	6.96^{a}	18.57^{c}	7.75^{a}	14.45^{d}	85.5^{c}	2.78^{d}	1.57^{d}	0.17^{d}	48.2^{d}
Pulping effluent	1330.4^{a}	1320^{a}	0.14^{d}	22.7^{a}	4.30^{d}	378.0^{a}	465.3^{a}	24.1^{a}	9.11^{a}	4.54^{a}	9413.3^{a}
Fermentation Effluent	1278.8^{a}	1280.7^{b}	0.62^{c}	22.6^{a}	5.30^{c}	146.7^{b}	445.6^{a}	19.42^{b}	6.05^{b}	3.82^{b}	7600^{b}
Down	445.67^{b}	550.7^{c}	2.98^{b}	20.37^{b}	6.23^{b}	48.22^{c}	280^{b}	12.19^{c}	4.58^{c}	1.46^{c}	450^{c}
LSD	81.6	28.4	0.24	0.507	0.201	7.83	24.247	1.187	0.31	0.09	477
SE (±)	43.33	15.06	0.13	0.27	0.11	4.16	12.88	0.63	0.16	0.05	253.4
CV (%)	6.10	2.02	4.08	1.28	1.75	2.96	4.65	5.00	4.03	2.26	5.88

LSD = least significant difference; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia and PO4 = phosphate,
Mean values within a column labeled with same letter(s) are not significantly different.

Table 5 Mean comparison for the physicochemical characteristics of Omobeko sampling points

Points of wastewater sampling	TSS (mg/l)	TDS (mg/l)	DO (mg/l)	Temp (^{0}C)	pH	Turb. (NTU)	EC (µS/cm)	NO_3 (mg/l)	NH_3 (mg/l)	PO_4 mg/l)	BOD_5 (mg/l)
Upper	131^{d}	89.00^{c}	5.80^{a}	18.50^{b}	7.43^{a}	11.61^{d}	99.60^{c}	4.08^{d}	2.23^{d}	0.93^{d}	71.10^{d}
Pulping effluent	1811^{a}	1694.00^{a}	0.15^{d}	23.43^{a}	3.37^{d}	653.3^{a}	548.00^{a}	21.6^{a}	11.10^{a}	4.87^{a}	8220.00^{a}
Fermentation effluent	1166^{b}	996.00^{b}	0.39^{c}	23.3^{a}	5.05^{c}	149.43^{b}	341.03^{b}	17.96^{b}	8.52^{b}	2.07^{b}	5936.70^{b}
Down	335^{c}	262.70^{c}	3.99^{b}	19.00^{b}	6.62^{b}	56.86^{c}	201.30^{c}	9.43^{c}	6.51^{c}	1.49^{c}	458.70^{c}
LSD	68.79	78.63	0.18	0.88	0.23	19.39	7.95	0.48	0.32	0.09	240.67
SE (±)	36.54	41.76	0.09	0.465	0.12	10.30	4.22	0.26	0.17	0.05	127.82
CV (%)	4.37	5.68	3.65	2.21	2.17	5.57	1.90	2.42	4.11	2.67	3.48

LSD = least significant difference; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia and PO_4 = phosphate.
Mean values within a column labeled with same letter(s) are not significantly different.

Table 6 Mean comparison for the physicochemical characteristics of Bulbulo sampling points

Points of wastewater sampling	TSS (mg/l)	TDS (mg/l)	DO (mg/l)	Temp (ºC)	pH	Turb. (NTU)	EC (µS/cm)	NO_3 (mg/l)	NH_3 (mg/l)	PO_4 mg/l)	BOD_5 (mg/l)
Upper	142.00[d]	133.3[d]	4.67[a]	18.0[d]	7.46[a]	12.60[d]	86.53[d]	5.36[d]	2.32[d]	0.33[d]	127.00[d]
Pulping effluent	1240.7[a]	1440[a]	0.04[d]	24.5[a]	2.95[d]	537.30[a]	463.30[a]	24.91[a]	9.84[a]	3.98[a]	11633.3[a]
Fermentation effluent	1063.0[b]	1200.0[b]	0.58[c]	22.3[b]	4.46[c]	180.38[b]	365.00[b]	20.00[b]	7.64[b]	3.07[b]	5433.30[b]
Down	680.30[c]	520.0[c]	1.67[b]	19.0[c]	5.61[b]	65.99[c]	200.99[c]	11.94[c]	6.54[c]	1.03[c]	751.67[c]
LSD	53.82	20.02	0.71	0.71	0.27	7.47	14.27	0.43	0.46	0.12	428.13
SE +/-	28.58	10.63	0.38	0.38	0.14	3.97	7.58	0.23	0.24	0.06	227.39
CV	4.45	1.43	15.1	1.81	2.63	2.50	3.72	1.89	6.29	3.47	5.12

LSD = least significant difference; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia and PO_4 = phosphate.
Mean values within a column labeled with same letter(s) are not significantly different.

Table 7 Mean comparison for the physicochemical characteristics of Geruke sampling points

Points of wastewater sampling	TSS (mg/l)	TDS (mg/l)	DO (mg/l)	Temp (^{0}C)	pH	Turb. (NTU)	EC (µS/cm)	NO_3 (mg/l)	NH_3 (mg/l)	PO_4 mg/l)	BOD_5 (mg/l)
Upper	170^{d}	150.00^{d}	4.07^{a}	20.53^{c}	7.14^{a}	27.44^{d}	120.97^{c}	5.48^{d}	3.22^{d}	1.02^{d}	167.3^{d}
Pulping effluent	1736.0^{a}	1244.30^{a}	0.02^{c}	23.03^{a}	3.21^{d}	424.67^{a}	392.30^{a}	25.89^{a}	11.33^{a}	4.51^{a}	12150^{a}
Fermentation effluent	1293.3^{b}	1095.0^{b}	0.12^{c}	22.03^{b}	4.48^{c}	155.27^{b}	372.00^{b}	18.48^{b}	9.19^{b}	2.65^{b}	7613^{b}
Down	720.33^{c}	644^{c}	1.38^{b}	21.36^{b}	5.22^{b}	112.2^{c}	210.43^{c}	10.91^{c}	6.03^{c}	1.45^{c}	1200^{c}
LSD	73.62	26.79	0.35	0.67	0.13	7.68	12.73	1.07	0.42	0.25	467
SE (±)	39.10	14.23	0.18	0.36	0.07	4.08	6.77	0.57	0.22	0.13	248.07
CV (%)	4.91	2.34	6.94	1.63	1.22	3.16	2.83	4.75	5.08	7.26	4.964

LSD = least significant difference; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia and PO_4 = phosphate.

Mean values within a column labeled with same letter(s) are not significantly different.

In agreement with the results of the present, study Solomon *et al.* (2008) and Yared *et al.* (2010) reported that NO_3 (26.4 mg/l), NH_4^+ (12.6 mg/l). Alemayehu and Devi (2008) also noted accumulation of inorganic ions such as nitrate (23 - 10.5 mg/l), phosphate (7.3 - 4.6 mg/l) in coffee wastewater. Similarly, Tsigereda (2011) also reported nutrient deposition where NH_3 ranged between 8.44 to 17.08 mg/l while the range was between 1.75 and 3.27 for PO_4. Abebe *et al.* (2012) stated that range was 6.1 to 12 mg/l for NO_3 and 5 to 30 mg/l NH_3. However, another study (JARC 2007) put ammonia concentrations in the range of 1.35 to 8.02 mg/l.

4.2. Physicochemical Characteristics of Treated Wastewater with $EMAS_1$ and $EMAS_2$

The wastewater from the pulping, which treated with different concentration levels of EM, and the wastewater from different processing location analyzed. The results showed a significant variation with regard to these physicochemical characteristics. EM technology may help to mitigate water crisis by purifying wastewater by devouring all of its toxins and the stench of solid waste is eliminated almost immediately (Mohammed *et al.,* 2011). From the experiment done by Firdaus (2007) on industrial wastewater treatment using EM, it was learnt that EM was effective in reducing BOD, nitrate and phosphate up to 24.47%, 18.87%, and 80.47%, respectively while increasing the pH by 45.61%.

From the physicochemical analysis of variance of this study the probability values for all physicochemical parameters except for temperature (for one location) had a highly significant difference ($P<0.01$) (Appendix Table 2 and 3). The result of pulping wastewater treatment with different concentration levels of $EMAS_1$ and $EMAS_2$ revealed a highly significant difference among locations.

The wastewater from the pulping treated with EMAS showed a reduction in the value of BOD_5, NO_3, NH_3, PO_4, Turbidity, TSS and TDS and increment in pH and DO. Even though the value of some parameters was not satisfactory, when compared with the permissible limits for irrigation water set by WHO (1995), the reasonable purification capacity of EMAS was demonstrated.

Table 8 Physicochemical characteristics of treated wastewater and the pulping effluent value with permissible limits for discharge.

Parameters	WHO MPL	EEPA MPL	Pulping Effluent		Treated with $EMAS_1$ (4ml)		Treated with $EMAS_2$ (6ml)	
			Min	Max	Min	Max	Min	Max
Temp. (ºC)	20	-	22.6	24.5	22.27	22.57	22.47	22.57
pH	6.5-8.5	6-9	2.94	4.3	5.8	6.06	5.02	5.99
BOD_5 (mg/l)	100	80	8220	12150	790	870	450	646
TDS (mg/l)	450	300	1244	1690	245	404	241	388
TSS (mg/l)	200	100	1240	1811	310	427	311	429
PO_4 (mg/l)	5	5	3.99	4.87	1.28	1.57	3.08	3.67
NO_3 (mg/l)	5	5	21.6	25.89	4.94	5.6	4.77	5.99
NH_3	5	5	9.11	11.33	3.08	3.21	1.46	2.61
Turb. (NTU)	5–10	-	378	653	58	72	46.1	69.8
EC (µS/cm)	100	-	392	548	319	469	273	372.67
DO mg/l	2	-	0.02	0.15	2.1	2.8	2.28	2.94

TSS = total suspended solid; TDS = total dissolved solid; BOD_5 = Biochemical oxygen demand; DO = dissolved oxygen; pH = power of hydrogen; NTU = Nephelometric turbidity unit; Turb. = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3= ammonia and PO_4 = phosphate.

4.2.1. Biochemical oxygen demand

The biological oxygen demand of wet coffee processing treated pulping wastewater with effective microorganism which was activated with 5% molasses, 5% EM_1 and 90% distilled water ($EMAS_1$) showed a highly significant difference both for concentration level and location ($P<0.01$). Likewise, the Biochemical oxygen demand of the pulping effluent treated with effective microorganisms activated with 2.5% EM_1, 2.5% molasses, 5% effluent and 90% distilled water ($EMAS_2$) also resulted in a highly significant change($p<0.01$) (Appendix table 2 and 3).

With regard to the effect of $EMAS_1$, concentration level and location on the BOD_5 of wet coffee processing effluent reveled that there was a highly significant interaction effect on the level of BOD_5 after treatment with $EMAS_1$. Accordingly, treatment coffee processing wastewater from different location with $EMAS_1$ revealed that the lowest BOD_5 achieved by treating the effluent from OB and GR with 4 ml $EMAS_1$. On the other hand, the BOD_5 was remained to be the maximum when the pulping effluent from BU was not treated with any EMAS, which was followed by effluent from same place treated with 8 ml $EMAS_1$ (Table 9).

The reduction in BOD_5 due to EM treatment was not same across all the treatments. The maximum BOD_5 reduction across all location observed, as compared to the control, when the effluent treated with 4ml $EMAS_1$. The level of reduction was 57.8%, 60.5%, 54.3%, and 49.6% for BO, BU, GM and GR locations, respectively. The mean value of wastewater from different locations treated with different concentrations of $EMAS_1$ showed highly significantly difference at $p<0.01$ (Appendix table 5). The concentration level of 2ml and 6ml showed slight reduction in BOD_5 while treatment with 8ml did not show much reduction in BOD_5, for OB and GR the value were higher than the control.

In another experiment, the interaction effect of $EMAS_2$ concentration level and location was similarly highly significant. More specifically, it was observed that the minimum BOD_5 value was recorded when the wet coffee processing wastewater from GM pulping station was treated with 6ml $EMAS_2$ followed by the treatment of effluent of OB with 6 ml $EMAS_2$. To the contrary, the BOD_5 level was remained highest when effluent from BU that was not treated with any $EMAS_2$ followed by BU effluent treated with 12ml $EMAS_2$. The BOD_5 levels were brought down to 555mg/l, 635mg/l, 450mg/l and 646.7mg/l for OB, BU, GM and GR respectively when the effluent were treated with 6ml $EMAS_2$ (Table 10). When expressed in percentage, the reductions achieved due to treatment with 6ml $EMAS_2$, were 71.0%, 71.1%, 75.7%, and 58.7% for OB, BU, GM and GR, respectively.

The use of EMAS for the treatment of wet coffee processing wastewater decreases the amount of BOD_5 because of which the pollutant load of wastewater decreased. The use of EM activated with effluent molasses and EM1 ($EMAS_2$) showed a higher reduction in BOD_5 level

than activation of EM with molasses and EM1 ($EMAS_1$). The level of concentration used had also a highly significant effect. The mean values of the two experiments showed that application of 4ml for activation of first experiment ($EMAS_1$) and 6ml for second experiment ($EMAS_2$) demonstrated a high reduction level (Table 9 and 10).

The achievements observed in the present study due to EM treatment are in coherence with reduction of BOD_5 from an industrial wastewater by 24.47% reported by Firduas (2007). Though the reduction of BOD_5 did not reach to the permissible limit for irrigation water (100mg/l) by WHO (1995), treatment with EM had demonstrated a great capacity to reduce the BOD_5 from the wet coffee processing wastewater. This significant reduction of BOD_5 might be due to digestion of organic compounds, which are biodegradable by the bacteria present in $EMAS_1$ and $EMAS_2$. These beneficial bacteria use organic matter contained in the wastewater as a food or nutrient supply. When the effluent become more concentrated, the BOD_5 increased which might be due to the use of more microorganisms beyond the purification capacity (Nicholas, 1996).

The BOD_5 from the wastewater reduced due to the action of microorganism. These microorganisms tend to collect as a biological flock called biomass and generally possess good settling characteristics. As biological oxidation stabilization of organic matter, proceeds high rate of BOD_5 removed from wastewater upon contact with active biomass BOD_5 utilized in proportion to cell growth. Enzymes of living cells decompose materials that concentrate on the biomass surface; new cells synthesized; decomposition by products washed into the water or escape into the atmosphere. Biological cell material oxidized through endogenous respiration when food supply becomes limited. The biomass converted to settle able material or removable solids (Nicholas 1996). These beneficial bacteria used organic matter contained in the wastewater as a food or nutrient supply (Firduas, 2007).

Table 9 Mean comparison of NO_3, NH_3, PO_4, BOD and DO for concentration by location interaction of wastewater treated with $EMAS_1$

Treatment combination	BOD_5	DO	NO_3	NH_3	PO_4
OB X 0ml L^{-1}	1916.67^{d}	0.78^{j}	6.62^{ef}	5.62^{C}	2.71^{bc}
OB X 2mlL^{-1}	1016.67^{m}	1.85^{d}	5.81^{h}	3.05^{lm}	0.99^{k}
OB X 4ml L^{-1}	808.33^{p}	2.19^{b}	5.10^{jk}	3.17^{jk}	1.28^{j}
OB X 6ml L^{-1}	1200.0^{jk}	1.58^{f}	6.82^{d}	3.35^{hi}	2.17^{f}
OB X 8ml L^{-1}	1950.00^{c}	1.26^{h}	8.81^{a}	4.67^{e}	2.14^{f}
BU X 0ml L^{-1}	2203.33^{a}	0.15^{m}	6.76^{de}	5.45^{d}	2.46^{e}
BU X 2ml L^{-1}	1210.00^{j}	1.20^{h}	5.46^{i}	2.97^{m}	1.61^{g}
BU X 4ml L^{-1}	870.00^{n}	2.10^{c}	4.94^{k}	3.08^{kl}	1.57^{gh}
BU X 6ml L^{-1}	1350.00^{i}	1.44^{g}	6.43^{f}	3.22^{j}	2.48^{de}
BU X 8ml L^{-1}	2106.67^{b}	0.34^{l}	8.81^{a}	4.62^{e}	2.38^{e}
GM X 0ml L^{-1}	1850.00^{e}	0.29^{l}	6.93^{d}	5.92^{a}	2.16^{f}
GM X 2ml L^{-1}	1100.00^{l}	1.61^{f}	6.23^{g}	3.49^{g}	1.51^{ghi}
GM X 4ml L^{-1}	845.00^{o}	2.81^{a}	5.14^{j}	3.21^{j}	1.28^{j}
GM X 6ml L^{-1}	1185.00^{k}	1.27^{h}	6.88^{d}	3.44^{gh}	2.74^{b}
GM X 8ml L^{-1}	1615.00^{g}	0.84^{j}	8.17^{b}	4.65^{e}	3.22^{a}
GR X 0ml L^{-1}	1566.67^{h}	0.46^{k}	7.40^{c}	5.80^{b}	1.47^{hi}
GR X 2ml L^{-1}	1220.00^{j}	1.88^{d}	6.48^{f}	2.85^{n}	1.41^{i}
GR X 4mlL^{-1}	790.00^{p}	2.83^{a}	5.60^{i}	3.08^{kl}	1.28^{j}
GR X 6ml L^{-1}	1110.00^{l}	1.77^{e}	6.76^{de}	3.32^{i}	2.59^{cd}
GR X 8ml L^{-1}	1756.67^{f}	1.11^{i}	8.70^{a}	4.17^{f}	2.12^{f}
S.E±	12.96	0.05	0.12	0.07	0.08
LSD (5%)	21.17	0.08	0.19	0.11	0.13
CV	2.25	3.39	2.4	1.67	3.95

LSD= least significant difference SE = standard error CV= coefficient of variance, NO_3 = nitrate, NH_3 = ammonia PO_4=phosphate, BOD_5=Biochemical oxygen demand, DO=dissolved oxygen.
Means followed by the same letter/s within the column are not statistically different
All value is in mg/l

Table 10 Mean comparison of concentration by location interaction for NO_3, NH_3, PO_4, BOD and DO of wastewater treated with $EMAS_2$

Treatment combination	BOD_5	DO	NO_3	NH_3	PO_4
OB X 0ml L^{-1}	1916.67^{c}	0.78^{h}	6.62^{def}	5.62^{c}	2.71^{d}
OB X 3mlL^{-1}	836.67^{k}	1.93^{c}	6.31^{g}	3.36^{f}	2.04^{g}
OB X 6ml L^{-1}	555.00^{m}	2.97^{a}	4.77^{k}	2.61^{h}	3.09^{c}
OB X 9ml L^{-1}	933.33^{j}	1.65^{e}	3.72^{n}	1.57^{k}	1.29^{k}
OB X 12ml L^{-1}	1203.33^{h}	0.38^{k}	5.90^{h}	3.34^{fg}	1.10^{l}
BU X 0ml L^{-1}	2203.33^{a}	0.15^{m}	6.76^{cd}	5.45^{d}	2.46^{e}
BU X 3ml L^{-1}	1340.00^{f}	2.29^{b}	6.45^{fg}	2.33^{i}	1.91^{h}
BU X 6ml L^{-1}	635.00^{l}	2.98^{a}	5.02^{j}	1.45^{l}	3.11^{c}
BU X 9ml L^{-1}	1250.00^{g}	1.93^{c}	4.37^{l}	0.96^{n}	1.69^{i}
BU X 12 ml L^{-1}	2086.67^{b}	0.30^{l}	6.55^{ef}	3.26^{g}	1.11^{l}
GM X 0ml L^{-1}	1850.00^{d}	0.29^{l}	6.93^{bc}	5.92^{a}	2.16^{f}
GM X 3ml L^{-1}	1043.33^{i}	1.31^{g}	6.70^{de}	2.27^{i}	2.56^{e}
GM X 4ml L^{-1}	450.00^{n}	2.28^{b}	5.53^{i}	1.56^{k}	3.38^{b}
GM X 9ml L^{-1}	1166.67^{h}	1.52^{f}	4.11^{m}	1.05^{m}	1.34^{k}
GM X 12ml L^{-1}	1876.67^{cd}	0.40^{k}	6.57^{def}	3.39^{ef}	1.26^{k}
GR X 0ml L^{-1}	1566.67^{e}	0.46^{j}	7.40^{a}	5.80^{b}	1.47^{j}
GR X 3ml L^{-1}	965.00^{j}	1.74^{d}	6.62^{def}	2.12^{j}	2.03^{g}
GR X 6mlL^{-1}	646.67^{l}	2.95^{a}	5.95^{h}	0.96^{n}	3.67^{a}
GR X 9ml L^{-1}	1050.00^{i}	1.49^{f}	4.04^{m}	1.37^{l}	1.60^{i}
GR X 12ml L^{-1}	1610.00^{e}	0.62^{i}	6.96^{b}	3.46^{e}	1.04^{l}
S.E±	28.38	0.036	0.118	0.0541	0.0731
LSD (5%)	46.37	0.058	0.192	0.088	0.1194
CV	0.94	2.499	2.5	1.87	3.56

LSD= least significant difference SE = standard error CV= coefficient of variance, NO_3 = nitrate, NH_3 = ammonia PO_4=phosphate, BOD_5=Biochemical oxygen demand, DO=dissolved oxygen.
Means followed by the same letter/s within the column are not statistically different
All value is in mg/l

4.2.2. Dissolved oxygen (DO)

The amount of dissolved oxygen in wet coffee processing effluent treated with $EMAS_1$ (Experiment 1) was observed to be highly significant affected by concentration level, location and their interaction ($P<0.01$) (Appendix table 2). Treatment of wet coffee processing wastewater With $EMAS_1$ raised the concentration of DO to the maximum when effluents from GM and GR were treated with 4ml $EMAS_{1.}$ On the other hand, the level of DO was the lowest in the effluent from BU which had not received any EM treatment at all followed by the DO value registered due to the treatments of effluent from BU and GR with 8 and 0 ml EMAS1, respectively.

The amount of dissolved oxygen in wet coffee processing effluent treated with it observed that from the mean dissolved oxygen value, the application of 6ml $EMAS_2$ had a highest increment than other concentration levels. The interaction effect of location with concentration level on DO value was highly significant ($P<0.01$) (Appendix table 3). The highest increments for the different locations were 2.97, 2.98, 2.28 and 2.95 mg/l for OB, BU, GM and GR respectively when effluents treated with 6ml $EMAS_2$ (Table 10). However, further increase in the concentration levels of $EMAS_2$ resulted in the decrease of DO value.

Concisely, the increments in DO observed (Table 9 and 10) for the treated wastewater with $EMAS_1$ and $EMAS_2$, respectively. The highest DO values observed when the wastewater treated with 6 ml $EMAS_2$ than 4ml $EMAS_1$. The use of $EMAS_2$ was cost effective due to the decrease in the use of EM1 and molasses for activation by 33.3%. The mean square value showed that treated wastewater for both experiment were significantly different (Appendix table 4 and 5). The significant increment in the concentration of DO is due to the biodegradable organic matters, which used by microorganisms as a nutrient or food, as the demand for oxygen decreased this lead to availability of dissolved oxygen in the wastewater. The oxygen from the atmosphere also may contribute for the increment in treated wastewater (Nicholas, 1996). The DO value obtained after treatment almost met the permissible limit set by WHO (1995) for irrigation water.

4.2.3. Nitrate (NO_3)

The amount of nitrate in the wastewater after treatment with $EMAS_1$ showed a highly significant difference ($P<0.01$) (Appendix table 2) for the interaction effect of concentration level by location. Similar to this the pulping effluent treated with $EMAS_2$ also resulted in a highly significant change ($p<0.01$) (Appendix table 3).

Hence the wet coffee processing wastewater from the four locations treated with $EMAS_1$ demonstrated that there was highly significant reduction in the amount of NO_3 from effluent treated with 4ml of $EMAS_1$ was 22.9%, 26.9%, 25.8 % and 24.3% compared to the control obtained for OB, BU, GM and GR wastewater, respectively (Table 9). Nevertheless, as the $EMAS_1$ concentration level increased beyond 4ml, a significant increment of nitrate value observed. Whereas the nitrate value remained maximum for effluent treated with 8ml and the control.

A similar scenario was observed for nitrate value in experiment wherein wastewater was treated with $EMAS_2$ and resulted in a highly significant ($P<0.01$) change. Highest reduction of nitrate was achieved when 9 ml of $EMAS_2$ was used for all location of wastewater when compared to other concentration levels. But the extent of reduction achieved with at 6ml $EMAS_2$ was also observed to meet the permissible limit of WHO and EEPA. The nitrate levels were brought down to 4.77 mg/l, 5.02 mg/l, 5.53 mg/l and 5.59 mg/l for OB, BU, GM and GR, respectively (Table 10). When expressed in percentage, the reductions achieved due to treatment with 6ml $EMAS_2$, were 43.8%, 35.4%, 40.7% and 45.4% for OB, BU, GM and GR, respectively. More specifically, it was observed that the minimum nitrate value was recorded when the wet coffee processing wastewater from OB pulping station was treated with 6ml $EMAS_2$ followed by the treatment of effluent of BU with 6 ml $EMAS_2$ (Table 10).

This study demonstrated that the use of EMAS to treat wet coffee processing wastewater decreases the amount of nitrate as a result; the pollution load of wastewater decreased. The use of $EMAS_2$ showed the maximum decrease than the use of $EMAS_1$. The mean value of the two experiments showed that the application of 4ml for activation of first experiment and 6ml

for second experiment demonstrated a high reduction level (Table 8). This value met the permissible limit for irrigation water (5mg/l) (WHO 1995).

The observed reduction in the nitrate content of wastewater due to treatment with EMAS could attributed to the microbial utilization of these nutrients, thereby making the water suitable for recycling for agricultural purposes (Mohana *et al.,* 2011). This study was in agreement with the result of a research done on industrial wastewater by Firduas (2007) with the reduction of 18.8% of NO_3. Rashid and West (2007) also report the similar result with this finding reduction of NO_3 by 43% from industrial wastewater and with the research done by Okuda and Higa (2008) who reported that EM application decreased the nitrate level by favoring the nitrification and denitrification reaction.

4.2.4. Ammonia (NH_3)

Ammonia is one of the physicochemical characteristics to determine the pollution load of wastewater. Therefore, the amount of ammonia in the treated effluent with effective microorganism to reduce the pollution load observed. The use of $EMAS_1$ to treat the wet coffee processing wastewater showed a highly significant change ($P<0.01$) for the interaction (Appendix table 2). Treatment with $EMAS_1$ decreased the amount of ammonia in wet coffee processing wastewater. The minimum value of ammonia obtained from application of 2 ml $EMAS_1$ was 2.85 and 2.97 mg/l, which observed in the effluents of GR and BU respectively (Table 9). On the other hand, the level of NH_3 remained to be the highest in the effluent for all locations, which had not received any EM treatment at all followed by the NH_3value registered due to the treatment of effluent with 8ml $EMAS_1$(Table 9).

The high decrement in NH_3 concentration of the effluent from OB, BU, and GR was 3.05, 2.97 and 2.85 mg/l respectively when treated with 2 ml $EMAS_1$ and 3.32 mg/l for GM treated with 4ml $EMAS_1$ (Table 9). The higher concentration (the low reduction) observe from the effluent of GM (5.92 mg/l) when the effluent which receive no EM treatment (Table 9). The percentage of reduction for effluents from OB, BU, GM and GR were 43.5%, 43.5%, 45.7% and 46.8% when treated with 4 ml $EMAS_1$ as compared to the 0 ml applied effluent (Table 9).

Pulping wastewater treated with $EMAS_2$ had highly significant difference for the interaction effect concentration level by location at $P<0.01$ for NH_3 (Appendix table 3). The highest reduction value observed from the effluent of all location when treated with 9 ml of $EMAS_2$ except for GR, which show lowest, value at 6 ml $EMAS_2$ application (Table 10). More specifically, it observed that the minimum NH_3 value was recorded when the wet coffee processing wastewater from OB (1.57 mg/l), BU (0.96 mg/l), and GM (1.05 mg/l) pulping station treated with 9 ml $EMAS_2$. The treatment of pulping effluent from OB (2.61 mg/l), BU (1.45 mg/l), GM (1.56 mg/l) and GR (0.96 mg/l) also when treated with 6ml $EMAS_2$. To the contrary, the NH_3 level remained highest when effluent from GM (5.9 mg/l) and GR (5.8 mg.l) was not treated with any $EMAS_2$ followed by OB (5.45 mg/l) and BU (5.64mg/l). When expressed in percentage, the reductions achieved due to treatment with, 72.0%, 82.4% and 82.3% for OB, BU and GM effluent when 9 ml $EMAS_2$ used and 83.4% for GR effluent treated with 6 ml $EMAS_2$ (Table 10).

As depicted this study the use of EMAS for the treatment of wet coffee processing wastewater decreases the amount of NH_3 as a result of which the pollutant load of wastewater decreased. The use of EM activated with effluent molasses and EM1 ($EMAS_2$) showed a higher reduction in level of NH_3 than activation of EM with molasses and EM1 ($EMAS_1$). The level of concentration used had also a highly significant effect. The mean values of the two experiments showed that application of 2 ml and 4 ml for activation of first experiment ($EMAS_1$) and 9 ml and 6 ml for second experiment ($EMAS_2$) demonstrated a high reduction level. The use of $EMAS_2$ had a great reduction capacity and cost effective than $EMAS_1$.

The achievement in both treatment are below the maximum permissible limit for WHO irrigation and EEPA discharge standards so for to the near by river. Since the NH_3 concentration was lower than the permissible limit where the effluent treated with 6 ml and 9 ml of $EMAS_2$. The use of 6ml $EMAS_2$ is cost effective. The similar report was released on the reduction of ammonia by using EM by Firdues (2007) with 48% and Rashid and West (2007) with 44% reduction. The reduction of ammonia in wastewater was due to the

nitrification and denitrification process and the consumption of nitrogen by the microorganism (Nicholas 1996).

4.2.5. Phosphate (PO_4)

Even though the amounts of phosphate in wet coffee, processing wastewater was lower than that of nitrate and ammonia, there was still reduction when treated with activated EM. The extent of reduction in phosphate load of the wastewater depended on the level of EM concentration and location of wastewater, both of which bearing a highly significant variation due to their interaction effect ($P < 0.01$) (Appendix Table 2 and 3). With regard to the effect of $EMAS_1$ concentration level and location on the PO_4 of wet coffee processing effluent, it was apparent that there was a highly significant interaction effect on the level of PO_4 after treatment with $EMAS_1$. Accordingly, treatment of coffee processing wastewater from different locations with $EMAS_1$ depicted that the lowest PO_4 achieved by treating the effluent from OB (0.99 mg/l) with 2 ml. On the other hand, the PO_4 remained to be the maximum when the pulping effluent from GM (3.22 mg/l) which was treated with 8ml of $EMAS_1$ followed by OB (2.72 mg/l) and BU (2.48 mg/l) which was not treated with any EMAS (Table 9).

At concentration level, 2 ml and 4 ml when treated with $EMAS_1$ the reduction were high when compared to the control and other level but reduction in PO_4 due to EM treatment was not same across all the treatments. At the application of 2 ml $EMAS_1$ on wet coffee processing wastewater the high reduction were observed (Table 8) for all location with the mean values of PO_4 0.99, 1.61, 1.35 and 1.33 mg/l for OB, BU, GM and GR respectively followed by 4ml $EMAS_1$ 1.28 mg/l for OB, BU, and GR and 1.57 mg/l for GM. However, when the concentration level increased the amount of PO_4 also increment observed when compared to the control. The percentage reductions from 2ml $EMAS_1$ application were 63.47%, 34%, 21.96 and 6.34% for OB, BU, GM and GR respectively with highest reduction of PO_4 from Omobeko wastewater (Table 9).

In another experiment, the interaction effect of $EMAS_2$ concentration level and location was similarly highly significant. The application of activated $EMAS_2$ on wastewater of different location had an effect on phosphate concentration. The PO_4 result obtained after treatment showed a highly significant difference at $P<0.01$ (Appendix table 3). The highest decrement of PO_4 in pulping wastewater were 1.04, 1.10, 1.11 and 1.26 mg/l for GR, OB, BU and GM respectively (Table 10) but this value by far lower than the permissible limit for irrigation. Unlike other parameters wastewater PO_4 value show the highest reduction when the application of 12ml of $EMAS_2$ with the percentage of decrement for these treatment were 59.6%, 54.9%, 41.6% and 28.9% for OB, BU, GM and GR respectively. This reduction parentage similar with study by Firdaus (2007) the application of EM on industrial west water decrease the PO_4 value by 80.47%. The wastewater from OB, BU, GM and GR treated with 6 ml $EMAS_2$ was revealed relatively the good value which were 3.09, 3.11, 3.38 and 3.38 mg/l (Table 10) for the irrigation standard by WHO (5 mg/l) .

The use of EMAS had a great potential economically, application of $EMAS_1$ was more visible because at the application of 2 ml the high reduction observed. The reduction of phosphate at the application of EM were due to the some heterotrophic bacteria such as poly-phosphate microbes were able to remove solubilized phosphate by accumulating them intracellularly in the form of polyphosphate (Nicholas, 1996).

4.2.6. Temperature

The temperature of wastewater from all locations showed a slight decrease due to treatment with different concentrations of EM. The interaction effect between location and concentration level of EM on temperature for treated effluent was found to be significant ($P<0.05$) (Appendix table 2). Accordingly, treatment with 4 ml activated $EMAS_1$ resulted in the lowest observed temperature (Table 11).

The temperature of wastewater treated with activated $EMAS_2$ in the second experiment was significantly different among treatments at $P<0.05$ (Appendix table 3). The least significant

difference at 5% was 0.11 among concentration level and 0.099 among locations (processing locations) (Table 11 and 12).

4.2.7. Electric conductivity (EC)

With regard to the mean value of EC for effluents treated with $EMAS_1$ (activated EM with molasses, EM1 and distilled water), the ANOVA showed a highly significant ($P<0.01$) interaction effect of concentration level EM and location (Appendix table 2). The maximum reductions value of 310.7, 379.3, 366.0 µS/cm from OB, BU and GM respectively were observed when 4ml $EMAS_1$ and 349 µS/cm from 6 ml of $EMAS_1$ (Table 11).

The reduction of electrical conductivity from the second experiment observed with the application of 6 ml and 3ml activated with $EMAS_2$ for OB and BU where as at 6 ml for GM and GR. From the ANOVA (Appendix table 3) the electrical conductivity was significantly different ($P<0.01$) among concentration levels and locations. The reductions in the EC of wastewater treated with $EMAS_2$ were 56.3%, 62.8%, 33.2% and 41% for OB, BU, GM and GR, respectively lower as compared with the control (Table 12). The use of $EMAS_2$ had a greater reduction effect than the use of $EMAS_1$. In agreement with findings of the present study, Szymanski and Patterson (2003) observed a general trend of decreasing EC after completion of dosages of EM application.

Table 11 Mean comparison of temperature, pH, EC, turbidity, TSS and TDS for concentration by location interaction of wastewater treated with $EMAS_1$

Treatment combination	Temp (ºC)	pH	EC (µS/cm)	Turbidity (NTU)	TSS (mg/l)	TDS (mg/l)
OB X 0ml L^{-1}	22.50^{bcd}	4.35^{j}	546.00^{c}	157.36^{a}	612^{c}	532.33^{c}
OB X 2mlL^{-1}	22.57^{bc}	5.28^{c}	497.33^{efg}	56.10^{i}	446.3^{j}	404.67^{g}
OB X 4ml L^{-1}	22.27^{def}	5.80^{b}	408.00^{h}	57.97^{i}	310.7^{r}	245^{k}
OB X 6ml L^{-1}	22.73^{ab}	4.97^{fg}	547.33^{c}	56.83^{i}	406^{m}	355. 33^{i}
OB X 8ml L^{-1}	23.07^{fg}	4.73^{hi}	620.33^{ab}	47.67^{j}	495.7^{f}	430.67^{ef}
BU X 0ml L^{-1}	22.53^{bcd}	4.18^{k}	482.67^{fg}	137.67^{b}	673.3^{a}	571.0^{a}
BU X 2ml L^{-1}	22.50^{bcd}	5.06^{ef}	332.00^{j}	76.83^{ef}	462.7^{i}	418.67^{f}
BU X 4ml L^{-1}	22.50^{bcd}	5.87^{b}	319.67^{j}	72.47^{fg}	427.7^{k}	322.0^{j}
BU X 6ml L^{-1}	22.43^{cde}	4.90^{g}	532.33^{cd}	84.27^{d}	379.3^{o}	250.0^{k}
BU X 8ml L^{-1}	22.77^{ab}	4.67^{i}	605.00^{b}	68.27^{gh}	485.3^{g}	439.67^{e}
GM X 0ml L^{-1}	22.53^{bcd}	4.40^{j}	490.33^{fg}	106.33^{c}	633.3^{b}	557.0^{b}
GM X 2ml L^{-1}	22.53^{bcd}	5.18^{cd}	468.33^{g}	78.07^{e}	478.3^{h}	391.0^{h}
GM X 4ml L^{-1}	22.57^{bc}	6.06^{a}	368.00^{i}	68.50^{gh}	412.7^{l}	404.3^{g}
GM X 6ml L^{-1}	22.53^{bcd}	4.93^{g}	512.67^{def}	58.83^{i}	366.0^{p}	354^{i}
GM X 8ml L^{-1}	22.67^{bc}	4.78^{h}	604.00^{b}	42.07^{k}	537.3^{e}	501.33^{d}
GR X 0ml L^{-1}	21.97^{g}	4.72^{hi}	530.00^{cde}	110.40^{c}	590.0^{d}	553.33^{b}
GR X 2ml L^{-1}	22.53^{bcd}	5.12^{de}	511.00^{def}	84.67^{d}	433.0^{k}	388.33^{h}
GR X 4mlL^{-1}	22.57^{bc}	5.99^{a}	469.67^{def}	67.63^{h}	394.3^{n}	321.67^{j}
GR X 6ml L^{-1}	22.17^{efg}	5.11^{de}	591.67^{b}	56.33^{i}	349.0^{q}	339.67^{m}
GR X 8ml L^{-1}	23.00^{a}	4.95^{g}	649.33^{a}	59.30^{i}	487.0^{g}	413.0^{fg}
S.E±	0.184	0.06	20.21	2.84	3.74	2.65
LSD (5%)	0.30	0.10	33.02	4.64	8.087	11.03
CV	0.813	1.21	4.01	3.67	0.80	0.64

LSD = least significant difference, SE = standard error CV= coefficient of variance, TSS = total suspended solid TDS = total dissolved solid, and EC = electric conductance

Means followed by the same letter/s within the column are not statistically different

Table 12 Mean comparison of temperature, pH, EC, turbidity, TSS and TDS for concentration by location interaction of wastewater treated with $EMAS_2$.

Treatment combination	Temp (ºC)	pH	EC (µS/cm)	Turbidity (NTU)	TSS (mg/l)	TDS (mg/l)
OB X 0ml L^{-1}	22.50^{def}	4.35^{k}	546.00^{e}	157.36^{a}	613.33^{c}	533.33^{b}
OB X 3mlL^{-1}	22.40^{f}	5.38^{d}	466.00^{hi}	39.77^{m}	446.3^{j}	393.67^{i}
OB X 6ml L^{-1}	22.53^{cdef}	5.02^{f}	323.67^{k}	46.10^{kl}	311.67^{o}	241.67^{p}
OB X 9ml L^{-1}	22.50^{def}	4.36^{k}	540.33^{ef}	41.40^{lm}	406.00^{i}	356.33^{k}
OB X 12ml L^{-1}	23.00^{a}	4.34^{k}	767.33^{a}	43.20^{lm}	495.67	430.00^{fg}
BU X 0ml L^{-1}	22.53^{cdef}	4.18^{l}	482.67^{g}	137.67^{b}	686.67^{a}	571.67^{a}
BU X 3ml L^{-1}	22.90^{ab}	5.58^{c}	336.33^{k}	71.10^{g}	460.33^{h}	418.67^{g}
BU X 6ml L^{-1}	22.47^{ef}	5.16^{e}	273.67^{l}	69.83^{gh}	429.33^{k}	321.00^{m}
BU X 9ml L^{-1}	22.97^{a}	4.86^{g}	495.67^{g}	77.93^{f}	376.67^{l}	248.33^{op}
BU X 12ml L^{-1}	22.67^{cde}	4.46^{j}	600.00^{c}	93.50^{e}	490.00^{g}	442.00^{f}
GM X 0ml L^{-1}	22.53^{cdef}	4.40^{jk}	490.33^{g}	106.33^{cd}	633.33^{b}	557.00^{b}
GM X 3ml L^{-1}	22.70^{bcd}	4.92^{g}	529.33^{f}	66.13^{h}	431.67^{k}	387.67^{j}
GM X 6ml L^{-1}	22.47^{ef}	5.99^{a}	376.67^{j}	49.50^{jk}	425.00^{kl}	388.67^{j}
GM X 9ml L^{-1}	23.00^{a}	4.69^{h}	480.67^{gh}	41.80^{lm}	368.67^{m}	335.00^{l}
GM X 12ml L^{-1}	22.63^{cde}	4.43^{jk}	566.33^{d}	50.97^{j}	535.00^{e}	456.00^{e}
GR X 0ml L^{-1}	21.97^{g}	4.72^{h}	530.00^{ef}	110.40^{c}	586.67^{d}	486.67^{d}
GR X 3ml L^{-1}	23.00^{a}	5.73^{b}	525.00^{f}	59.67^{i}	433.67^{k}	372.00^{jk}
GR X 6mlL^{-1}	22.57^{cdef}	5.18^{e}	372.67^{j}	65.43^{h}	385^{m}	302.33^{n}
GR X 9ml L^{-1}	23.10^{a}	4.98^{g}	449.67^{i}	82.27^{f}	342^{n}	253.67^{o}
GR X 12ml L^{-1}	22.73^{bc}	4.58^{i}	625.33^{b}	102.80^{d}	483.67^{f}	411.33^{h}
S.E±	0.1335	0.058	10.142	2.936	6.50	8.21
LSD (5%)	0.218	0.096	16.57	4.797	4.80	6.05
CV	0.59	1.2	2.07	3.88	1.07	3.80

LSD= least significant difference, SE = standard error CV= coefficient of variance, TSS= total suspended solid TDS= total dissolved solid, and EC=electric conductance,
Means followed by the same letter/s within the column are not statistically different

4.2.8. pH in treated effluent

Linich (2001) reported that a change in microbial populations may impact upon pH. The pH value determines the condition of the wastewater to be weather acidic or alkaline. Most microorganisms prefer for their action pH between 6 and 8 whereas some microorganisms in EM like lactic acid bacteria prefer the pH to be acidic.

The pH value for the effluent treated with EM activated with molasses, EM1 and distilled water ($EMAS_1$) was observed to be highly significant ($P<0.01$) (Appendix table 2) due to the interaction effect between concentration level of $EMAS_1$ and location. Treatment of wet coffee processing wastewater with $EMAS_1$ raised the pH value to the maximum from GM (6.06) followed by GR (5.99) were effluent treated with 4 ml $EMAS_1$. In contrast, the level of pH remained to be the lowest (slightly acidic) in the effluent from BU (4.18) which had not received any $EMAS_1$ treatment followed by the pH value registered due to the treatments of effluent from OB (4.35) and GM (4.4) with 0 ml $EMAS_1$ (Table 11).

Though the increase in pH of effluent was very low, the analysis of variance for pH show that the interaction effect between location and concentration level of EM was observed to be significant ($P<0.01$). The maximum increment in pH observed for effluent from GR (5.73). However, the pH for effluent from OB (4.34) and GM (4.43) which was treated with 12ml $EMAS_2$ showed no increment rather a similar value revealed from the effluent of the same location, which did not receive any EM application. The pH increments were high for effluents from GM (5.99) treated with 6 ml $EMAS_2$ followed by OB (5.38), BU (5.58) and GR (5.73) which was treated with 3ml $EMAS_2$ (Table 12). Based on the results obtained from the two experiments, application of 4ml of $EMAS_1$ and 3ml $EMAS_2$ resulted in high increment in pH, however, as the concentration level for EM increased, the pH value went down making the effluent acidic. Despite the observed increment in pH, most treated wastewater remained in slightly acidic condition. This could probably be attributed to the lactic acid bacteria in the EM that produce lactic acid as the major metabolic end product from the digestion of organic matter in addition to the acidic nature of wet coffee processing effluent. On the other hand, the slight increment of pH in the EM treatment was due to the

decrease in the organic matter content that contribute for acid condition by releasing the organic acid.

4.2.9. Turbidity

Turbidity of water is an important parameter as it contributes to the aesthetics of water and leads to its acceptance or rejection for consumption. The treated effluent depict that the turbidity value were highly significantly different for the interaction effect at $P<0.01$ (Appendix table 2). The mean value for all concentration and location were much lower than that of the control with small difference among treated effluent value under both experiments. The lowest mean value obtained in experiment one was for effluent from collected OB (56.1NTU), when treated with application 2ml $EMAS_1$ followed by 56.83 NTU, 73.57 NTU, 47.4 NTU and 58.5 NTU for OB, BU, GM and GR, respectively when treated with 8ml (Table 11).

The turbidity levels wastewater treated with $EMAS_2$ was highly significantly ($P<0.01$) affected by the interaction of concentration level and location (Appendix table 3). But the level of reduction for each location was different. For effluent from Omobeko, though high reduction was observed with application of 3 ml activated $EMAS_2$ the other concentration levels also had similar reduction in turbidity. At the application of 3 ml and 6ml $EMAS_2$ for Bulbulo, 9 ml for Gembe and 6 ml for Geruke, high reduction levels were observed (Table 12). The decrease in turbidity is due to the EM activity on decreasing the other physicochemical parameters like nutrients and solids. Though the reduction in turbidity level did not reach to the permissible limit, the result of the present study is an indicator of the potential of EM in turbidity reduction. In line with this, the action of effective microorganism using the toxic microorganisms and organic matter as a food source had been opined by Nicholas (1996) hence EM may contribute to the reduction of turbidity level in wastewater.

4.2.10. TSS and TDS

The TSS (total suspended solid) and the TDS (total dissolved solid) wastewater from wet coffee processing were significantly affected due to the interaction effect of concentration level and location at $P<0.01$ (Appendix table 2 and 3). Pertaining to the effluent treated with $EMAS_1$ the maximum reduction value observed from OB with TSS (356.67 mg/l) and TDS (253.33mg/l) followed by GR with TSS (389.33mg/l) and BU with TDS (315.00mg/l) which was treated with 4ml $EMAS_1$. On the other hand the effluent from BU remained highest for TSS (687.67mg/l) and TDS (571.67mg/l) as it had no EMAS treatment received.

As illustrated in Table 11 the level of TSS and TDS decreased when the application of $EMAS_1$ was raised from 2 ml to 4 ml whereas the value slowly decreased as the concentration level increased. Accordingly, the reduction of TSS and TDS for effluent from GM and GR following the application of 6 ml and 8ml $EMAS_1$ resulted in lowest decrease in value. The percentage reduction in value, as compared to the control (0 ml treated) for the effluent from OB, BU, GM and GR in terms of TSS were 41.8%, 37.3%, 38.2% and 33.6%, respectively and TDS were 52.5%, 44.9%, 37.5% and 39.9% after application of 4ml $EMAS_1$.

As a follow up effort, $EMAS_2$ was used instead of $EMAS_1$ to treat wet coffee processing wastewater and it was observed that from the mean TSS and TDS value, the application of 6ml $EMAS_2$ demonstrated the maximum decrease than other concentration levels. The interaction effect of location with concentration level on TSS and TDS value was highly significant ($P<0.01$). The highest decrease for the different locations was 368.67 and 342.42 mg/l 335.35, 253.67 for BU, GM and GR, respectively when effluent was treated with 9ml $EMAS_2$ while for OB (331.) it was effective with 6ml $EMAS_2$. The minimum value for the TDS was 241.67 67mg/l which was obtained with application of 6ml $EMAS_2$ for the effluent from OB while for locations BU, GM and GR observed values were 248.33, 335.00 and 253.67 mg/l with the application of 9ml $EMAS_2$ (Table 12). However, further increase in the concentration levels of $EMAS_2$ resulted in the increase of TSS and TDS value. The reduction percentage for the wastewater treated with $EMAS_2$ was in terms of TSS 47%, 50%, 53% and 44%; and TDS 44%, 49%, 39% and 25% for effluents from OB, BU, GM and GR,

respectively when treated with 6ml $EMAS_2$. A similar finding was reported by Rashid and West (2007) on the treatment of industrial wastewater using EM with the reduction of TSS by 71% by Firdaus (2007) by 94%.

Concisely, decrease in TSS and TDS observed (Table 11 and 12) for the treated wastewater with $EMAS_1$ and $EMAS_2$, respectively. The highest TSS and TDS values observed when the wastewater treated with $EMAS_2$ than $EMAS_1$. The use of $EMAS_2$ was cost effective due to the decrease in the use of EM1 and molasses for activation by 33.3%. The mean value TSS and TDS for the main effects of treated wastewater for both experiment were significantly different (Table 11 and 12). The significant decrease in the value of TSS and TDS is due to the biodegradable organic materials, which were used by microorganisms as a nutrient or food, decrease the organic matter available in the wastewater as a result the decrement in TSS and TDS was observed.

The study on dairy wastewater treatment using EM by HortResearch and Biological Husbandry Unit of New Zealand in 2003 shows the similar result with this study that at the beginning of reaction the values of TSS in the EM extended treatments are much lower than that control. This may explained by the activity of certain microorganisms (such as Microzyme in the EM extended) which can excrete flocculent material, thereby improving the settlement performance of sludge. In contrast to this study, in municipal sludge there was a significantly higher level of solids in the treated tanks compared to the control there were not sufficient changes to sludge volume (CHWTP) or suspended solids (septic tanks) to indicate a clear benefit from the use of EM in wastewater (Szymanski and Patterson 2003).

4.2.11. Correlation for EMAS treated wastewater

4.2.11.1. Correlation of physicochemical characteristics treated wastewater-using $EMAS_1$

The current study unveiled that the BOD was highly significant and negatively related with the dissolved oxygen (r^2 = -0.91). This is because of the microorganism in wastewater which need oxygen for their survival because of EMAS used. Biochemical oxygen demand, but

inversely related to the dissolved oxygen (Balasubramanian *et al.,* 2012). The BOD was highly significant and positively correlated with NO_3 (r^2 = 0.50), NH_3 (r^2 = 0.72) and PO_4 (r^2 = 0.52) and $p<0.01$. The BOD also highly positively significant and related with TSS (r^2=0.50), TDS (r^2 = 0.48), EC (r^2 = 0.49) and turbidity (r^2 = 0.63) but negatively related with pH (r^2 = -0.74) (Table 13).

The NO_3 was positively and highly significant with $p<0.01$ related with NH_3 (r^2=0.55) the it also positively related with temperature (r^2 = 0.34), EC (r^2 = 0.65), TSS (r^2 = 0.72) and TDS (r^2=0.73). The NO_3 amount in treated coffee effluent negatively related with DO (r^2 = -0.51). The NH_3 from this study was positively and highly related with $p<0.01$ with PO_4 (r^2 = 0.44), EC (r^2 = 0.48), turbidity (r^2 = 0.69), TSS (r^2 = 0.89) and TDS (r^2 = 0.90). But NH_3 negatively related with DO (r^2 = -0.71) and pH (r^2 = -0.51). The PO_4 result of this experiment also positively and highly ($p<0.01$) related with EC (r^2 = 0.575), TSS (r^2 = 0.577) and TDS (r^2 = 0.496). Though PO_4 was negatively related with DO (r^2 = -0.579), pH (r^2 = -0.47) and temperature (r^2 = -0.65) (Table 13).

The DO was positively related only with pH (r^2 = 0.71) as the amount of dissolved oxygen in the treated wastewater increased the pH also increased. However the DO was inversely related with EC (r^2 = -0.45), turbidity (r^2 = -0.50), TSS (r^2 = -0.42) and TDS (r^2 = -0.43). The electrical conductivity of effluent treated with $EMAS_1$ was positively related with TSS (r^2 = 0.39) and TDS (r^2 = 0.51). TSS was positively related with TDS (r^2 = 0.93) (Table 13).

Table 13 Correlation of physicochemical characteristics treated wastewater-using $EMAS_1$

	NO_3	NH_3	PO_4	BOD	DO	Temp.	pH	EC	Turb.	TSS	TDS
NO3	1.00										
NH3	0.55**	1.00									
PO4	0.59**	0.44**	1.00								
BOD	0.50**	0.72**	0.52**	1.00							
DO	-0.51**	-0.71**	-0.62**	-0.91**	1.00						
Temp	0.34*	-0.47**	-0.27*	0.01^{ns}	-0.04^{ns}	1.00					
pH	-0.07^{ns}	-0.51**	-0.47**	-0.74**	0.71**	-0.09^{ns}	1.00				
EC	0.65**	0.48**	0.44**	0.49**	-0.45**	0.14^{ns}	-0.57**	1.00			
Turb.	-0.48**	0.69**	0.05^{ns}	0.63**	-0.50**	-0.25^{ns}	-0.43**	-0.09^{ns}	1.00		
TSS	0.72**	0.89**	0.18^{ns}	0.50**	-0.42**	-0.48**	-0.22^{ns}	0.39**	0.67**	1.00	
TDS	0.73**	0.90**	0.12^{ns}	0.48**	-0.43**	0.52**	-0.23^{ns}	0.51**	0.61**	0.93**	1.00

NO_3 = nitrate; NH_3= ammonia; PO_4 = phosphate BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp=temperature; pH=power of hydrogen; Turb = turbidity; EC = electrical conductivity; TSS = total suspended solid and TDS = total dissolved solid.

4.2.11.2. Correlation of physicochemical characteristics treated wastewater-using $EMAS_2$

The correlation of parameters in experiment two using $EMAS_2$ for effluent treatment reveals that the BOD was highly significant and negatively related with the dissolved oxygen (r^2= -0.91) this is because of the microorganism in wastewater which need oxygen for their survival decrease because of $EMAS_2$ used. The BOD was highly significant and positively correlated with NO_3 (r^2 = 0.50), and NH_3 (r^2 = 0.73) where as unlike experiment one the BOD was negatively related with PO_4 (r^2 = -0.52) the p values were $p<0.01$. The BOD also highly significant and related with EC (r^2 = 0.70), turbidity (r^2 = 0.64), TSS (r^2 = 0.76) and TDS (r^2 = 0.82), where as negatively related with pH (r^2 = -0.65) (Table 14).

The NO_3 was positively and highly significant with $p<0.01$ related with NH_3 (r^2 = 0.71) this due to the nitrification and the denitrification process. The NO_3 also positively related with EC (r^2 = 0.74), TSS (r^2 = 0.56) and TDS (r^2 = 0.59). The nitrate amount in treated coffee effluent negatively related with DO (r^2 = -0.51) and the correlation with pH (r^2 = -0.08) was not significant. The ammonia from this study was positively and highly related with $p<0.01$

with EC (r^2 = 0.35), turbidity (r^2 = 0.70), TSS (r^2 = 0.49) and TDS (r^2 = 0.51). But NH3 negatively related with DO (r^2 = -0.71) and pH (r^2 = -0.51) (Table 14).

The DO was positively related with pH (r^2 = 0.71) and PO_4 (r^2 = 0.63) as the amount of dissolved oxygen in the treated wastewater increased the pH also increased. However the DO was inversely related with EC (r^2 = -0.79), turbidity (r^2 = -0.51), TSS (r^2 = -0.83) and TDS (r^2 = -0.77). The pH in this study was negatively related with EC (r^2 = -0.59), turbidity (r^2 = -0.434), TSS (r^2 = -0.48) and TDS (r^2 = -0.71). The EC in this study was positively related with TSS (r^2 = 0.67) and (r^2 = 0.79). The turbidity was significantly and positively correlated with BOD (r^2 = 0.64), ammonia (r^2 = 0.70), nitrate (r^2 = 0.49), TSS (r^2 = 0.71) and TDS (r^2 = 0.61) and negatively and significantly with DO (r^2 = -0.51) and pH (r^2 = -0.48). The TSS also positively related with TDS (r^2 = 0.96) (Table 14).

Table 14 Correlation of physicochemical characteristics treated wastewater using $EMAS_2$

	NO_3	NH_3	PO_4	BOD	DO	Temp.	pH	EC	Turb.	TSS	TDS
NO_3	1.00										
NH_3	0.71**	1.00									
PO_4	0.002^{ns}	-0.13^{ns}	1.00								
BOD	0.50**	0.73**	-0.52**	1.00							
DO	-0.51**	-0.71**	0.63**	-0.91**	1.00						
Temp	-0.34**	-0.47**	-0.29*	-0.004^{ns}	0.04^{ns}	1.00					
pH	-0.08^{ns}	-0.51**	0.47**	-0.65**	0.71**	0.09^{ns}	1.00				
EC	0.74**	0.35**	-0.71**	0.70**	-0.79**	0.19^{ns}	-0.59**	1.00			
Turb.	0.49**	0.70**	0.05^{ns}	0.64**	-0.51**	-0.25^{ns}	-0.43**	0.13^{ns}	1.00		
TSS	0.56**	0.49**	-0.58**	0.76**	-0.83**	0.16^{ns}	-0.48**	0.67**	0.71**	1.00	
TDS	0.59*	0.51**	-0.62**	0.82**	-0.77**	0.11^{ns}	-0.708**	0.79**	0.61**	0.96^{**}	1.00

NO_3 =nitrate; NH_3= ammonia; PO_4 = phosphate BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH=power of hydrogen; Turb = turbidity; EC = electrical conductivity; TSS = total suspended solid and TDS = total dissolved solid.

5. SUMMARY AND CONCLUSION

Coffee plays a great role in Ethiopian economy and its production is increasing widely. The coffee commerce currently supports millions of Ethiopians and serves as main source of GDP for the country. The coffee, which goes for export market, processed in dry and wet processing method and coffee processed in wet method generate more income than the dry. The government also gives more attention to expand wet method which known bringing premium price although coffee though the high amount of wastewater released from this method pollutes the river and its surrounding environment due to generation of organic matter from the coffee berries during pulping and fermentation processes. Jimma zone is known for its high production of coffee specially Gomma and Mana weredas hence this study was done on wet coffee processing effluent at four selected locations three from Gomma and one from Mana weredas with the objective of characterization and assessing impact of wet coffee processing effluent and treating the effluent with effective microorganism at laboratory scale.

Based on the results of the two laboratory experiments conducted to treat effluents with $EMAS_1$ and $EMAS_2$, the effect of treatment of wastewater with EMAS was significant when compared to the control. The physicochemical property of treated wastewater revealed a reduction of 49.5-60.5% BOD, 23.05-26.88% NO_3, 41-46% NH_3 36-52% PO_4, 24-33% EC 35-47% turbidity, 16-23% TSS and 33-41% TDS and increment of pH by 37-52% and DO by 1.4-2.5mg/l accountable to the treatment of effluent with 4ml $EMAS_1$. Moreover, treatment with $EMAS_2$ demonstrate much more reduction than treatment with $EMAS_1$; with observed reduction of 49.5-60.5% BOD_5, 40-45% NO_3, 72-82% NH_3 and 30-52% PO_4 when treated with 9ml $EMAS_2$ and 29-40% EC, 40-53% turbidity, TSS and 44-53% TDS and increment of pH by 25-49% and DO by 1.99-2.8mg/l when treated with 6ml $EMAS_2$.

For most physicochemical characteristics, the use of $EMAS_2$ with 6 ml and $EMAS_1$ relatively had a better purification capacity than other levels on average. However, from the two experiments the use of $EMAS_2$ with 6 ml was economical than $EMAS_1$ of 4 ml due to the use of molasses and EM1 were lower than $EMAS_1$ and the percentage of purification were better than $EMAS_1$. Though this study was done at laboratory scale, the findings show that EM has a

potential on purification of wet coffee processing wastewater which can help to reduce the problem created due to wastewater. When compared with the WHO and EEPA permissible limits, the quality obtained after treatment of wet coffee processing wastewater with EMAS in terms of NO_3, NH_3, PO_4 and DO meet the standard for the discharge the BOD_5, TSS, TDS, pH and Turbidity were highly reduced but do not meet the standard to the permissible limit of WHO and EEPA. As the observation was done for short period, the temperature and EC showed only a slight change. Since this study was done only once under laboratory scale in order to determine the efficacy of EM, it is better to repeat on constructed lagoon or pond. In the future, this study should be done using constructed ponds and lagoons to determine the best concentration level and activation as well as the potential of purification at large scale.

As future line of work

- Detailed comprehensive study on the impact of wet coffee processing wastewater on near by community requires multidisciplinary study.
- The findings achieved in wet coffee processing wastewater with EM need to be confirmed with another study that involve evaluation at both in-situ and ex-situ condition.
- Additional researches on the treatment method of wet coffee processing wastewater on different location have to be done.
- There should be a multidisciplinary approach on developing strategies and treatment methods and implementing to reduce pollution load.

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APPENDIX

Appendix Table 1 ANOVA Mean Square Values for physicochemical parameters of four locations.

Parameters	DF	Omobeko	Bulbulo	Gembe	Geruke
NO_3	3	349.10**	445.85**	345.96**	447.35**
NH_3	3	77.04**	60.30**	44.86**	80.36**
PO_4	3	15.57**	12.44**	17.46**	14.27**
BOD_5	3	49104770.4**	85985787.2 **	72088233.9**	105741992.6**
DO	3	23.05**	21.15**	33.34**	28.02**
Temperature	3	21.75**	23.24**	11.86**	3.35**
pH	3	9.68**	12.66**	7.35**	9.8**
EC	3	142372.84**	93184.93**	128035.57**	82592.43**
Turbidity	3	293105.20**	192252.96**	86321.30**	117653.27**
TSS	3	1916405.35**	1060163.64**	1414624.17**	2169150.44**
TDS	3	1733030.5**	1377383.41**	1236432.53**	910802.97**

DF = degree of freedom, TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; pH=power of hydrogen; EC = electrical conductivity; NO3=nitrate; NH3= ammonia and PO_4 = phosphate

Appendix Table 2 ANOVA P value for physicochemical characteristics of effluents treated with $EMAS_1$

SV	DF	BOD_5	DO	NO_3	NH_3	PO_4	EC	pH	Temp.	Turb.	TSS	TDS
Con. level	3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Location	4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<.0.074^{ns}	<0.01	<0.01	<0.01
Con*loc	12	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<.0001	<0.01	<0.01	<0.01
CV (%)		0.94	3.385	2.4	1.673	3.95	0.81	1.21	4.01	3.67	0.71	1.1
R-Square		0.99	0.99	0.99	0.99	0.99	0.99	0.99	0.75	0.99	0.99	0.99
SE+/-		12.83	0.05	0.11	0.07	0.08	6.30	0.06	0.18	2.88	3.74	2.65

SV = source of variation; DF = degree of freedom; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp = temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO3=nitrate; NH_3 = ammonia; PO_4 = phosphate; con = concentration; loc=location; and Trt=treatment.

Appendix Table 3 ANOVA P value for physicochemical characteristics of effluents treated with $EMAS_2$

SV	DF	BOD5	DO	NO3	NH3	PO4	EC	pH	Temp.	Turb	TSS	TDS
Con.	3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<.01	<0.01	<0.01	<0.01
Location	4	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<.107^{ns}	<0.01	<0.01	<0.01
Con*loc	12	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<.0001	<0.01	<0.01	<0.01
CV (%)		0.937	2.5	2.01	1.87	3.56	2.99	1.20	0.59	3.98	1.05	0.93
R-Square		0.99	0.99	0.99	0.99	0.99	0.99	0.99	0.86	0.99	0.99	0.99

SV = source of variation; DF = degree of freedom; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp=temperature; pH = power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia; PO_4 = phosphate; con = concentration loc = location; and Trt = treatment.

Appendix Table 4 Mean square for ANOVA for physicochemical characteristics as influenced by $EMAS_1$ and location and their interaction.

SV	DF	NO_3	NH_3	PO_4	BOD	DO	Temp	pH	EC	Turbidity	TSS	TDS
Location	3	0.68**	0.27**	0.56**	201202**	0.93**	0.08^{ns}	0.15**	26202**	811**	9487**	34119**
EMAS1	4	19.53**	15.50**	3.90**	2621265**	7.40**	0.44**	3.81**	91773**	10176**	56852**	68646**
EMAS1* Location (con*loc)	12	0.23**	0.07**	0.39**	93364.**	0.16**	0.12**	0.03**	3408**	614**	2540**	3749**
CV (%)		1.74	1.67	3.95	2.25	3.38	0.81	1.21	4.01	3.67	0.58	0.85

*** = highly significant; ^{ns} = non significant; SV=source of variation; DF=degree of freedom; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp=temperature; pH=power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO_3 = nitrate; NH_3 = ammonia; PO_4 = phosphate; con = concentration; loc = location; and Trt=treatment.*

Appendix Table 5 Mean square for ANOVA for physicochemical characteristics as influenced by $EMAS_2$ and location and their interaction

SV	DF	NO_3	NH_3	PO_4	BOD	DO	Temp	pH	EC	Turbidity	TSS	TDS	
Location	3	1.40**	1.17**	0.08**	201203**	0.48**	0.39^{ns}	0.31**	21708**	2704**		11568**	19462**
$EMAS_2$	4	16.55**	37.6**	8.40**	2621265**	12.26**	0.51**	2.72**	14128461**	1066711**		58456**	51061**
$EMAS_2$* Location	12	0.16**	0.40**	0.35**	51394**	0.19**	0.18**	0.26**	10038**	1044.90**		20767**	2510**
CV (%)		2.01	1.87	3.56	0.93	2.50	0.58	1.20	2.07	3.88		0.75	1.00

*** = highly significant; ^{ns} = non significant; SV = source of variation; DF=degree of freedom; SE = standard error; CV = coefficient of variation; TSS = total suspended solid; TDS = total dissolved solid; BOD = Biochemical oxygen demand; DO = dissolved oxygen; Temp=temperature; pH=power of hydrogen; Turb = turbidity; EC = electrical conductivity; NO3=nitrate; NH3= ammonia; PO_4 = phosphate; con = concentration; loc = location; and Trt=treatment.*