WOLKITE UNIVERSITY

COLLEGE OF ENGINEERING AND THECNOLOGY

DEPARTMENT OF CHEMICAL ENGINEERING

Final project on: Extraction and formulation of perfume from lemon grass

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Table contents

List of acronyms

AOAC	Associates of Analytical Chemistry
GC	Gas chromatography
MS	Mass-spectrometer
SCFE	Supercritical fluid extraction
Esdp	Esprit de Parfum
EDP.	Eau de Parfum
EDT	Eau de toilette
EDC	Eau de Cologne
CIF	Cost insurance and freight
ANOVA	Analysis of variance
ASTM	American Society of Testing and Material
MSD	Mass selective detector

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Abstract

Lemon grass is perennial herb which has been cultivating in Ethiopia and known as tej-sar which has been using to decorate the house rather than for perfume application. The objective of this study was to develop a perfume at optimum extraction point using soxhlet extraction from tej-sar (Cymbopogon citratus). The effect of different parameters, such as extraction time and particle size was determined on the extraction yield of essential oil. The percentage oil yield in dried samples small size was higher as compared to dried samples of large size due to solvate formation.For soxhlet extraction, the minimum essential oil yield has been obtained 0.65% for large size particles with range of 2.5-5mm after the extraction time of 3:00 hr and 0.79% for small size particle of lemon grass after the extraction time of 2.30 hours with ranges of from 0.1-2.5mm. From the experiment what was obtained was applicable for perfume due to it consisted of citral number that has lemony aroma. The extracted oil from tej-sar was developed into perfume using a fixative.

1. Introduction

1.1. Background

Perfume is a mixture of fragrant essential oils and aroma compounds, fixatives and solvents used to give the human body, objects and living spaces a pleasant smell.

Since the beginning of recorded history, humans have attempted to mask or enhance their own odor by using perfume which emulates nature's pleasant smells. Many natural and man-made materials have been used to make perfume to apply to the skin and clothing to put in cleaners and cosmetics or to scent the air. This is because of differences in body chemistry, temperature and body odors (Auracacia, 2016). Perfumes and scented articles were in use from pre Vedic and Vedic periods for religious practices, social customs and domestic rituals and later gradually became part and parcel of human life. Perfumes were also used in cosmetics and beauty aids. Medicinal values of many perfume was well known to ancient and were used in both rituals and to treat diseases. Perfumes as luxury accessory or one of the most popular must have items on the aspirational buyers' list as well (Auracacia, 2016). Perfume is extremely difficult to produce the large required a quantity of different ingredients to produce a very small essence. The difficulty of production resulted in scarcity and hence made the substance even more valuable and expensive. In Greek society, perfume or odor was one of the main symbols of beauty (Donato and Seefried, 1980).

Perfume is presently the most important in Ethiopia that used to provide a pleasant odor, cover the scent of the base of the product, give a product an identity, provide product concept support and signify a change in a product. Ethiopia is one of the countries which have been using perfume traditionally for many years. According to Ethiopian revenue and custom authority 2015, Ethiopia import perfume an average of 112,589.908kg annually from different countries to satisfied the demand of the buyers (ERCA, 2015). Due to this reason Ethiopia loss its foreign currency as much. Lemongrass is perennial herb largely cultivated in tropical and subtropical countries. The scientific name of lemongrass is Cymbopogon citratus. The Cymbopogon word derives from the Greek words “kymbe” (boat) and “pogon” (beard) referring to the arrangement of the spike of the flower. The word citratus derives from the old Latin meaning lemon-scented leaves (Shah *et al.*, 2011).

It is a perennial tropical grass; resistant to different temperatures and can grow in warm, semi-warm and temperate climates. It is from 60 to120 centimeters high, its leaves are green, long and slats and have pleasant aroma and taste.

This grass is native to India (Parikh & Desai, 2011). The essential oil of the C. citratus is one of the most important volatiles oils which is economically important component of lemon grass (Lewinsohn *et al*., 1998).The oil used in brazil for medicinal purposes, perfume, food and pharmaceuticals (Koshima *et al.*, 2006).These days the demand for the lemongrassperfume has soared because of its unique fragrance.While it is widely used in the different types of herbal teas and non-alcoholic beverages. Lemongrass also known in Ethiopia as (tej-sar) .There is two common lemongrass species that were registered in the book of Ethiopia and Eritrea flora (Tewolde and sue Edwards, 1997). Those are cymbopogon citratus and nardus. Cymbopogon citratus grass has been growing for many years in Ethiopia as backyard plant.This herb plant used by the farmer for decoration of the house during holiday instead of using for perfume and other application. From this Ethiopia has capacity to cultivate this grass due to available land, high cultivation period and good whether condition and that used as raw material for perfume development.

1.2 Statement of Problems

The problem of formulation of perfume involves knowing the proportion in which essential oil and other materials to be mixed to avoid skin irritation and increase the intensity and longevity of the perfume. Most imported perfumes are synthetic odorant which are not pure natural fragrance and it is a mixture of organic compounds that are harmful when it applied. Today there is a high demand of perfume for various purposes such as medicinal, soap making household, personal hygiene and insecticides even if it is significant, no perfume factories that are erected in Ethiopia rather than imported. Imported perfume are also very expensive to meet the demand of our local consumer industries besides direct human use, In particular perfumes that are usually imported can be produced locally from a vast variety of oil bearing plants yet to be explored. Therefore it becomes necessary to source and extract this oil from local source such as lemongrass that used to develop perfume by extracting the essential oil with soxlet extraction.

1.3 OBJECTIVES

1.3.1 General objective

The main objective of this project is to extract and formulate perfume from Lemon grass (Cymbopogon citratus).

1.3.2 Specific objectives

- Raw material preparation and Extraction of essential oils from lemon grass
- Characterization of essential oil extracted from lemongrass.

1.4 Significance of the study

- To secure our perfume supply as well as to reduce its perfume import bill through the development of an indigenous perfume production.
- To reduce any side effect resulting from synthetic chemicals by substituting natural one.
- To create awareness for stockholders as it is possible to establish local industries for perfume and oil production because of available land and cheap raw materials.

2. Literature Review

2.1 The History of Perfumery

Perfumery is the art of making perfumes began in ancient Mesopotamia and Egypt and was further refined by the Romans and Persians. Knowledge of perfumery came to Europe as early as the 14th century due partially to the spread of Islam. The first modern perfume made of scented oils blended in an alcohol solution was made in 1370 at the command of queen Elizabeth of Hungary and was known throughout Europe as Hungary Water.The art of perfumery prospered in Renaissance Italy and in the 16th century. Italian refinements were taken to France by Catherine de' Medici‟s personal perfumer (Elizabeth, 2011). The most detailed perfume recipes for composite scented preparations were written down ancient Egypt only during the Ptolemaic Period. During this period the doors to the classical World were opened wide to the ancient Egyptians and besides the Egyptian language Latin and Greek were understood. These were inscribed in hieroglyphs on the walls of perfume incense „laboratories‟ of temples. Other sources such as actual remains of plants found in excavations and wall paintings and reliefs in temples and tombs provide us with knowledge of the plants used by the Egyptians for perfume production and the methods they used .The bulk of our knowledge about perfume recipes from ancient world and the plants used in them, however, it isderived from various classical authors both Greek and Roman("The essence and use of perfume in ancient Egypt by Sheila Ann by submitted in accordance with the requirements for the degree of master of arts in the subject ancient near eastern studies at the University of South Africa Supervisor : Professor P S Vermaak joint supervisor : Mrs a Ferreira February 2012," 2012). Most modern perfumes are alcohol-based and contain synthetic scents. While the term perfume usually refers to fragrances in general, in the more technical language of the perfumer, a perfume must contain over 15% of fragrance oils in alcohol. The preferred fragrances for perfumes are no means universal, but differ according to cultural dictates and fashions. In the sixteenth century, for example pungent animal scents such as musk and civet were very popular. In the nineteenth century by contrast such animal scents were generally considered too crude and light floral fragrances were favored (Calkin and Jellinek, 1994).

The ancient Egyptians highly prized their botanical treasures in which Egypt was richly endowed and for the growing of which its climate was admirably suited, being moderate and uniform. The Egyptians made various types of perfumes from fragrant plants oils and fats employing various methods and involving various professions (Brun, 2000; Dayagí-Mendels, 1989). There are two main categories of perfumery products which toiletries and household products. Toiletries are defined as fine fragrances, personal care products, cosmetics and deodorants. Household products are considered to be air fresheners, laundry products, surface cleaners and disinfectants.

2.2 Concentration of Perfume

Perfume types reflect the concentration of aromatic compounds in a solvent (which in fine fragrance) is typically ethanol or a mix of water and ethanol. Various sources differ considerably in the definitions of perfume types. The intensity and longevity of a perfume is based on the conc entration, intensity and longevity of the aromatic compounds (natural essential oils or perfume oils) used: As the percentage of aromatic compounds increases, so does the intensity and longevity of the scent created. Specific terms are used to describe a fragrance's approximate concentration by percent/volume on perfume oil which is typically vague or imprecise.

- A list of common terms (Perfume-Classification) is as follows (Burr and chandler, 2008).
- Perfume extract or simply perfume (Extract): 15-40% aromatic compounds.
- Esprit de Parfum (ESdP): 15-30% aromatic compounds a seldom used strength
- concentration in between EdP and perfume.
- Eau de Parfum (EdP), Parfum de Toilette (PdT): 10-20% (typical 15%) aromatic compounds.
- Eau de toilette (EdT): 5-15% (typical 10%) aromatic compounds.
- Eau de Cologne (EdC): Chypre citrus type perfumes with 3-8% (typical 5%) aromatic compounds.

Perfume mist: 3-8% aromatic compounds (typical non-alcohol solvent).

- Splash (EdS) and Aftershave: 1-3% aromatic compounds.

2.3 Perfume Notes

Perfume is described in a musical metaphor as having three sets of notes making the harmonious scent accord. The notes unfold over time with the immediate impression of the top note leading to the keeper middle notes and the base notes gradually appearing as the final stage. These notes are created carefully with knowledge of the evaporation process of the perfume (Burr and chandler, 2008).

Top notes: The scents that are perceived immediately on application of a perfume. Top notes consist of small light molecules that evaporate quickly. They form a person's initial impression of a perfume and thus are very important in the selling of a perfume.

Middle notes: The scent of a perfume that emerges just prior to when the top notes dissipate. The middle note compounds form the "heart" or main body of a perfume and act to mask the often unpleasant initial impression of base notes which become more pleasant with time. They are also called the heart notes.

Base notes: The scent of a perfume that appears close to the departure of the middle notes. The base and middle notes together are the main theme of a perfume. Base notes bring depth and solidity to a perfume. Compounds of this class of scents are typically rich and “deep" and are usually not perceived until 30 minutes after application. The scents in the top and middle notes are influenced by the base notes as well the scents of the base notes will be altered by the type of fragrance materials used as middle notes.

2.4 Component of Perfume

There are three basic parts of perfume that make up components for the formulation:

Basics**:** The base is the most important component of any perfume. A base can be any fragrance or scent that is a not as volatile. This scent can be added to the perfume which will result in a product that meets consumer desires.

Fixatives: Fixatives are ingredients in the perfume that prolong the odor effect and delay the evaporation rate of volatile materials. The component tends to have no odor to be miscible in polar and non-polar solutions and to be at a higher boiling point temperature.

The fixative bonds to polar compounds within the perfume through hydrogen bonding reducing the overall vapor pressure of the mixture. They retain a high concentration of the top and middle notes and release them slowly over time.

Examples of fixatives are vetyver, dipropylene glycol, diethyl phthalate
and glycerin.

Solvents: The solvent is the portion of the perfume in which all the components are dissolved. Solvents are used to dilute the mixture in order to increase the surface area of application without ting an excessive amount of the fragrance. It is also used to reduce the intensity of the fragrance of the solution. Solvents decrease the price of the perfume per milliliter while increasing the amount of applications per bottle. Ethanol is the most common solvent that used for perfume formulation.

2.5 Perfume import in Ethiopia

Perfume is very significant in Ethiopia for different purpose in addition to body perfume.

Table 2.1 perfume imports from different countries, 2011-2015

Year	Net wt.(kg)/yr	Average /yr in kg	CIF Value (ETB)/yr	Average in annual(Birr)
2015	98,931	112,589.908	123,723,953.19	129,616,394
2014	164,898.94		200,754,677.02	
2013	126,095.36		126,607,519.23	
2012	86,207.57		95,911,419.74	
2011	86,816.67		101,084,402.80	

(Source: Ethiopian revenue and custom authority, Addis Ababa Ethiopia 2016)

2.6 Classification of Perfumery Materials

The raw materials employed in perfumery have natural origin (animal or plant) or synthetic origin (natural-identical or new artificial fragrant molecules called aroma chemicals).

- Aroma chemicals synthesized from crude oil making up to 70 to 100% of the perfume concentrate.
- Natural essential oils and plant extracts-constituting up to 30% of perfume composition.
- Animal products used in very small amounts up to 0.1% in the formulation.

Aromatics Source

Plant Sources:

Plants have long been used in perfumery as a source of essential oils and aroma compounds. These aromatics are usually secondary metabolites produced by plants as protection against herbivores, infections as well as to attract pollinators. Plants are by far the largest source of fragrant compounds used in perfumery. The sources of these compounds may be derived from various parts of a plant. A plant can offer more than one source of aromatics for instance the aerial portions and seeds of coriander have remarkably different odors from each other.

Animal Sources:

Ambergris: Lumps of oxidized fatty compounds whose precursors were secreted and excelled by the sperm whale. Ambergris should not be confused with yellow amber which is used in jewelry.

Because the harvesting of ambergris involves no harm to its animal source, it remains one of the few animalic fragrance agents around which little controversy now exists.

Castoreum: Obtained from the odorous sacs of the North American beaver.

Civet: Also called civet musk, this is obtained from the odorous sacs of the civets, the world society for the protection of animals investigated African civets caught for this purpose.

Hyraceum: Commonly known as "Africa Stone"is the petrified excrement of the Rock Hyrax.

Honeycomb: From the honeycomb of the honeybee both beeswax and honey can be solvent extracted to produce an absolute.

Deer musk**:** Originally derived from the musk sacs from the Asian musk deer, it has now been replaced by the use of synthetic musk sometimes known as "white musk".

Synthetic Source

Many modern perfumes contain synthesized odorants. Synthetics can provide fragrances which are not found in nature.

For instance, Calone, a compound of synthetic origin imparts a fresh ozonous metallic marine scent that is widely used in contemporary perfumes. Synthetic aromatics are often used as an alternate source of compounds that are not easily obtained from natural sources.

2.7 Essential Oil

Essential oils or volatile oils are found in many different plants. These oils are different from fatty oils because they evaporate or volatilize on contact with the air and they possess a pleasant taste and strong aromatic odor. They are readily removed from plant tissues without any change in composition. Essential oils are very complex in their chemical nature. The two main groups are the hydrocarbon terpenes and the oxygenated and sulphured oils (Gunther, 1994).

2.7.1 Essential Oil plant for perfumery in the world

Lavender Lavender perfumes are very old and were used by the Romans in their baths. It is still one of the most important scents. It is a low shrub with terminal spikes of very fragrant bluish flowers. The oil is important in the manufacture of Eau de Cologne and other perfumes and is also used in soaps, cosmetics and medicine as a mild stimulant. Lavender water a mixture of the oil in water and alcohol is popular in England (Yardley brand).

Jasmine

A highly esteemed perfume, jasmine is cultivated in southern France and surrounding areas. The flowers are picked as soon as they are open and the oil is extracted by enffleurag(Seminar, 2014).

2.7.2 Essential Oil plant in Ethiopia for perfumery application

Ethiopia has a long history of spice and herb production for the domestic market and has a unique, indigenous product. Modest quantities of several spices have been exported for centuries to countries in the Middle East and exports to Europe have developed over the past twenty years (Wikipedia).In general use, herbs are any plants used for flavoring, food, medicine and perfume. In Ethiopia several culinary herbs are produced: chervil, chives, coriander, dill, green basil, lovage, mint, oregano, rocolla, thyme, vernonia are Aromatic plant(Council & Willems, n.d).

- **Lavender**

Is a genus of 39 species of flowering plants in the mint family Lamiaceae.

The genus includes annual or short-lived herbaceous perennial plants and suffrutescent perennials, sub shrubs or small shrubs. Leaf shape is diverse across the genus. They are simple in some commonly cultivated species.

Frankincense

Frankincense and myrrh are phytotoxically safe raw materials in industries like pharmaceuticals and food industries. They are used in folk medicines, flavoring, beverages and liqueurs, cosmetics, detergents, creams and perfumery, paints, adhesives and dyes manufacturing. Both myrrh and frankincense are highly valued for their aromatic fragrances and are common ingredients in incense perfume and potpourris, soaps, detergents, creams and lotions and are often included in meditation blends, as it strengthens the psyche and aids in deepening the meditative state (FAO, 1995). Three types of frankincense products are recognized in Ethiopia:

Tigray, Ogaden and Borena Incense are aromatic biotic material which releases fragrant smoke when burned.

Myrrh

Myrrh is the aromatic resin of a number of small, thorny tree species of the genus Commiphora which is an essential oil, termed an oleoresin. Myrrh resin is a natural gum. It has been used throughout history as a perfume incense and medicine. It can also be ingested by mixing it with wine. In pharmacy, myrrh is used as an antiseptic in mouth washes, gargles and toothpastes.

Gum Arabic

Gum Arabic is used as thickening, stabilizing, emulsifying and suspending agent in food and drink industries; as tablet-binding agent, cream, lotions- suspending and emulsifying agents in pharmaceuticals, as film forming and sizing agent in printing and textile industries. It is also usedin ceramics, paints, inks, textiles and adhesives. In cosmetics, gum Arabic functions as a stabilizer in lotions and protective creams where it increases viscosity, imparts spreading properties and provides a protective coating and a smooth feel. It is used as an adhesive agent in blusher and as a foam stabilizer in liquid soaps.

Opoponax

Opoponax or sweet myrrh is a cousin of the healing Myrrh Commiphora.Myrrh with a warm balsamic and sweet, honey-like aroma. It is a natural oleo-gum-resin like myrrh and frankin cense. The color of its resin is brown; however, good quality crude botanical resin is dark red.

Opoponax has been a component of incense and perfumes since Biblical times. Talking of perfumery in particular,Opoponax qualities from several Commiphora are widely used especially in oriental fragrances to impart sweet balsamic notes.

Lemongrass (Cymbopogon Citratus)

A perennial plant with long, thin leaves, is one of the largely cultivated medicinal plants for its essential oils in parts of tropical and subtropical areas of Asia, Africa and America (Ranitha, Nour, Sulaiman, Nour, & S, 2014).Cymbopogon (lemon grass) is a genus of about 55 species of grasses (of which the type species is Cymbopogon citratus) native to warm temperate and tropical regions of the Old World and Oceania. It has a citrus flavor and can be dried and powdered or used fresh. Lemongrass oil is used as a pesticide and a preservative. Chemical investigations of essential oils in the nineteenth century revealed that many of the compounds responsible for the pleasant odors contained exactly ten carbon atoms.

These ten carbon compounds came to be known as terpenes if they were hydrocarbons and terpenoids if they contained oxygen and were alcohols, ketones or aldehydes. Eventually, it was found that there are also minor and less volatile plant constituents with fifteen, twenty, thirty and forty carbon atoms. Because compounds of ten carbons were originally called terpenes, they came to be called terpenoids if they contained oxygen and were alcohols, ketones and aldehydes (Pavia *et al*, 2005).There are two type of lemongrass(Teji sar) which are found in Ethiopia cymbopogon citratus and nardus.

2.8 Chemical Constituents of Essential Oils.

Essential oils are a mixture of fragrant compounds including terpenoids, aldehydes (geranial, citronellal), alcohols (geraniol, citronellol, nerol), esters (linalyl acetate, citronellyl acetate, isobornyl acetate (Burt, 2004), which accumulate in different organ like leaves, barks, woods, roots, rhizomes, fruits and seeds. Essential oils are synthesized and accumulated in specialized organs (trichomes, secretory cavities or canals) located near surfaces. Volatile fraction: Essential oil constituting of 90–95% of the oil in weight, containing the monoterpene and sesquiterpene hydrocarbons as well as their oxygenated derivatives along with aliphatic aldehydes, alcohols and esters.Non-volatile residue**:** that comprises 1–10% of the oil containing hydrocarbons, fatty acids, sterols, carotenoids, waxes and flavonoids.

Hydrocarbon Essential Oils consist of Chemical Compounds that have hydrogen and carbon as their building blocks. Basic Hydrocarbon found in plant is isoprene.

Terpenes

Generally have names ending in "ene." For examples: Limonene, Pinene, Piperene, Camphene, etc.Terpenes are anti-inflammatory, antiseptic, antiviral and bactericidal. Terpenes can be further categorized in monoterpenes, sesquiterpenes and diterpenes. Referring back to isoprene units under the Hydrocarbon heading when two of these isoprene units join head to tail, the result is a monoterpene when three join, it"s a sesquiterpene and four linked isoprene units are diterpenes.

Monoterpenes [C10H16]

Properties: Analgesic, Bactericidal, Expectorant and Stimulant.Monoterpenes are naturally occurring compounds, the majority being unsaturated hydrocarbons (C10). But some of their oxygenated derivatives such as alcohols, Ketones and carboxylic acids known as

monoterpenoids.The branched-chain C10 hydrocarbons comprises of two isoprene units and is widely distributed in nature with more than 400 naturally occurring monoterpenes identified.

Sesquiterpenes

Properties**:** anti-inflammatory, anti-septic, analgesic, anti-allergic.Sesquiterpenes is biogene tically derived from farensyl pyrophosphate and in structure may be linear, monocyclic or bicyclic. They constitute a very large group of secondary metabolites, some having been shown to be stress compounds formed as a result of disease or injury.

Sesquiterpene Lactones

Over 500 compounds of this group are known; they are particularly characteristics of the Composite, but do occur sporadically in other families. Not only have they proved to be of interest from chemical and chemotaxonomic viewpoints, but also possess many antitumor, anti-leukemia, cytotoxic and antimicrobial activities.

They can be responsible for skin allergies in humans and they can also act as insect feeding deterrents. Chemically the compounds can be classified according to their carboxylic skeletons. Thus, from the germacranolides can be derived the guaianolides, pseudoguaianolides, eudesmanolides, eremophilanolides, xanthanolides, etc.

Diterpenes

Properties: anti-fungal, expectorant, hormonal balancers, hypotensive .Diterpenes are made of up four isoprene units. This molecule is too heavy to allow for evaporation with steam in the distillation process, so is rarely found in distilled essential oils. Diterpenes occur in all plant families and consist of compounds having a C20 skeleton. There are about 2500 known diterpenes that belong to 20 major structural types. Plant hormones Gibberellins and phytol occurring as a side chain on chlorophyll are diterpenic derivatives. The biosynthesis occurs in plastids and interestingly mixtures of monoterpenes and diterpenes are the major constituents of plant resins. In a similar manner to monoterpenes, diterpenes arise from metabolism of geranyl geranyl pyrophosphate (GGPP).

◆ **Alcohols**

Anti-septic, anti-viral, bactericidal and germicidal. Alcohols are the compounds which contains Hydroxyl compounds. Alcohols exist naturally, either as a free compound or combined with a terpenes or ester. When terpenes are attached to an oxygen atom and hydrogen atom, the result is an alcohol. When the terpene is monoterpene, the resulting alcohol is called a monoterpenol. Alcohols have a very low or totally absent toxic reaction in the body or on the skin. Therefore, they are considered safe to use.

◆ **Aldehydes**

Properties: anti-fungal, anti-inflammatory, anti-septic, anti-viral, bactericidal, disinfectant, sedative. Medicinally, essential oils containing aldehydes are effective in treating Candida and other fungal infections disease.Example Citral in lemon, Lemongrass and lemon balm which has been used.

◆ **Acids**

Properties: anti-inflammatory. Organic acids in their free state are generally found in very small quantities within Essential oils. Plant acids act as components or buffer systems to control acidity.

◆ **Esters:**

Esters are formed through the reaction of alcohols with acids. Essential oils containing esters are used for their soothing, balancing effects. Because of the presence of alcohol, they are effective antimicrobial agents. Medicinally, esters are characterized as antifungal and sedative, with a balancing action on the nervous system. They generally are free from precautions with the exception of methyl salicylate found in birch and wintergreen which is toxic within the system.

◆ **Ketones**

Properties: anti-catarrhal, cell proliferant, expectorant, vulnery. Ketones often are found in plants that are used for upper respiratory complaints. They assist the flow of mucus and ease congestion. Essential oils containing ketones are beneficial for promoting wound healing and encouraging the formation of scar tissue. Ketones are usually (not always) very toxic**.** The most toxic ketone is Thujone found in mugwort, sage, tansy, thuja and wormwood oils. Other toxic ketones found in essential oils are pulegone in pennyroyal and pinocamphone in hyssops. Some non-toxic ketones are jasmone in jasmine oil, fenchone in fennel oil, carvone in spearmint and dill oil and menthone in peppermint oil.

Lactones:

Properties: anti-inflammatory, antiphlogistic, expectorant, febrifuge. Lactones are known to be particularly effective for their anti-inflammatory action, possibly by their role in the reduction of prostaglandin synthesis and expectorant actions. Lactones have an even stronger expectorant action then ketones (A, 2012)

2.9 Uniqueness of essential oils

In early work, the term essential oil was defined as the volatile oils obtained by the steam distillation of plants. This definition was clearly intended to make a distinction between "fatty oils" and the oils which are easily volatile. It was found that the oils contain many classes of organic substances with varying volatility. Although a list of all the known oil components would include a variety of chemically unrelated compounds. It is possible to classify these into main groups of essential oils (Guenther, 1960).

Terpenes, related to isoprene

Straight-chain compounds, not containing any side branches

Benzene derivatives

Essential oils are volatile:

Essential oils are the volatile fragrant components from various indigenous and exotic plants which have been traded internationally for several centuries (Becker, 2005).

Essential oils are aromatic:

Essential oils are highly aromatic and therefore, many of the benefits can be obtained by simply inhaling them. This can be done by breathing in the fragrance from the bottle or they can be diffused into the room.

Essential oils when diffused can be the best air filtration system in the world. They will purify the air by removing metallic particles and toxins from the air increase atmospheric oxygen; increase ozone and negative ions in the house which inhibits bacterial growth, destroy mold, cigarettes and animal odours; fill the air with a fresh herbal aromatic scent (Becker, 2005).

Essential oils have penetrating characteristics:

The penetrating characteristic of essential oils greatly enhances their ability to be effective. Essential oils will penetrate into the body when applied to the skin. Essential oils rubbed into the feet will be distributed to every cell in the body in minutes. They will even penetrate a finger or toe nail to treat fungal infection underneath. Other vegetable oils do not have this propensity to penetrate (Becker, 2005).

Pure essential oils have very high frequency:

The effectiveness of essential oils is sometimes also described in terms of frequency.It has been reported that the human body has an electrical frequency and that much about a person's health can be determined by frequency.Tainio has determined that the average frequency of the human body during the day time is 62-68 MHz (a healthybody frequency is 62-72). When the frequency drops, the immune system is compromised. If the frequency drops to 58 MHz, cold and flu symptoms appear, at 55 MHz, diseases like Candida take hold, at 52 MHz, Epstein bar and at 42 MHz, cancer. According to Dr. Royal R. Every disease has a frequency He found that certain frequencies can prevent the development of disease and that others would destroy disease. Substances with higher frequency will destroy diseases of a lower frequency. The study of frequencies raises important questions, concerning the frequencies of substances we eat breath and absorb. Many pollutants lower healthy frequency.

Processed canned food has a frequency of zero. Fresh produce has up to 27 MHz Essential oil start at 52 MHz and go as high as 320 MHz, which is the frequency of rose oil. Clinical research shows that essential oils have the highest frequency of any natural substance known to man, creating an environment in which disease; bacteria, virus, fungus, etc. cannot live (Becker, 2005).

2.10. Essential Oil Extraction Processes for Perfume Formulation

There are a few conventional and modern methods of extracting essential oils. It can be extracted by hydro-distillation, cold pressing, enfleurage, hydro-diffusion, supercritical fluid extraction, vapo-cracking,soxhlet extraction turbo-extractor and microwave extraction.

A. Hydro-distillation

Essential oils can be extracted by hydro-distillation; water, steam and water / steam distillation

i. Steam distillation

Many of the essential oils presently used in perfumery are obtained by steam distillation of flowers, leaves, bark; etc (John, 1982).Steam is widely used because of its high latent heat of evaporation, relatively cheaper and widely available. There are two types of steam distillation: water / steam distillation and steam distillation. This process involves the use of steam to percolate and vaporize out the essential oils from the plant material, with the subsequent condensation of steam and essential oil prior to their separation. It can be seen from the experimental work done that there is an art to distillation and that especially for low yield plants much skill is needed. The role of the distiller is to achieve oil as close as possible to the oil as it exists in the plant. During distillation, only very tiny molecules can evaporate so they are the only ones which leave the plant. These extremely small molecules make up an essential oil. The most advanced type of distillation is by direct steam provided from a separate boiler. The advantages of this type of "dry" steam distillation are that it is relatively rapid, therefore charging and emptying the still is much faster and energy consumption is lower. The rapid distillation is also less likely to damage those oils which contain reactive compounds, e.g. Esters. As a general rule all stills should be insulated ("lagged") to reduce heat losses. Their design and construction should also facilitate loading and unloading (Guenther, 1972).

ii. Water distillation

In this method the charge (which is usually comminuted) is immersed totally in water, which is boiled. The stills are of the simplest type and are used extensively by small holder producers of essential oils (Guenther, 1972). Often they are heated over an open fire. The disadvantages are that the heat is difficult to control and hence the rate of distillation is variable. Also the possibility exists for local overheating and "burning" of the charge which can lead to poorerquality oil. Improved distillation control can be obtained by using steam from a separate boiler, which is passed into jacket around the still or through a closed coil in the bottom of the still, to heat the contents of the still.

A further disadvantage of this system is that it requires the heating of a large quantity of water adding to costs and time needed for each distillation. However, it is necessary for the efficient distillation of certain woody materials e.g. sandalwood and cinnamon barks (Noor Azian, 2001).

iii. Water and Steam distillation

In water and steam distillation, the steam can be generated either in a satellite boiler or within the still, although separated from the plant material. Like water distillation, water and steam distillation is widely used in rural areas. Moreover, it does not require a great deal more capital expenditure than water distillation. Also, the equipment used is generally similar to that used in water distillation, but the plant material is supported above the boiling water on a perforated grid. In fact, it is common that persons performing water distillation eventually progress to water and steam distillation. It follows that once rural distillers have produced a few batches of oil by water distillation, they realize that the quality of oil is not very good because of its still notes (subdued aroma). As a result, some modifications are made. Using the same still, a perforated grid or plate is fashioned so that the plant material is raised above the water. This reduces the capacity of the still but affords a better quality of oil. If the amount of water is not sufficient to allow the completion of distillation, a cohobation tube is attached and condensate water is added back to the still manually, thereby ensuring that the water, which is being used as the steam source, will never run out. It is also believed that this will, to some extent, control the loss of dissolved oxygenated constituents in the condensate water because the re-used condensate water will allow it to become saturated with dissolved constituents, after which more oil will dissolve in it.

B. Cold Pressing

Cold pressed expression or scarification is used to obtain essential oils for the peels and seeds of citrus's, such as bergamot, grapefruit, lemon, lime, mandarin, orange and tangerine oils (Arnould, 1981). In this process, the outer layer of the fruit peel contains the oil are removed by scrubbing. Then the whole fruit is pressed to squeeze the juice from the pulp and to release the essential oil from the pouches. The essential oil rises to the surface of the juice and is separated from the juice by centrifugation.

C. Enffleurage

Enfleurage is an expensive process and is rarely used today except in a few places in France and India (Arnould, 1981).

Some flowers, such as jasmine or tuberose have very low Contents of essential oil and are extremely delicate; heating them would destroy the blossoms before releasing the essential oils (Furia, 1972). In such cases, enfleurage is sometimes used to remove the essential oils (Guenther, 1972). Flower petals are placed on solid sheets of warm which will absorb the flowers' essential oils. When all the fragrance is transferred from the flowers to the fat, they are removed and replaced with fresh ones (Billot and Wells, 1975). This process is repeated several times until the fat becomes saturated with the essential oil. A solvent most of the time alcohol is then added which separates the essential oil from the fatty substance. The alcohol will then evaporate leaving only the essential oil. This method is no longer
commercially viable (Poucher, 1974).

D. Hydro-diffusion

Although introduced more recently than carbon dioxide extraction, hydro-diffusion is similar to steam distillation except that the steam is produced above the plant material and percolates down through it (Chrissie, 1996). The advantage of hydro-diffusion over distillation is that the process is quicker, especially for fibrous material such as woods and barks. The resultant oils are reported to have a superior aroma and a richer colour obtained by ordinary distillation. Nevertheless, oils captured by hydro-diffusion process are not widely available.

E Soxhlet extraction method

Soxhlet extraction techniques involve solid/liquid contact for removing one or more chemical compounds from solid materials by dissolution in liquid reflux.

F. Supercritical Fluid Extraction (SFE)

Carbon dioxide is a new method of extraction using carbon dioxide gas, which is kept under high pressure and at a constant temperature (Kelly *et al*, 2002). Plants are placed in a stainless steel tank and as carbon dioxide is injected into the tank, pressure inside the tank builds. Under high pressure, the carbon dioxide turns into a liquid and acts as a solvent to extract the essential oils from the plants. When the pressure is decreased, the carbon dioxide returns to a gaseous state, leaving no residues behind. The equipment for this process is very expensive and so are the resulting oils. Carbon dioxide extractions have fresher, cleaner, and crisper aromas than steam distilled essential oils and they smell more similar to the living plants because high heat is not used.

This extraction method produces higher yield. Many essential oils that cannot be extracted by steam distillation can be obtainable with carbon dioxide extraction. Nevertheless, this technique is very expensive and it is not easily handled.

G. Vapo-cracking

This is a new continuous process, which is French patented (Martel, 1978) allows the recovery of the most volatile aromas directly from solids, like fruits, vegetables and other aromatic raw materials. As a first step in processing, either batch wise or continuously, it simultaneously replaces a press, particularly when that conventional process would yield very little juice. The main plant divides in two parts: Heating is obtained by direct steam injection by steam jacket and hollow screws or by any suitable range of radiations. Condensates may be recovered from that "bleaching" if some preliminary extraction has occurred. A flash chamber is kept under vacuum, where the pre-heated solids fall after passing a sluice. At this point 10 percent of the water content of the material is vaporized. The recovered condensates are remarkably rich in the most volatile aromatic fractions. They are further concentrated by continuous distillation or any other suitable process.

H. Solvent extraction

Solvent extraction uses very little heat so it's able to produce essential oils whose fragrance would otherwise be destroyed or altered during steam distillation. Solvent extraction is used on delicate plants to produce higher amounts of essential oils at a lower cost (Chrissie, 1996). In this process, a chemical solvent such as hexane is used to saturate the plant material and pull out the essential oils. The plant is removed and this renders a solvent. The solvent is then boiled off under a vacuum or in a centrifugal force machine to help separate it from the essential oil. Because the solvent has a lower boiling point than the essential oil it evaporates and the oil is left. The solvent is cooled back into liquid and reclaimed. Along with the essential oil, the fats, waxes and heavier oils can be extracted. This produces a substance called a concrete. The process continues by dissolving oils into warm alcohol. The alcohol is removed under vacuum and pure essential oil is left. Although more cost-efficient than enfleurage, solvent extraction is more expensive than steam distillation so it is reserved for costly oils which cannot be distilled. A solvent extracted essential oil is called an absolute.

I. Micro wave extraction

Microwave radiation interacts with dipoles of polar and polarizable materials. The coupled forces of electric and magnetic components change direction rapidly (2450 MHz). Polar molecules try to orient in the changing field direction and hence get heated. In non-polar solvents without polarizable groups, the heating is poor (dielectric absorption only because of atomic and electronic polarizations).

This thermal effect is practically instantaneous at the molecular level but limited to a small area and depth near the surface of the material. The rest of the material is heated by conduction. Thus, large particles or agglomerates of small particles cannot be heated uniformly,which is major drawback of microwave heating. It may be possible to use high power sources to increase the depth of penetration. However, microwave radiation exhibits an exponential decay once inside a microwave-absorbing solid.

In microwave-assisted extraction (MAE): 1) the heat of the microwave irradiation is directly transferred to the solid without absorption by the microwave-transparent solvent; 2) the intense heating of step 1 causes instantaneous heating of the residual microwave-absorbing moisture in the solid; 3) the heated moisture evaporates,creating a high vapor pressure; 4) the vapor pressure generated by the moisture breaks the cell; and 5) breakage of cell walls releases the oil trapped within it(Sukhdev Swami *.et al* ,2008).

2.11 Selection of Extraction Processes of Essential Oils

2.11.1 Soxhlet extraction method

Soxhlet extraction techniques involve solid/liquid contact for removing one or more chemical compounds from solid materials by dissolution in liquid reflux. In a conventional Soxhlet extractor, the solid material is put into the thimble of the extractor. It is gradually filled up with the extracting liquid phase by condensing the vapors from the distillation flask. When the solvent gets to a particular level, a siphon pulls the thimble contents into the distillation flask, thus carrying the extracts into the bulk liquid . The process is continued for the chosen contact time, and each is replicated. Moreover, has reported that many industrial applications require mathematical models for design and effective systems control. Models are simplified mathematical representations of systems at a particular point in time and intended to promote understanding of the real system [2]. Therefore, process modeling involves relating together the properties of a system that are influenced by the process.

The outcome is a set of mathematical equations, which is the process model . The process model comprises a set of mathematical formulations or equations that permit us to predict a chemical process's dynamics. Sometimes, to optimize or maximize process operating variables, engineers cannot choose the best operating variables that will minimize operating costs or maximize the profit of a chemical process plant. In a situation like this, the process model and appropriate economic information are used to analyze the prevailing situation and determine the most profitable process conditions . It is worthy to note that mathematical models are useful in developing scale-up procedures from laboratory scale up to pilot plant scale and then industrial scale-up allowing alternative strategies to evaluate the selection of the process variable conditions . In modeling with Microsoft Excel, different trendlines, including linear, polynomial (quadratic and cubic), exponential, logarithmic, and power regression models, can be obtained. But, Middleton [14] suggested that the exponential and power model transform data before the fit, resulting in inaccurate best fit and regression (R2). In addition, both power and exponential curves are used to fit data that increase or decrease at a high rate, and (SSRG International Journal of Chemical Engineering Research (SSRG-IJCER) - Volume 7 Issue 2 May to Aug 2020)neither curve can fit harmful data or data equal to zero. Much research work had been conducted on kinetic modeling of the steam distillation technique, but information regarding Soxhlet extraction modeling of lemongrass oil is scarce in the literature. Therefore, this study aims to formulate Soxhlet extraction models to help achieve maximum lemongrass oil yield by fitting the observed experimental data into the different regression models.

2.12 Raw material selection

Aromatic and Perfume Plants: There are number of indigenous and introduced aromatic and perfume plants in Ethiopia. Some of these are Commiphora spp,Boswellia spp, Cinnamomum cassia, Juniperus procera, Echinops spp, Olea europaea subsp. Africana, Otostegia spp, Ocimum spp, Artemisia spp,Cymbopogon citratus, Cyperus bulbosus, Myrtus communis (Genetic, 1996).

2.12.1. Lemongrass as raw material for perfumes.

Lemongrass oil which is used in a range of industrial products where a lemon flavor is required, can also provide citral that can be modified into β-ionone and methyl-ionone and serves as a starting precursor for vitamins A and E synthesis (Robbins 1983).Lemongrass is one of the best important perfumery material and flavoring. Large quantities of lemongrass generated as result of the high demand of flavor in worldwide. Most of which are native to south Asia, Southeast Asia and Australia. Two major types have considerable relevance for commercial use: East Indian lemongrass (cymbopogon flexuous*)* is native to India, Sri Lanka, Burma and Thailand, whereas the West Indian oil is extracted from cymbopogon citratus (Dc) stapf that is mainly cultivated in central and South America and also known in parts of Africa, south East Asia and the Indian Ocean islands. There are two types of lemongrass species that are cultivated in Ethiopia. Those are cymbopogon citratus and nardus that were experimentally recognized by the book of Ethiopian and Eritrea flora.

2.12.2. Chemical composition of lemongrass essential oil

The main chemical components of lemongrass oil are myrcene, citronellal, geranyl acetate, nerol, geraniol, neral and traces of limonene and citral.The major terpenes in C citratus include citral-α or geranial (10–48%) and citral-β or neral (3–43%), borneol (5%), geraniol (2.6–40%), geranyl acetate (0.1–3.0%), linalool (1.2–3.4%) and nerol (0.8–4.5%) (Akhila *et al*, 2010).

2.13 Production Process of essential oil from lemongrass

Lemon grass essential oil was extracted by soxhlet extraction of the dried leaves of lemon grass; the oil of lemon grass was yellow in colour with a citrus grass (lemon fragrance).

Drying

Lemon grass was obtained from garden, were dried as soon as possible so as to diminish the risk of fungal growth before oil extraction or mushroom colonization. Results were achieved at room temperature with exposure to direct sunlight and frequent spreading.

Size Reduction

The dried plant material was disintegrated by feeding it into a grinder that was used to reduce the size of the leaves into desired size. This can increase the yield of the oil during the process. Cutting of leaves enhance and help catalyze the production of the oil during the process. Furthermore, size reduction maximizes the surface area, which in turn enhances the mass transfer of active principle from plant material to the steam.

It is an advantage also because cutting can increase the quantity of the grass fed during the operation.

2.14 Perfume Formulation from lemongrass essential oil.

2.14.1 Production Process of perfume from lemongrass essential oil

Blending:

Once all of the oils for the perfume have been extracted, they need to be blended together. The oils are blended accorded to a formula that has been predetermined by a master in the perfume industry, often referred to as a "nose." One perfume can contain hundreds of different ingredients and can take multiple years to develop. Once the perfume oils have been blended, the scent is mixed with alcohol. The amount of alcohol added to the perfume oils varies depending on what the final product will be being. Eau de parfum will have a lower concentration of alcohol while scents such as body mists will have a much higher concentration of alcohol. Most perfumes are made of about 10-20% perfume oils dissolved in alcohol and a trace of water.

Aging:

Higher quality or fine perfumes are oftentimes aged for months or potentially even years after the scent has been blended. This is done to ensure that the proper scent has been created. Aging allows the different scents or notes to really blend together. Perfumes are made up of top notes that provide the scent with body as well as base notes which create an enduring fragrance.

3. Materials and Methods

The experimental work has been done in laboratory of wolkite university college of engineering and technology department of Chemical Engineering,Wolkite Ethiopia.

3.1 Materials and Equipment's

Materials used during extraction and characterization of lemongrass essential oil were:1000ml Separation funnel , 250ml and 1000 Beakers, Electronics weighting balance , Water bath, 2000ml Round bottom flask, Knife, Electric heater ,Distilled water, centrifugal miller, Scissors, aluminum foil, density separator, perfume bottle, centrifugal separator, oven, sieve,, etc.

3.2. Methods

3.2.1. Raw material preparation

Lemongrass (Cymbopogon Citratus) leaves were purchased from home garden in gurage zone at gubre town and farmer who live around church in south of wolkite ,

Ethiopia.

Figure 3.1 Lemon grass from a domestic backyard gubre

They were collected according to (Edwin *et al*., 2012).Some dirty material was removed. The plant material divided in two parts to do an experiment, on the varied the size particle of dried lemon grass. Fresh Cymbopogon citratus leaves were dried at room temperature for five days. Lastly it was kept in plastic bag after drying, and for both size reductions were done based on required size, then experiment was done by putting it into still to extract the oil based on the weight size which was decided.

3.2.2. Determination of moisture content of the lemongrass

Fresh lemon grass was collected on April 2023; after it was collected half of them were dried by sun for 5 days. According to Buggle *et al*. 1997 carried out an experiments to study the effects of drying temperature on the amount and quality of essential oils extracted from C. citratus and Leaf blades were cut into small parts (about 1 –1.5cm in length) and dried for several days and grinded about 0.2-5mm, until a constant weight was achieved the higher amount of oil was collected at lower size particles. So drying atmospheric temperature is good. two sample of the lemon grass randomly were weighted for small size 60, 58.7 grams and two samples for large size of dried lemon grass 60, 58.7 grams then dried in oven at 1050C and the weight was measured every two hours. The procedure was repeated until a constant weight was obtained and the percentage moisture content of the lemon grass was determined (General, 2005). The moisture content of the sample was determined using equation (3.1).

$$moisture\% = \frac{w1 - w2}{w1}$$

Where, W1 = Original weight of the sample before drying

W2 = weight of the sample after drying

3.2.3. Size reduction

After the moisture was removed by sundry then cut with scissors for size of 20mm and 30mmand it was milled in crush miller for dry with sieve size 0.1-5mm.

3.2.4 Essential Oil extraction

3.2.4.1. Soxhlet extraction of essential oil from lemon grass

240gm dry sample of lemongrass will be weighed and placed in a 500ml clean flat bottom flask. Next, 1200ml N- hexane solvent will be poured into the 500ml flask and stopped. The flask and content will be allowed to stand for 4hrs; this will be done to extract all the oil content in the lemongrass and for complete extraction. After which the extract will be decanted into another 500ml beaker. 100ml of Ethanol will be added to extract the essential oil since essential oil is soluble in Ethanol. Then, the mixture will be transferred to 500ml separating funnel and separated by liquid/liquid separation process. Finally, The content of the separating funnel will be allowed to come to equilibrium, which separated into two layers (depending on their different density).The lower Ethanol extract and the upper Hexane layer will be collected into two separate 250ml beaker and will be placed in a water bath at 78oC.

This will be done to remove the Ethanol leaving only the natural essential oil.

Figure 3.2: soxhlet extractor

3.2.4.2.3. Determination of percentage of oil extracted

The percentage yield was calculated in two forms i.e. oil yield and extraction yield using the formula below.

Since lemongrass have an oil content of 1-2%, average 1.5% oil content was taken for calculating the yield, Therefore:-

$$percentage\,oilyeild = \frac{massoil}{0.015 * massofsample} * 100$$

3.3. Characterization of essential oil extraction from lemongrass

Materials used during extraction and characterization of lemongrass essential oil were:500ml Separation funnel , 250ml and 500 Beakers, Electronics weighting balance , Water bath, 500ml Round bottom flask, Knife, Electric heater ,Distilled water, , centrifugal miller, Scissors, aluminum foil, density separator,, centrifugal separator, and etc

3.3.1 To develop perfume and characterize physical property using sensor analysis.

Materials that used during perfume development and sensor analysis were: Micro-Pipette, Funnel, 50ml and 120ml beakers, perfume bottle, shaker, centrifuge separator, (glycerin and castor oil), carrier solvent ethanol (96%), Distilled water, Lemon grass essential oil,panalists, paper and pen.

3.3.2 Characterization of essential oil extracted from lemongrass

The specific gravity at 250C, density, viscosity, PH value, was analyzed using standard procedures. The result was compared with standard lemongrass oil. The acid value.

3.3.4. Determination of some of physical properties of lemongrass oil

3.3.4.1. Determination of PH

Two gm of the sample was taken and putted in a clean dry 25ml beaker and 13ml of hot distilled water was added to the sample in the beaker and stirred slowly. And then it was cooled in cold water bath to 250C.The PH electrode was standardized with a buffer solution first and then immersed in the sample and the PH was read (A.O.A.C official method analysis 960.19, 2000)

3.3.4.2. Determination of boiling temperature of the essential oil

0.5ml of lemon grass oil poured in to beaker and a thermometer was inserted and placed on a heating mantle, it was observed that the oil in the beaker started circulating leading to boiling of oil and read temperature on thermometer.

3.3.4.3.Determination of moisture and volatile matter of essential oil

Moisture content of oils and fats is the loss in mass of the sample on heating at 105 0C under operating conditions specified.0.5gm of oil was weighted and putted in a dish and then dried in an oven at 1050C for 1hour.The dish was removed from the oven and cooled in a dissector and weighed. The process was repeated until a constant weight was observed and the moisture and volatile matter of the oil was determined (Hand book of food analysis, 1984)

3.3.4.4. Determination of the specific gravity

The density of the oil was determined by using density bottle. A clean and dry bottle of 25ml capacity was weighed (W0) and then the bottle was filled with the oil, stopper inserted and reweighed to give (W1).

The oil was substituted with water after washing and drying the bottle and weighed to give (W2) (General, 2005).The expression for specific gravity (Sp.gr) is

$$sp.gr = \frac{(x2 - x1)}{(x3 - x1)} = \frac{massofsamp\,le}{massofaneq\,ualvolumeo\,fwater}$$

3.4. Characterization of the chemical properties of lemongrass oil

3.4.1. Acid value

25ml of toluene and 25ml of ethanol was mixed in a 250ml beaker. The resulting mixture was added to 0.5gm of oil in a 250ml conical flask and few drops of phenolphthalein were added to the mixture. The mixture was titrated with 0.1M KOH to the end point with consistent shaking for which a dark pink colour was observed and the volume of 0.1M KOH (V0) was noted. The Acid value was calculated as (General *et al*., 2005)

$$Acidvalue = \frac{56.1 * V * N}{M}$$

Where V = Volume in ml of potassium hydroxide solution,

N = Normality of the potassium hydroxide solution,

M = Mass in gm of the material taken for the test.

3.5. To develop perfume and characterize physical property using sensor analysis

3.5.1. Perfume development from lemongrass essential oil

Procedure:

Lemon grass essential oil extract was measured and placed in 120ml beaker with measured containing carrier solvent of ethanol then it was shaked by shaker and again fixative was added in solution mixture to improve the longevity of the perfume. In addition to that distilled water was also added to the mixture to homogenized undissolved ingredient. The mixture was poured into black bottle then it was placed in the dark area to concentrate the perfume for two week. Finally it was filled in the bottle of perfume.

4. Results and Discussion

4.1 Characterization of essential oil extraction from lemongrass

Table 4.1 Moisture content of dry lemongrass

	Sample weight in gram	
	Sample 1	Sample 2
Initial mass	60	58.7
Mass after 2hr	58.7	57
Mass after 4hr	57	56.12
Mass after 6hr	56.12	54.17
Mass after 8hr	54.17	54.1
Mass after 10hr	54.1	54.1
Final moisture content	9.83%	7.8%

Table 4.1 Moisture content of dry lemongrass

The moisture content of the two lemongrass sample having mass of 60 ,58.7 gm were 9.83, and 7.8 percent respectively. The mean average moisture content of the two samples will be 8.815%.The result obtained was different from Vázquez-briones *et al*., 2015 which moisture content of the dried C.citratus 9.14 ± 0.70% due to geographical variation of it.

4.1.1.2 Essential Oil extraction

4.1.1.2.1 Percentage yield of extraction for dry lemongrass

In this section outline of the results obtained from soxhlet extraction lemongrass essential oil experiments are discussed.

The percentage extraction yield was calculated by using equation 3.3 and the result is summarized in the table 4.2 below.

Factors			
Time(minutes)	Particle size(mm)	Average(grams)	Extraction yield(%)
150	0.1-2.5	0.711	0.79
	2.5-5	0.675	0.75
180	0.1-2.5	0.621`	0.69
	2.5-5	0.585	0.65

Two factors were selected because of uncontrollable of equipment especially temperature. Even if temperature was not taken as factor, the recommended temperature was around boiling point (Guenther, 1972). Due to that the extraction temperature was 94.50C and water boil at 92.5 0C in wolkite. So it was not that much difference based on the recommended. Based on that the maximum extraction of lemongrass oil was 0.79% at particle size ranges from 0.1-2.5mm for the extraction time of 2.5 hours and the minimum yield obtained was 0.65 % at maximum Particle size and maximum extraction time. According to Boruah *et al.*1995 Healthy and uninfected leaves yielded 0.80% oil, while leaves with a disease index of (60–75%) yielded 0.50% oil.From that what was concluded that extraction of essential oil high from health leave, properly prepared and uninfected by micro bacteria.

4.1.1.3 Effect of process parameters in percentage essential oil yield of lemongrass

4.1.1.3.1 Effect of process parameters in percentage essential oil yield of dry lemongrass.

- Effect of extraction time on percent yield of essential oil.

Effect of Extraction time Figure 4.1 shows the influence of extraction time on the extraction yields of Lemongrass (Cymbopogon Citratus) over the range 150-180 min under a fixed temperature and different raw material size. As being depicted in figure.4.1 most of the oil was being extracted at 150 minutes and the amount of yield obtained up to this period was 0.79%. In general, the rate of extraction was high at the beginning of the extraction but get slow gradually by time. These results confirmed the Fick"s second law of diffusion which stated about the final equilibrium achieved by the solute concentrations in plant matrix and in the solvent after a certain time.

This caused into no significant improvement in oil yield when prolonging the extraction time.

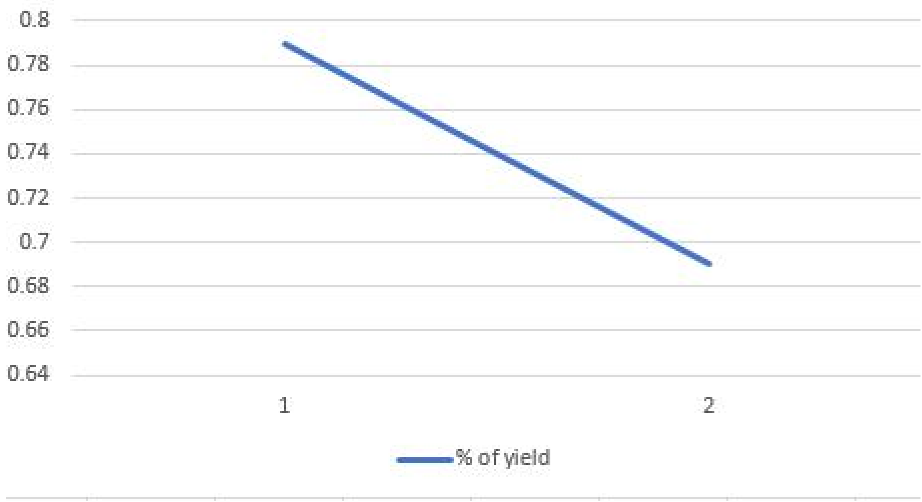


Figure 4.1 The effect of time on lemongrass oil yield at particle size (a) 0.1-2.5mm, b) 2.5-5mm

- Effect of particle size on percent yield of lemongrass of essential oil.

Particle size plays a great role on the yield of lemongrass oil. Smaller the size gives high yield while samples with large particle size deliver low yield.

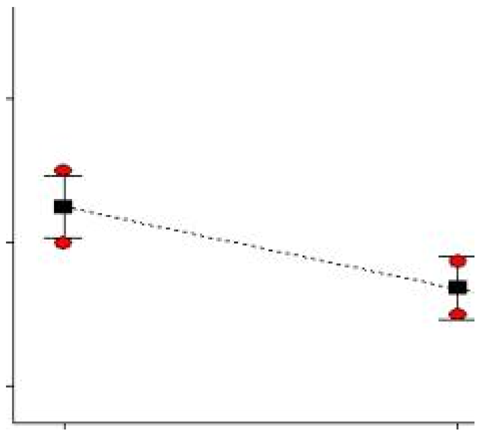


Figure 4.2 effect of particle size on the percentage of essential oil yield.

From above figure 4.2 what we observed less essential oil is extracted from the larger particles (>2.5mm) compared to the small size of the particles. The reason is that larger particles with smaller contact surface area have more resistant to steam entrance and carried out. Therefore, less amount of oil will be transferred from inside to the surrounding vapor in comparison with the smaller ones. Thus, an increase in particle size will decrease the oil yield. Nevertheless, we know that when the particle is too small (very fine chopped) , the extracted oil become small in its amount, even though the contact surface area for small particle is supposed to be significantly higher than that for the larger particles. This May be due to the agglomeration of the fine particles which reduces the effective surface area available for the free flow of steam towards inside the solid particles (Spider sayyar *et al* 2009) reported the result of extraction of oil from jatropha seed for three different particle size range<0.5mm, 0.5-0.75mm and >0.75mm.The highest percentage of oil yield was obtained with the intermediate particle size (0.5-0.75) which indicate that decreasing the particle size below certain particle size doesn't increase the percentage of oil yield and may even decrease the yield.

4.1.2 Characterization of essential oil extracted from lemongrass

4.1.2.1 Determination of some physical properties of the lemongrass oil

4.1.2.1.1 Moisture and volatile matter of essential oil

The moisture and volatile matter of the oil determined by oven method 0.5gm of oil was taken and put in oven and the weight was recorded at 2hr and 4hr .

The result obtained is summarized in the table below.

Table 4.3 moisture and volatility of essential oil

Time (hr)	0	2	4	dry
Weight(0.5gm)	0.5	0.49899	0.49899	0.202%

From equation (3.1), moisture and volatility of essential oil was determined.Then 0.5-0.49899 =0.0101gm. W is weight in gm of oil taken for the test=0.5gm, Substituting the above values in the equation

$$moisture and volatile matter = \frac{initial - final weight}{initial weight} = 0.2015\%$$

4.1.2.1.2 Kinematic viscosity dry extracted oil

Kinematic viscosity dry lemongrass extracted oil

Dynamic viscosity of oil which was red from vibro viscometer was 1.72mpa.s at temperature 200c, substituting the dynamic viscosity oil =1.72mpa.s=1.72× 10−3 kg/m.s and density of oil =881.25 kg/m3

$$kinematic viscosity = \frac{1.72 * 10\text{^}{-3}\,\mathrm{kg/ms}}{881.25\,\mathrm{kg/m\text{^}3}} = 1.95173 * 10\text{^}{-6} m\text{^}2 / s$$

Therefore the kinematic viscosity of the lemongrass oil was 1.95173× 10−6m2 /s

4.1.2.1.3 PH value of lemongrass oil

The PH value of oil was determined by PH electrode as measuring experiment

Table 4.4 The PH value of lemongrass oil was triplicated and the results obtained

Type of product	run			average
	1	2	3	
PH value of dry lemongrass extracted oil	3.98	4.200	4.21	4.13

Therefore the PH value of lemongrass oil was 4.13. In the preparation of perfume the PH value is in the range of 3.5-6.5 (Mueller *et al*,2000).The obtained PH value of lemongrass oil is in the range of commercially recommended.

4.1.2.2 Determination of chemical properties of lemongrass essential oil

4.1.2.2.1 Acid value of the oil

Acid value is the measure of total acidity of the lipid involving contributions from all the constituent fatty acids that make up the glyceride molecule (Ekpa and Ekpe, 1995). Titration method was used to determine the acid value. 25ml of toluene and 25ml of ethanol was mixed in a 250ml beaker. The resulting mixture was added to 0.5g of oil in a 250ml conical flask and few drops of phenolphthalein were added to the mixture. The mixture was titrated with 0.1M KOH to the end point with consistent shaking for which a dark pink colour was observed and the volume 1.3 and 1.8 of 0.1M KOH (V0) was noted for extracted oil from dry and fresh lemongrass.

Table 4.5 Acid value of essential oil lemongrass

Raw material condition	The volume of KOH for The sample (ml)	Mass of sample (gm)	Acid
Dry	1.3	2	18.23

4.2 Result of perfume proportion

Perfume oils usually contain tens to hundreds of ingredients and these are typically organized in a perfume for the specific role they play. Here only one ingredient used which is lemongrass essential oil instead of ten to hundreds ingredient what was used to develop perfume.

Table 4.6 perfume proportion from lemongrass essential oil

Type of perfume	Fragrance	Carrier solvent	Fixative	Diluent
Esprit de parfum	Lemongrass oil	Ethanol (96%)	Glycerin	D.water
Proportion of the perfume				
	20% lemon grass oil	75% ethanol	0.33% of glycerin	4.67%

These ingredients grouped into Primary scents (Heart): which consisted of one main ingredient such as lemongrass essential oil. Alternatively, multiple ingredients can be used together to create an abstract primary scent that does not bear a resemblance to a natural ingredient. For instance, jasmine and rose scents are commonly blended for abstract floral fragrances.But, here only lemongrass essential oil used to develop perfume. Blenders: are a large group of ingredients that smooth out the transitions of a perfume between different layers or bases. These blending ingredients are linalool which existed in the oil by itself. Fixatives: They used to support the primary scent by bolstering it in addition to elongate its aroma. Using the above proportion, perfume was developed based on the procedure which was stated in chapter three. After it was developed, sensor analysis carried out.

4.3 Result of the Sensory Evaluation of perfume

Sensory quality is the ultimate measure of product quality and success. Sensory analysis comprises a variety of powerful and sensitive tools to measure human responses to products (perfume). Appropriate application of these tests enables specific product and consumer insights and interpretation of volatile compound analyses to aroma perception. Results of sensory evaluation in terms of sensory attributes such as colour, aroma and overall acceptability are presented in table 4.20 below.

Table 4.7 Sensory analysis results of perfume in terms of aroma, color and acceptability

Each attributes	scored of each attributes based on ten panelist rating								
	1	2	3	4	5	6	7	8	9
color								2	8
acceptability							7	2	1
aroma							1	8	1
Each attributes	weight in percent for each attributes								
	1	2	3	4	5	6	7	8	9
color								20	80
acceptability							70	20	10
aroma							10	80	10

Hedonic scales: 1 = dislike extremely, 2 = dislike very much, 3 = dislike moderately, 4 = dislike slightly, 5 = neither like nor dislike, 6 = like slightly,7 = like moderately,8 = like very much,9 = like extremely.

According to the sensory analysis results in Table 4.20 showed that, about 80% of the panelists preferred like very much of aroma sample of the perfume at concentration of 20% essential oil developed. The results of sensory evaluation showed that perfume can be developed from lemongrass essential oil.

5.1 Material, energy balance and cost estimation

5.1.1 Material and energy balance

5.1.1.1 Material balance

Assumption: lemongrass collected on the harvesting period based on literature cited.

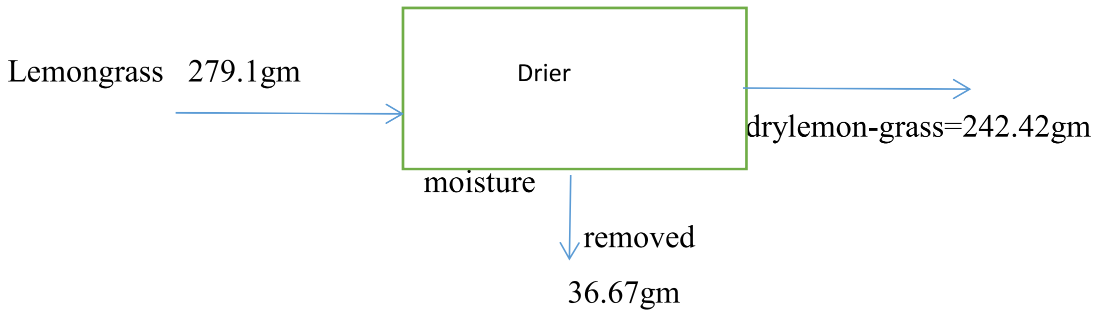
Note: All material balances had performed based on the experimental work in the laboratory. Material balances are nothing more than the application of the conservation law for mass According to Treybal (1981), matter is neither created nor destroyed.

Accumulation=Output + Consumption – Input – Generation (5.1)

Since there is no reaction, the generation and consumption terms are zero, no accumulation Total production feed for plant can be planted because we do have available climate condition and land for production of lemongrass but today the amount of lemongrass could not known how much we have. Even if it was not known but the people of Ethiopian have been using this herbal grass for anniversary ceremony to decorate the house and church. Only by 2015 Ethiopia imports 98,931 kg perfume from different countries to satisfy the demand of the customers (Ethiopian revenue and custom authority, 2015).So that perfume can be developed by substituted our indigenous raw material that used for perfume formulation. From this assume 40% for women. Assume if 35% will be substituted. As we know the concentration of fragrance perfume for women from 15-30%. Assume 20% concentrated. Then to produce 98,931kg of perfume, it needs 19,786.2 kg of fragrance required. Now the amount of land required to produce this essential oil 1,413.3 hectare needed. So let substitute 35% from the total import. The required essential oil needed 6990.47kg kg and the amount of land would be 285.3 hectare.

5.1.1.1.1 Material balance for dry lemongras

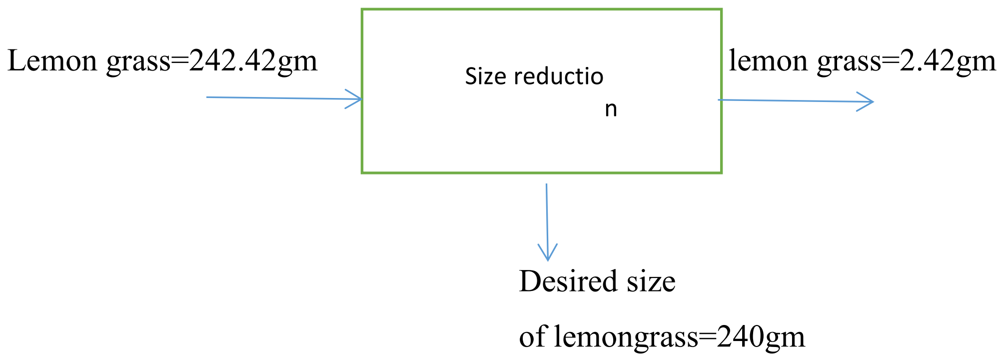
Drying, from experiment 13.29% moisture removed from 1kg lemongrass.



Balance on size reduction Machine

Note: assuming there is 1% of lemongrass loss .Then, the amount of raw material left to the

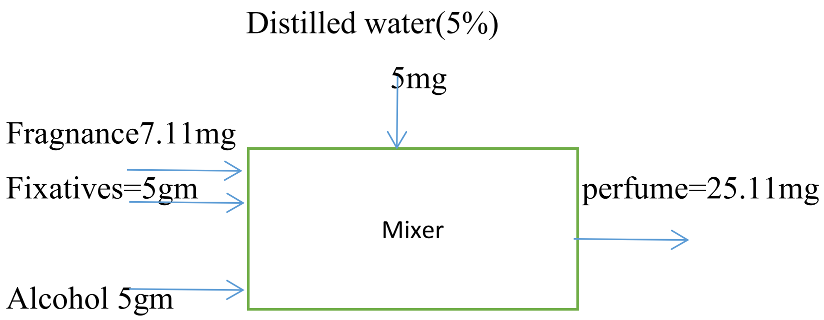
extraction vessel within required size.



5.1.1.1.2 Material balance in perfume formulation

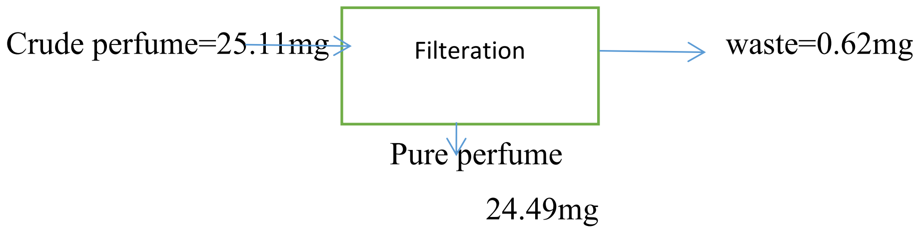
Mixing machine

Note: let 20% concentrated of perfume and 75%of alcohol



Filtration

Note: removal of undiluted ingredient 1%



5.1.1.1.3 Energy Balance on soxhlet extraction of oil from dry lemongrass

This energy balance was specifically calculated only for the water in the System. It showed that heat was absorbed into the system when boiling water and later released when water turns to vapor. Equation 5.2 shows the formula to calculate heat absorbed by the system:

$$QA = mCP\Delta T \tag{5.2}$$

Where, QA = heat absorbed by the system (kJ), m=mass of hexane (kg), Cp= heat capacity (kJ/kg. K), ΔT = temperature difference (0C)

At the end of the experiment there was 10 ml of water distilled out with lemongrass oil, Initial temperature 250 c, and final temperature 720c

QA = 50gm x 0.657 (KJ/kg.K) x (345 – 298) K = 1543.95 KJ

The heat released from the system, when water turns to vapor is illustrated in equation 5.3

$$QR = n\,\lambda \tag{5.3}$$

Where **n**=Number of moles of hexane (mol/g), λ =40. 656 x 103 J/mol

In this study, the experiment was carried out at atmospheric pressure (1atm). The heat capacity of steam at the constant pressure is 4.184kJ /kg.K (Perry and Green, 1997). The initial temperature of the system is 250C (298K) and the final temperature is 720C (367.5K). *QR* = 50g x (1mol / 86.17g) x 40.656E3 (J/mol) =23.590KJ

By using the data given, the total energy used by the system in this process, Qs is as follows:

$$QS = QA+QR \tag{5.4}$$

= 1543.95 KJ +23.590KJ= 1567.54KJ Where, QS= Energy used by the system in this process (kJ)

Where, QS= Energy used by the system in this process (kJ)

The total energy supplied into the system during the experiment, (kW.hr) is as follows:

$$Qp = \frac{1KWH}{3600KJ} * QS$$

$$QP = \frac{1KWH}{3600KJ} * 1567.54KJ = 0.435KWH$$

- **Sizing of major equipment**

Sizing and Equipment Listing For the Production of Perfume The engineer developing a process design must be the responsibility of preparing specific cation for individual places of equipment and design data must be developed. Safety factor: is a factor to represent the amount of our design that would be used to account for the changes in the operating performance with time. As a general, safety factor = (10 20) % of capacity of the equipment.

5.1.1.2 Economic analysis of perfume formulation from lemongrass additive

5.1.1.2.1 Building, equipment and manpower requirements

Plant parameters

Capacity per year = 34,602.82g/yr

Number of shifts /day = 2

Working days/year = 300 day

Machinery and equipment

Purchased cost for some basic plant

equipment‟s are estimated from

www.mhhe.com/engcs/chemical/peters/data/, and www. Matche/equipcost/index.com.

Table 5.1 Specification and purchased equipment cost.

Purchased equipment	Capacity	Qty	Material	Total cost (birr)
Rotary Cutter	2kg/s	1	_	267,653.5
Boiler	_	1	Stainless steel	355395
Soxhlet	_	1	Stainless steel	350000
Filter gravity	1mm2	1	Carbon steel	255,850
Condesor	0.07mm3/s	1	Carbon steel	179095
Total cost				1407993.5

Manpower requirement

Table 5.2 manpower requirement & cost

Sr.No	Manpower	Req. No	Monthly Salary (Birr)	Annual Salary (Birr)
1	General manager	1	5000	60000
2	Secretary	2	2000	48,000
3	Accountant	2	2500	60,000
4	Production and Technical Head	1	3000	36,000
5	Mechanic	2	2000	48,000
6	Electrical	2	2000	48,000
7	Store keeper	2	1800	43,200
8	Quality control	2	2500	60,000
9	Operators	6	2000	144,000
10	Ass. Operators	3	1800	64,800
11	Guards	2	1500	36,000
Total				648,000

5.1.2.2 Cost estimation

Cost of raw materials

Table 5.3 Cost of raw materials

Particulars	Unit price	Quantity per annum	Total cost(birr)
Lemon grass	5birr/gm	24gm	120
Ethanol	5.95birr/ml	200ml	1190
Water	0.05Bir/l	110L	55
Fixatives	10/ml	10ml	100
Distilled water	0.07/ml	10ml	0.7
Total raw material cost			1465.7

6. Conclusion and Recommendations

6.1 Conclusion

This work was intended to study the influence of different factors (Extraction timeand Particle sizes,) on the quality and quantity of extraction of essential oil from dried small particle size and raw material of lemongrass that used for perfume development. Variability of these operating conditions is the pre-dominant factors for the quantity of the oil. There are different methods of essential oil extraction from lemongrass. In this thesis, soxhlet extraction was used due to availability of the equipment. The percentage essential oil yield in dried small particle size(.1-2.5)mm samples is higher as compared to large particle size(2.5-5)mm samples. Characterization of essential oil was determined to see its quality. The citral was the main component of essential oil which has aroma that used to develop perfume. The maximum oil yield obtained was 0.79% for dried small particle size and 0.65% for large particle size lemon grass at the particle size of 2.5-5mm and the extraction time of 2.5 hours, the observed quantitative difference in the quantity of the oil was due to particle size and extraction time variability.Thus, determination of the appropriate size of the particles and optimal time needs to have a consideration to get the maximum amount of the required product for development of perfume. Finally perfume was developed from essential oil of tej-sar and sensor analysis was conducted to see its quality of aroma and the result was acceptable.

6.2 Recommendation

- Sedimentation analysis should be carried out on the yield obtained so that the approximate weight of the oil and water can be determined.
- It also recommends that formulation of perfume from lemongrass and other aromatic plant should have to be carried out to see the intensity and quality of it.
- We also recommend that in the formulation of perfume, the appropriate quantity of essential oil and other fixatives should be ensured to avoid skin reaction and to increase its intensity
- Here also recommend that the project to be implemented after detail feasibility studies.

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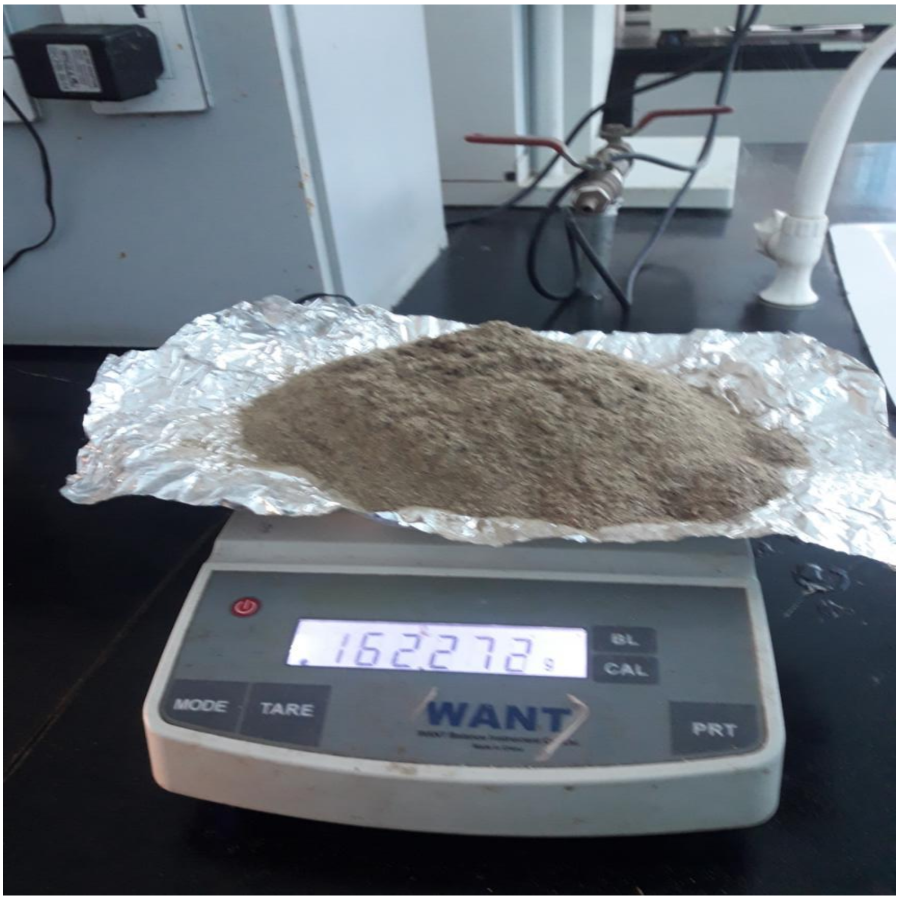
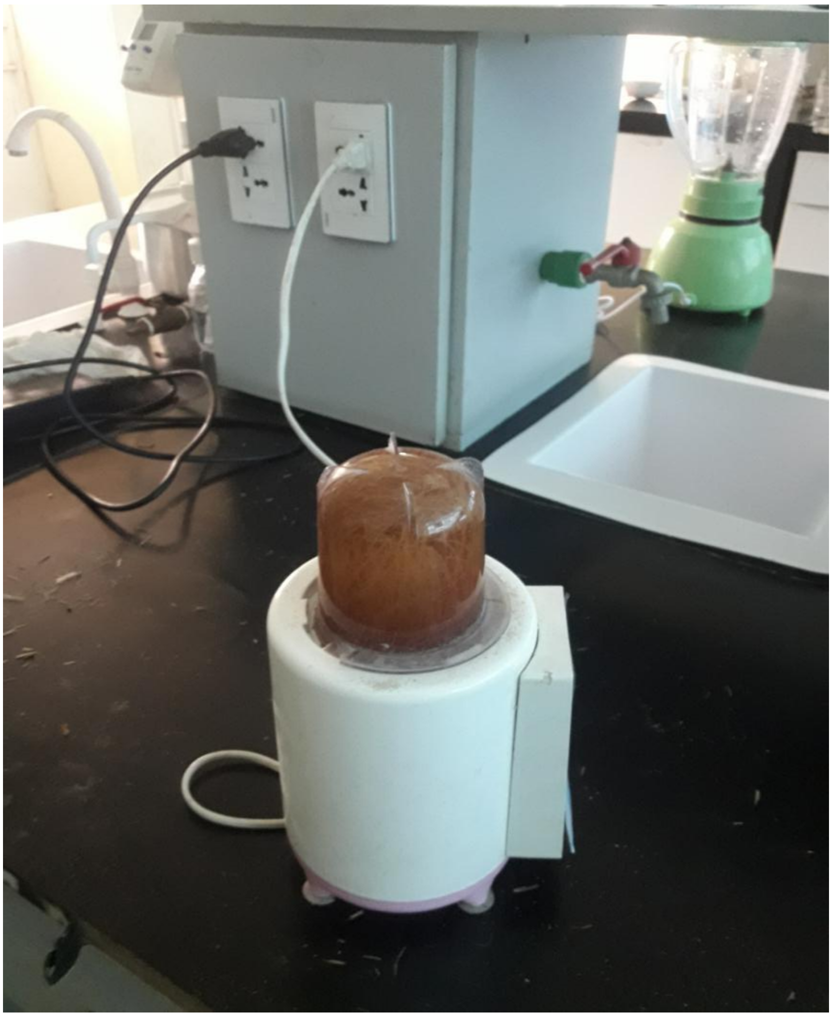
Appendix D

Laboratory equipment and samples photo

D1 Soxhlet extractor



D2: Balance



D3:Grinder

D4:Lemon grass powder


STSJ-5 High Frequency Sieve Shaker
Zhejiang Tugong Instrument Co., Ltd

D5:sieve