Development of a Water Disinfection Container Incorporating a Thermochromic Indicator

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Institute of Environmental Sciences and Engineering (IESE)

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APPROVAL SHEET

Certified that the contents and form of thesis entitled “Development of water disinfection container incorporating a thermochromic indicator”submitted by Ms Azra Nawar, Mariam Mahmood and Sarah Shahid have been found satisfactory for the requirement of the degree.

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ABSTRACT

The provision of safe and clean drinking water is of paramount importance for human health. More than one tenth of global population does not have access to safe drinking water (WHO/UNICEF, 2012).

Therefore, this research was focused on the design of water disinfection container for purification of water, using the minimum energy. For this purpose, a thermochromic strip was used as an indicator of the water temperature.

Microbial analyses have indicated that heating tap water up to 65 to 70^0C is effective in killing pathogenic microbes. Based on this, a thermochromic strip was designed to indicate the water temperature by changing color when it reaches the aforementioned range.

Laboratory synthesis of thermochromic paste was carried out by the method of Hughes (Hughes, 1998) with a few modifications. Using the correct chemical constituents the thermochromic paste was prepared and evenly spread on thin layer of filter paper. In the initial laboratory experimentation, the strip was observed to change color within the temperature range as specified above.

As a next step to this research work a suitable container was designed and fabricated, which would incorporate the thermochromic strip; an indicator of the change in water temperature. An aluminum container was designed with a special copper plate to mount the thermochromic strip. Experimental tests were performed which demonstrated the color change at a temperature of 65-70 ^{0}C accurately.

ACKNOWLEDGEMENTS

We extend our sincere gratitude to our supervisor *Prof. Dr.Ishtiaq A. Qazi* (Associate Dean, IESE) for guiding us through our project research. Without his help, we would not have achieved so much. Under his guidance we learned to work as a group and focus on different aspects of the project from chemical composition of the strip to designing and manufacturing of the container.

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Azra Nawar

Mariam Mahmood

Sarah Shahid

TABLE OF CONTENTS

List of Abbreviations

BGLB	Brilliant Green Lactose Broth
CFU	Colony Forming Unit
EC	Escherichia Coli
LTB	Lactose Tryptose Broth
MPN	Most Probable Number
MF	Membrane Filtration
SPC	Spread Plate Count
UNICEF	United Nations Children Fund
WAPIS	Water Purification Indicator Strip

List of Tables

List of Figures

Chapter 1

1 Introduction

1.1 Background

According to United Nations Children Fund (UNICEF) about 60% of rural families and 23% of urban families in developing countries are without safe drinking water and in some areas the water supplies may be heavily contaminated with pathogenic organisms (Hammer, 1996).

Diarrhea, a common disease of children in the developing countries, is caused by the use of contaminated water. The resulting dehydration is the leading cause of death in children, under the age of five, killing an estimated five million children annually (Andreatta, 1994).

Boiling can be used as a method of water disinfection. Bringing water to the boil is effective in killing or deactivating most bacteria and pathogens. Boiling is the most certain way of killing nearly all microorganisms. But boiling has its disadvantages like scalding, scaling of boiling vessels, removal of useful minerals and blandness of taste.

It is generally known that heating water at 65^0C for approximately six minutes will kill most of the pathogens (Andreatta, 1994). This process is referred to as pasteurization. This is carried out at a lower temperature than boiling, thereby making the whole process energy efficient.

Thermochromism: the ability of substance to change color due to change in temperature has been in use for quite some time now and has been applied in the form of paste on cups, pieces of jewellery, watches etc. Thermochromic pigment is a good visual indicator of

temperature change. By using the right chemicals and their right constituencies, a specific temperature range for color change can be achieved.

To design a suitable container various factors like thermal conductivity, economic feasibility etc. need to be taken into account. Aluminum is light, cheap and is also a good conductor of heat. It is sensitive to slight temperature change and is therefore able to conduct heat released by water at 65-70^{0}C efficiently.

1.2 Significance of Thermochromic Strip

Heating water to a lower temperature range would minimize the effects of scaling and blandness of taste. The main significance lies in the whole process being relatively less energy consuming than when water is put to boiling.

The development of an indicator strip will serve the purpose of indicating when the water reaches the temperature of 65^{0}C. This strip will work on the principle of thermochromism. An apparent color change from red to dark brown will take place between the temperature ranges of 65^{0}C to 70^{0}C.

It will be beneficial to devise a container, for heating water, which is energy efficient and less costly. The container will hold the strip in place thereby providing ease to the user. This energy efficient aluminum box, with the copper sheet incorporated within will be for long-term use and benefit. Also it is a cost-effective solution to problems associated with potable clean drinking water.

1.3 Aim

To design a water disinfection container with high energy efficiency and ease of use while incorporating a thermochromic indicator strip.

1.4 Objectives

- To carry out literature review
- Select appropriate chemical composition for the preparation of thermochromic pigment
- Perform initial experiments to verify the composition used for the preparation of the pigment
- Prepare a final thermochromic strip design based on initial experimentation
- Manufacturing an appropriate container to incorporate the strip
- Final experimentation.
- Report writing

1.5 Thesis Structure

Chapter 1 details the aim, objectives and thesis structure. Chapter 2 focuses on literature review. Chapter 3 details the materials and methods used to synthesize the thermochromic strip and the bacterial test methods employed. Chapter 4 explains the results obtained by three different techniques of microbial testing. Chapter 5 describes the designing, manufacturing and testing of the container for thermochromic strip and Chapter 6 gives the conclusions and recommendation.

Chapter 2

2 Literature Review

2.1 Water Disinfection

2.1.1 Background

The development of humanity has been tied in, to a large degree, with the state of health of the various groups that have inhabited the planet. On occasion, entire countries or regions have been decimated by pests and plagues that are often random, temporary and unique. Even so, there are diseases that appear to be as old as mankind itself, whose force and importance are a part of everyday life: the diarrheal diseases.

The edition of the *"World Health Report"* published by the World Health Organization (WHO, 2000) ranks diarrhea as the seventh cause of death in the world following heart disease, cerebrovascular accidents (strokes), acute respiratory infections, HIV/AIDS, chronic pulmonary obstructions and adverse prenatal conditions. While this ranking gives an idea of the relative importance of these causes of death, the finding of the Organization that diarrhea is by far the foremost cause of morbidity in human beings, being responsible for four billion cases a year, is much more significant. It is estimated that at any given time, almost one-half of the developing world's population is suffering from bouts of diarrhea.

Unfortunately, because of their longstanding presence in the lives of human beings, the scope and impact of diarrheal diseases on the health and quality of life of individuals and the economy of mankind as a whole tend to be overlooked.

Diarrhea can be traced to the existence of deficient nutrition, inappropriate excreta disposal, inadequate hygiene and poor drinking water quality. Proper treatment and delivery of safe water under favorable conditions, as practiced in developed countries, is one of the best ways to heavily reduce threats published by WHO. Within this context, disinfection of drinking water is of key importance for resolving the problem (Solsona and Vega, 2003)

Disinfection kills or inactivates disease-causing organisms in a water supply and must provide a 99.9 percent inactivation of *Giardia lamblia* cysts and enteric viruses to protect health and to comply with the U.S. Environmental Protection Agency (EPA) regulations. There are two kinds of disinfection: primary disinfection achieves the desired level of microorganism killing or deactivation, while secondary disinfection maintains a disinfectant residual in the treated water that prevents the re-growth of microorganisms.

Primary methods of disinfection are chlorination, chloramines, ozone, and ultraviolet light. Other disinfection methods include chlorine dioxide, potassium permanganate, and nano-filtration. Since certain forms of chlorine react with organic material naturally present in many water sources to form harmful chemical by-products, the U.S. Environmental Protection Agency has proposed maximum levels for these contaminants.(1)

(1) http://water.epa.gov/drink/contaminants/basicinformation/disinfectants.cfm#What%20are%20EPA%E2%80%99s%20drinking%20water%20regulations%20for%20disinfectants?

Boiling can be used as a method of water disinfection. Bringing water to the boil is effective in killing or deactivating most bacteria and pathogens. Boiling is the most certain way of killing nearly all microorganisms. But boiling has its disadvantages like scalding, scaling of boiling vessels, and removal of useful minerals and blandness of taste.

It is generally known that heating water at 65^0C for approximately six minutes will kill most of the pathogens (Andreatta, 1994). This process is referred to as pasteurization (Qazi *et. al*, 2002).

2.1.2 Household methods

There are many situations where individuals or families would need to resort to simple and effective methods for drinking-water disinfection. These include the following:

- Catastrophic conditions leading to displacement (earthquakes, floods, hurricanes, wars, or civil disturbances).
- Emergencies arising from flourishing waterborne diseases.
- Resident populations and foreigners at risk in endemic areas with unsafe water supplies.

Physical methods (boiling or the use of ceramic filters), chemical methods (chlorine compounds in solution or tablet form, e.g., sodium hypochlorite solutions, calcium hypochlorite tablets, organic chlorine compounds, iodine solution, and organic iodine compounds) and others have been recommended for such cases (Morris *et al*, 1953; Gershenfeld, 1957; Hadfield, 1957; Cox, 1969; O'Connor and Cooper, 1970 and WHO, 1972).

None of these methods, however, is entirely free from practical problems. Fuel wood for boiling, for instance, is no longer a tenable practice, particularly in areas where it is absent

or being depleted. Besides, the flat taste of boiled water discourages some consumers. The diverse types of ceramic filters have a wide range of pore sizes and present difficulties in selection. These suffer frequent clogging of the ceramic candles and often leak through disguised fine cracks. Chlorination frequently leads to consumer complaints and rejection because of the undesirable tastes and odors imparted to the water. It is especially so if high doses are applied inadvertently or as required in cases of heavily polluted waters. Relief agencies are often trapped in a dilemma by the requirements for importing and distributing, in addition to shortages, cost acceptability, and expiry dates. These issues encourage attempts to resolve them through the development of practical and effective techniques, simple enough to be applied by individuals or households.

2.1.3 Considerations Regarding Disinfection

As already stated, disinfection is a key process of any water treatment system. For that reason, it is important to emphasize a number of special considerations to be taken into account before undertaking disinfection to produce safe drinking water. Some of these are discussed below.

In designing a water treatment system, particularly in the rural areas, disinfection must not be approached as just one of several elements, but as a component vital to the system. Frequently, those who design water provision systems in small communities not only fail to take disinfection seriously, but they also give more importance to the flow (quantity) of water supply than quality of water supply.

Selection of the appropriate technology is very critical while designing a water treatment system. What is important when selecting a particular technology, however, is to take into account the determining factors, such as available resources and the possibility of technical support with regard to community social, economic and cultural aspects. When choosing the disinfection technique and system to be used, it is important to keep their

characteristics in mind and compare these with those of the plant, site and community. A good recipe is to complement the best conditions of the disinfection technique and system with those of the source, place, system and population and their cultural characteristics. This is very important and it must also be recognized that there is no ideal or perfect disinfectant or disinfection technique (Solsona and Vega, 2003)

2.2 Pasteurization

2.2.1 Background

Pasteurization was named after the French chemist Louis Pasteur and was first applied to inactivate spoilage microorganisms in wine then it was applied to milk (Dairy Science and Technology). Milk pasteurization can be defined as heating milk to certain temperature that is below the boiling point (100˚C) for a period of time. There are two major objectives of pasteurization. First, milk is pasteurized in order to kill all pathogenic microorganisms in milk so as to render it safe for human consumption (Kay, 1953), and the inactivation effect must be 100% for pathogenic germs (Spreer, 1998). The other objective is that pasteurization kills spoilage-causing microorganisms and that ensures safe milk. Also, pasteurization ensures the quality of milk and its products by destroying many undesirable enzymes in milk and extends the shelf life up to 16 days. [(2)]

2.2.2 Methods

There are two major types of heat treatment methods; batch and continuous heating systems.

2.2.2.1 Batch Method

In this method, the heat transfer is performed by using a vat pasteurizer which consists of a jacketed vat surrounded by either circulating water, steam or heating coils of water or steam and provided with a cover to prevent contamination. Also, a heated vat is provided

(2) http://www.foodsci.uoguelph.ca/dairyedu/pasteurization.html.

with an agitator to ensure uniform heating. Milk in the vat is heated at 63˚C and held at this temperature for 30 minutes throughout the holding period while it is being agitated. There is also a recording thermometer to trace a record permanent of time-temperature treatment (Energy technologies). The milk may be cooled in the vat or removed hot upon completing the holding period. This method has a better use of milk byproducts rather than the use of milk itself. It is mainly used in ice cream industry (Dairy Science and Technology).

2.2.2.2 Continuous Method

High Temperature Short Time (HTST) pasteurizer is used in this method. The heat treatment is performed by using a plate heat exchanger which consists of corrugated stainless steel plates clamped together in a frame. The heating medium is either hot water or vacuum steam.

Cold raw milk at 4˚C in a constant level tank is drawn into the regenerator of the pasteurizer. The milk here is warmed to approximately 57-68˚C by heat provided by hot pasteurized milk flowing in a counter current direction on the opposite side of thin stainless steel plates. The raw milk that is still under suction passes through a positive displacement timing pump which delivers it under positive pressure through the rest of the HTST system.

The raw milk is then forced through the heater section where hot water on the opposite sides of the plates heats the milk to a temperature of at least 72˚C. After that, the milk at this temperature and under pressure flows through the holding tube where it is held for at least 16 seconds. At the end of the holding tube there are temperature sensors, indicating thermometer and record-controller where the milk passes through.

Then milk passes through the Flow Diversion Device (FDD). The FDD assumes a forward-flow position if the milk passes the record-controller at temperature more than 72˚C which is called (preset cut-in temperature). However, the FDD remains in the normal position (in diverted-flow) if milk has not achieved preset cut-in temperature. This improperly heated milk flows through the diverted flow line of the FDD back to the raw milk constant level tank.

Properly heated milk flows through the forward flow part of the FDD to the pasteurized milk regenerator section where it gives up heat to the raw product and is cooled to approximately 32-9˚C. The warm milk passes through the cooling section where it is cooled to 4˚C or below by coolant on the opposite sides of the thin stainless steel plates. Then, it passes through a vacuum breaker at least 12 inches above the highest raw milk in HTST system then on to storage tank filler for packaging (Dairy Science and Technology)[(3)]

2.2.3 Water Pasteurization Indicators

In 1992, Dale Andreatta, a graduate engineering student at the University of California, Berkley, developed the Water Pasteurization Indicator (WAPI). The WAPI is a polycarbonate tube, sealed at both ends, partially filled with a soybean fat which melts at 69^0C ('MYVEROL' 18-06K, Eastman Kodak Co., Kingsport, TN 37662). The WAPI is placed inside a water container with the fat at the top of the tube. If heat from the water melts the fat, the fat will move to the bottom of the WAPI, indicating water has been pasteurized. The WAPI is reusable. After the fat cools and becomes solid on the bottom, the fish line string is pulled to the other end, placing the fat at the top of the tube.

[(3)] http://www.foodsci.uoguelph.ca/dairyedu/pasteurization.html#htst.

Another pasteurization indicator has been developed by Saye and Pejack (1994), which is based on expansion of a bi-metal disc which is housed in a plastic container. This also shows promise and is in the early testing stages.

2.3 Thermochromism

2.3.1 Background

Thermochromism is the ability of a substance to change color due to a change in temperature. Thermochromic pigments have a large range of possible color, and sensitively register changes in temperature by changing color or becoming transparent at a precisely-determined threshold temperature. The change is reversible, and can be repeated over long periods of time without substantial degradation in the pigment's thermochromic properties (Christie *et al.* 2007).

2.3.2 Thermochromic Liquid Crystal Reversible Temperature Indicating Strips

The Thermochromic Liquid Crystal (TLC) range of reversible temperature indicating labels forms a fast, easy to interpret indication of actual temperature. They are also convenient, easy to use and in-expensive.

2.3.2.1 What These Are ?

These self-adhesive labels consist of a series of temperature-sensitive elements containing microencapsulated TLC coated on a black backing. Each element changes colour distinctly as its rated temperature is reached, passing through the colors of the spectrum in sequence (Orange, Yellow, Green, and Blue before turning black at a higher temperature). The TLC strips are calibrated so that the indicator that shows green indicates the actual temperature.

The color changes are reversible and the reflected colors will be observed in the reverse order upon cooling.

2.3.2.2 How the labels work

The temperature-sensitive elements contain TLC molecules that are very sensitive to temperature and change position / twist in relation to changes in temperature. This change in molecular structure affects the wavelengths of light that are absorbed and reflected by the liquid crystals, resulting in an apparent change in the color of each temperature event.

When the rated temperature of an indicator is reached the TLC molecules twist slightly causing the TLC substance to absorb the red and blue portions of the visible light and reflect the green part. This causes the temperature event to appear green. When the temperature decreases, the molecules begin to twist in the opposite direction, and the TLC reflect a different portion of the spectrum.

2.3.2.3. Label Accuracy

The tolerance on all our products is ±1°C of the rated temperature, with the exception of the medical products (forehead thermometers and drug test strips) that have a tolerance of ±0.5°C of the rated temperature

2.3.2.3 Reading TLC Thermometers

The correct temperature is indicated by the square on the TLC thermometer that turns green. If green is not visible the temperature will be mid-way between the indicators that are illuminated tan and blue as follows:

Generally liquid crystal thermometer strips / temperature indicators can be read accurately to within half the increment between adjacent temperature events. For example the A296

series products, which have events sequenced every 5ºC, the temperature can be read to within 2.5ºC. For A523 series where events are sequenced every 1ºC or 2ºC and medical products (including forehead and drug test strips) the temperature can be read to within 0.5ºC and 1ºC respectively (Van der Maas *et al*, 2010).

2.3.3 Preparation of Reversible Thermochromic Materials

Since there are different seasons in nature, the solar energy absorbed by buildings varies largely in winter and summer. Therefore building thermal environment is not always comfortable for living and working.

To create a thermally comfortable building environment, a lot of measures have been taken for buildings, in which porous insulating materials are used widely. Although they can contribute essentially to the reduction of the fossil fuel consumption, the problems of fossil fuel consumption and environmental protection can't be resolved thoroughly by use of these materials. Reasonable usage and disposal of solar energy may be the final resolving method, which includes various solar walls and other solar devices, but the above material and devices may be complex and expensive for buildings.

The author and his colleagues have developed the so-called reversibly thermochromic building coatings to meet with the needs, whose color can be changed from red, blue etc. below a switching temperature to white above the switching temperature. During this process, solar energy is mainly absorbed below the switching temperature, and much of it is reflected above the switching temperature (Yiping Ma *et al*, 2008).

2.3.4 Solid State Thermochromic Materials

Solid-state thermochromic materials undergo semiconductor to metal transitions at a critical temperature, ***Tc***. This review begins by describing the phenomenon of thermochromism, whereby the optical properties of a material change reversibly as a result

of a change in temperature. The various different types of thermochromism will be introduced with a focus on the thermochromism exhibited by solid-state materials.

The fundamental chemical principles that describe the electronic structure and properties of solids, and the chronological developments in the theory behind the thermochromic transitions (such as, the effects of electron-electron interactions and structural phase changes due to lattice distortions) that led to the discovery of the semiconductor-to-metal transition, are presented.

An extensive discussion of vanadium and titanium oxides is presented with a particular focus on vanadium (IV) oxide since its transition temperature is closest to room temperature. Observations and current understanding of the nature of the semiconductor-to-metal transition exhibited by these materials is detailed. The possibility of fine-tuning the transition temperature by introducing various dopants (doping agent) into the vanadium (IV) oxide lattice is examined and the effects of dopant charge and size is examined.

Solid-state thermochromic materials may be exploited in areas such as microelectronics, data storage, or intelligent architectural glazing, thus are required to be synthesized as thin films for use in such applications.

The numerous synthetic techniques (PVD, sol-gel method, PLD, CVD, APCVD and AACVD), for making metal oxide thermochromic thin films are described in reference to the production of vanadium (IV) oxide and compared. Finally rare earth nickelates exhibiting thermochromism are described. (Kiria *et al*, 2010).

2.4 Applications of Thermochromic Strips

- The color indicating strip is very efficient in identifying the water pasteurization temperature.
- Use of these strips could help in providing good quality, palatable water to the user with considerable energy saving.
- The strips could also find their application in effective disinfection of water and can be used in situations like floods, earthquakes or influx of refugees.

Chapter 3

3 Materials, Methods & Experimentation

3.1 Materials

This chapter details the materials and methods used for the manufacturing and testing thermochromic strip. The chemical, media and apparatus used in the fabrication of the strip is listed below:

Chemicals:

- Mercuric nitrate $Hg(NO_3)_2$
- Potassium Iodide KI
- Sodium meta bi sulfite $Na_2S_2O_5$
- Copper Sulfate $CuSO_4.5H_2O$
- Distilled water H_2O

Media:

- Lauryal Tryptose Broth (LTB)
- Brilliant Green Bile Broth(BGLB)
- EC broth
- EMB agar

Apparatus:

- Test tubes
- Funnel
- Beakers
- Tripod stand
- Pipette
- Bunsen burner

- Colony counter
- Petri plates
- Stirrer
- Hot plate
- Mass balance Filtration assembly with vacuum pump.
- Membrane filter of 1.2 micro meter GF/C WHATMAN.
- Petri dishes etc.

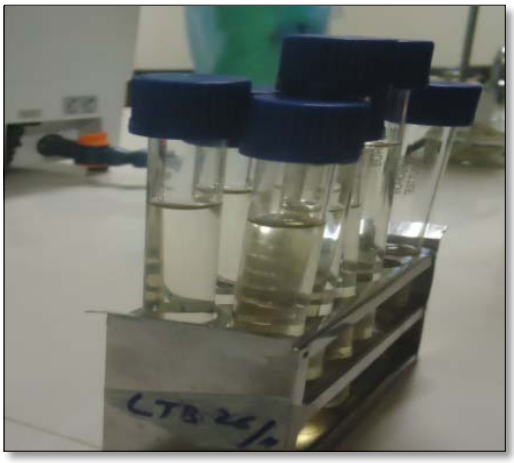


Figure 1- Lauryal Tryptos(BGLB)

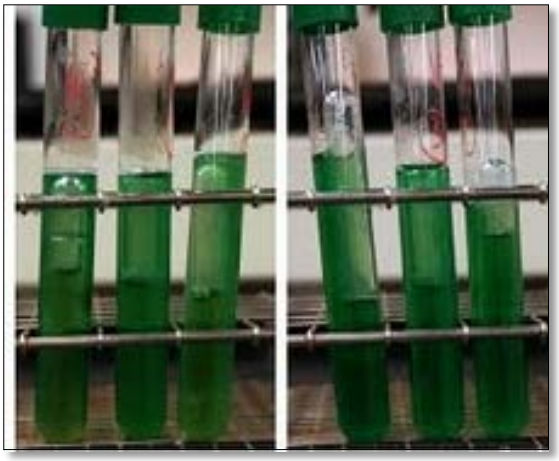


Figure 2- Brilliant Green Bile Broth

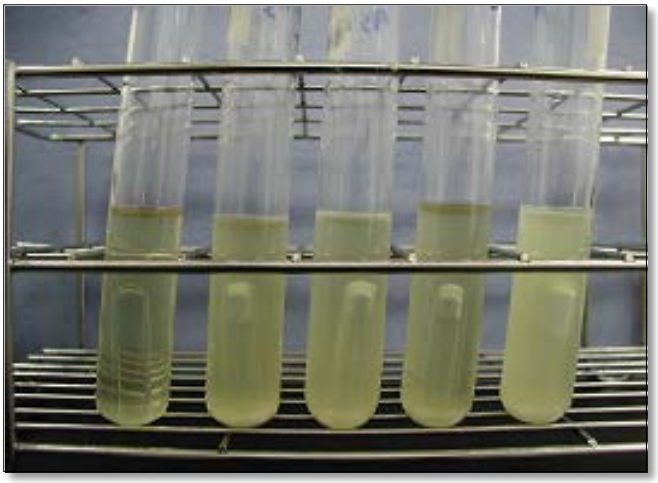


Figure 1-EC Broth

Figure 3-1: Media Used During This Study

3.2 Development of Thermochromic Strips

3.2.1 Synthesis of Thermochromic Pigment

Two different procedures were used for synthesis of thermochromic strip, the first one being unsuccessful. After making slight changes in the first procedure the right pigment was achieved. Both the procedures are detailed below:

First weigh out 1.6g of Mercuric Nitrate, 5g Potassium Iodide, 3g sodium meta bi sulfite and 2.5g copper sulfate. Figure 3-2 shows the digital balance used for measurement of the constituents.

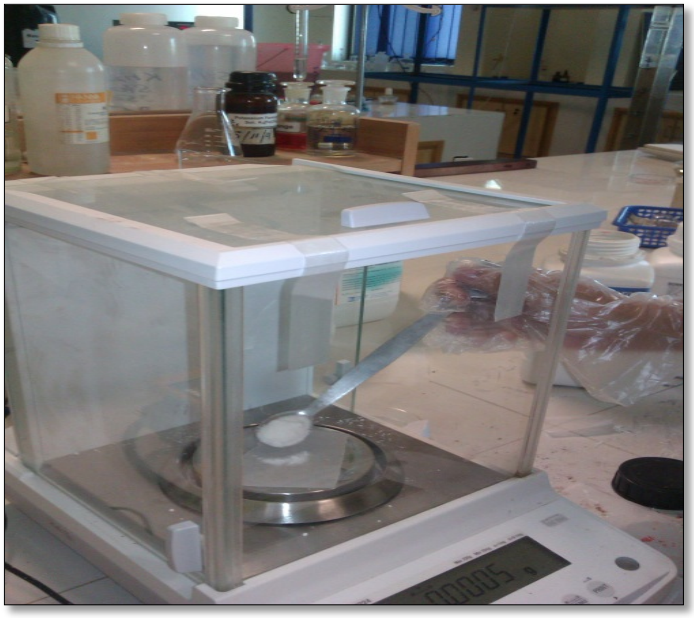


Figure 2-Digital Balance

3.2.1.1 First Procedure (Un-Successful)

- Add Mercuric nitrate in 25ml of boiled distilled water
- Add Potassium nitrate till the initial precipitate dissolves to give a clear solution

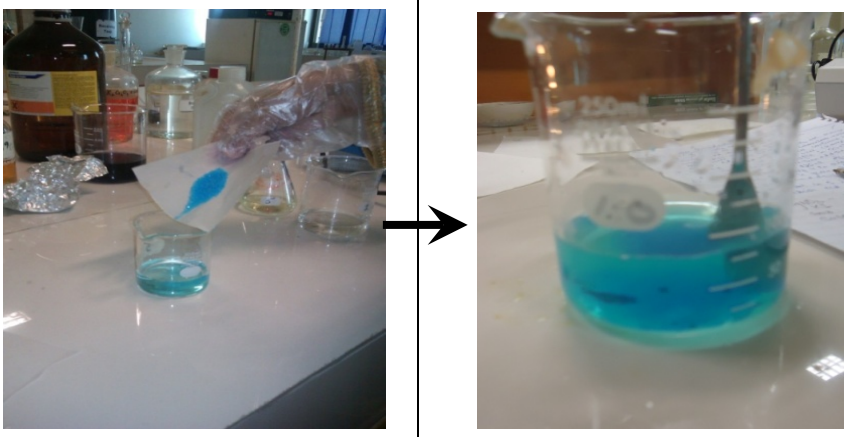
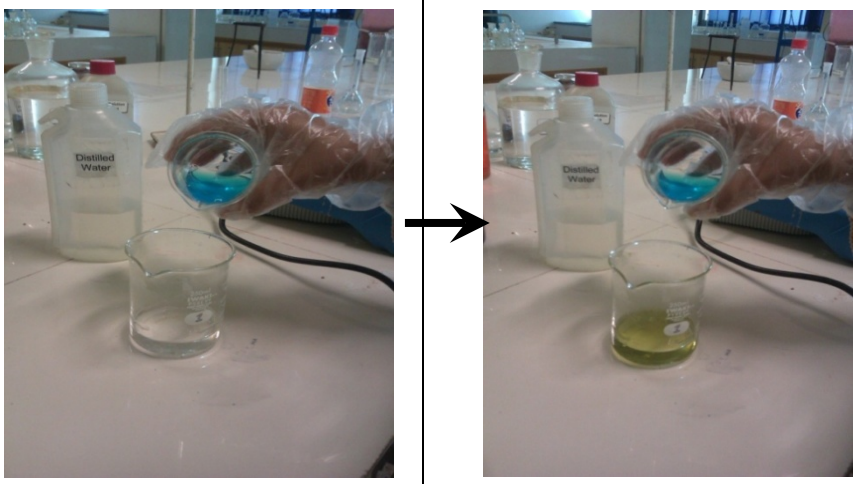
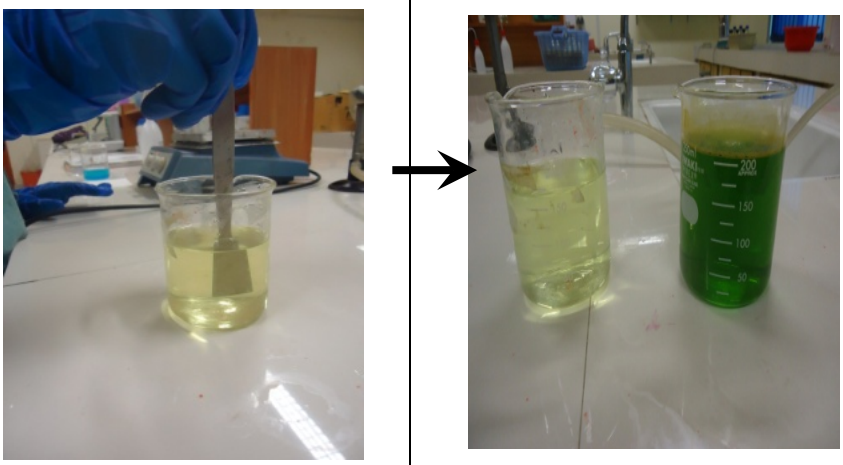
- Add Sodium meta bi sulfite in 50ml distilled water and boil in the potassium nitrate solution
- Add copper sulfate in 25ml distilled water.
- Add copper sulfate solution to boiled sodium meta bi sulfite solution.
- Mix both the solutions

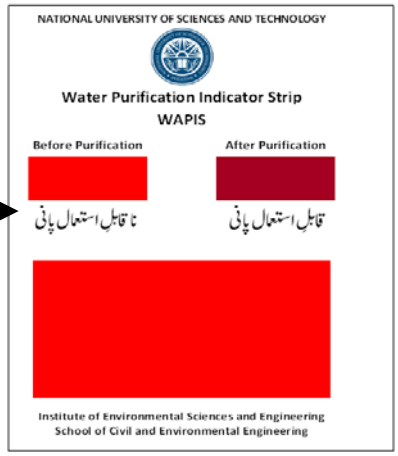
3.2.1.2 Second Procedure (Successful)

- Add mercuric nitrate in 25ml boiling distilled water.
- Add potassium iodide in 50ml distilled water.
- Slowly add this potassium iodide solution in the mercuric nitrate solution. Orange color will appear which will then become colorless; continue adding till it continues to remain orange. Add more of the solution and the colour changes to pale yellow. Stop adding.
- Add sodium meta bi sulfite in 50ml of distilled water, boil.
- Add copper sulfate in 25ml of distilled water.
- Add the copper sulfate solution to the sodium meta bi sulfite solution.

  Color change: green to yellow to muddy color
- Mix the above solution in the pale yellow solution. Pinkish red color appears

Table 1- Procedure for Synthesis of Thermochromic Pigment & Strip

Steps	Remarks
	Adding copper sulfate in 25ml of distilled water.
	Adding the copper sulfate solution to the sodium metabi sulfite solution.
	Adding mercuric nitrate in 25ml boiling distilled water.

		Mix the muddy green solution in the pale yellow solution. Pinkish red color appears
		Settling of the solution and then making a thermochromic strip.

3.2.2 Filtering the Pigment

The above solution is left for 24 hours to settle. Next the clear supernatant is removed carefully few times and the solution becomes concentrated. Then a filter paper is folded into the funnel which is fixed into the tripod stand. Next the prepared solution is poured in the funnel slowly. The solid part remains on the filter paper while the liquid portion is filtered out into the beaker. This liquid part undergoes the same procedure again, to achieve maximum amount of pigment.

This pigment can be smeared on the filter paper directly or by first powdering it and then dissolving it in ethanol. The latter is more suitable and is as follows:

After filtering the filter paper is left to air dry for a few hours. Later the dried pigment is grinded manually (Figure 3-3). This powder is then added into 1-2 ml of ethanol and stirred continuously till dissolved.

Figure 3-Grounded Pigment

3.2.3 Paste Application

A fine layer of the prepared paste is applied on the filter paper using a paintbrush, in thin strokes. The smearing is done uniformly and very carefully. Any remaining blank spaces are filled with the paste. The strip is then left to air dry. Figure 3-4 shows the application of paste onto the strip.

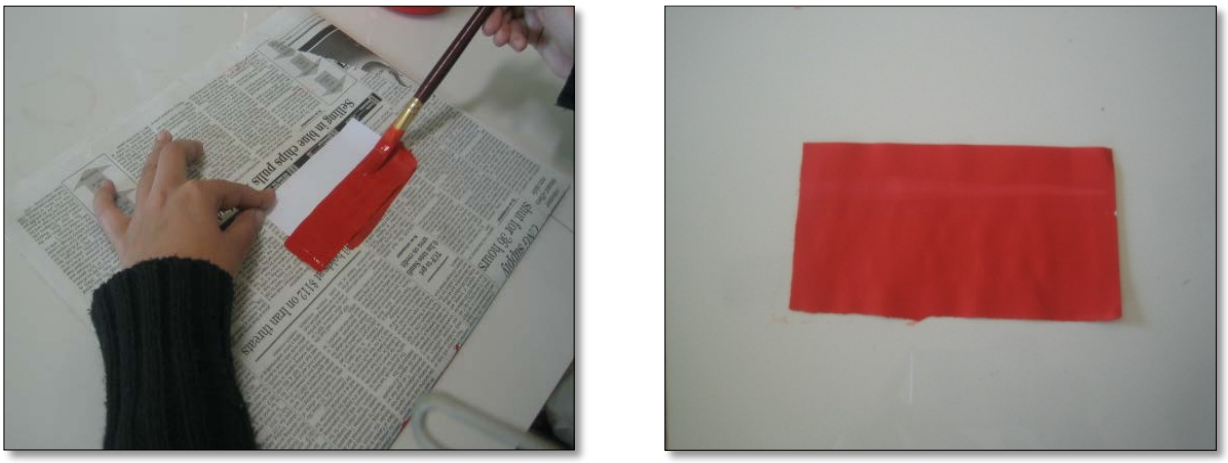


Figure 4 - Application of Thermochromic Paste on the Strip

3.3 Microbial Analysis of Drinking Water

Following three techniques were used for the microbial analysis of drinking water

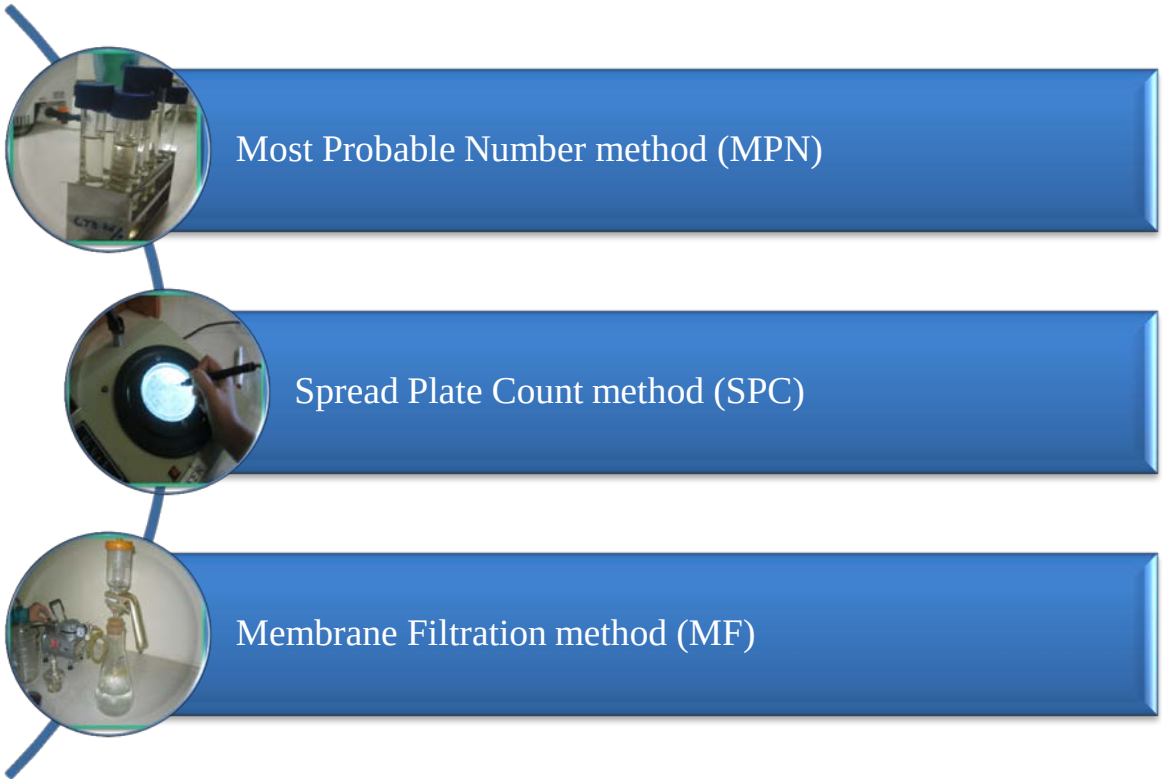


Figure 5-Methods for Microbial Analysis

3.3.1 Membrane Filtration (MF)

The Membrane Filtration (MF) Technique was introduced in the late 1950s as an alternative to the Most Probable Number (MPN) procedure for microbiological analysis of water samples. The MF Technique offers the advantage of isolating discrete colonies of bacteria and monitoring drinking, waste, and surface water.

Using the MF Technique, a 100 mL sample is passed through a 47 mm membrane using a filter funnel and vacuum system. Any organisms in the sample are concentrated on the surface of the membrane. The filter is then placed in a petri dish with nutrient medium. The

passage of nutrients through the filter facilitates the growth of organisms on the upper surface of the membrane.

Initially synthetic contaminated water was synthesized in the micro laboratory followed by the following dilutions.

- 5% distilled water and 95% contaminated water
- 10% distilled water and 90% contaminated water

3.3.2 Most Probable Number (MPN) Method

The Most Probable Number (MPN) technique is an important technique in estimating microbial populations in water. The MPN technique (Woomer 1994) estimates microbial population sizes in a liquid substrate. The methodology for the MPN technique is dilution and incubation of replicated cultures across several serial dilution steps. Tubes contain inverted Durham tubes to detect gas production (Lactose is converted to acid, gaseous H_2 and CO_2).

Sample: Tap water

It comprises of following steps:

a. Presumptive Test for Total Coliforms:

Ten test tubes method was used for the analysis. Initially Lauryl Typtose (LT) broth medium was used, single strength LT media were prepared. Dehydrated media was first weighed, pH was maintained and then media was poured into test tubes. Inverted Durham tubes were then added into these tubes and then media was sterilized in autoclave at 121ºC. Tubes were inoculated using 10 ml sample. Samples, after being inoculated into broth media, were incubated at 35±0.5 º C for 24-48 hours. After the time period of incubation, results were noted. Positive test was noted as the color change from light yellow to muddy

and the formation of bubbles in Durham tubes, whereas no color change and no bubble formation indicated a negative test.

b. Confirmatory Test of Coliforms:

Positive test tubes of LTB were then shifted into Brilliant green lactose bile broth (BGLB) for the confirmation of total Coliforms (Figure 8). A loop-full from positive LT tubes was then shifted into BGLB and incubated at 35±0.5ºC for 24 hours. After 24 hours any bubble formation/gas formation in the test tube showed positive test. The number of positive BGLB tubes was noted and this gave a confirmation for Total Coliforms. The number of positive test tube will give the MPN number using the MPN table.

c. Completed Phase Test for Fecal Coliforms:

After confirmation of Total Coliforms, 10 % of positive BGLB test tubes were shifted using metallic loop into EC broth and incubated at 44.5±0.2 ºC for 24 hours. Positive test tubes indicated the completed test for Total Coliforms.

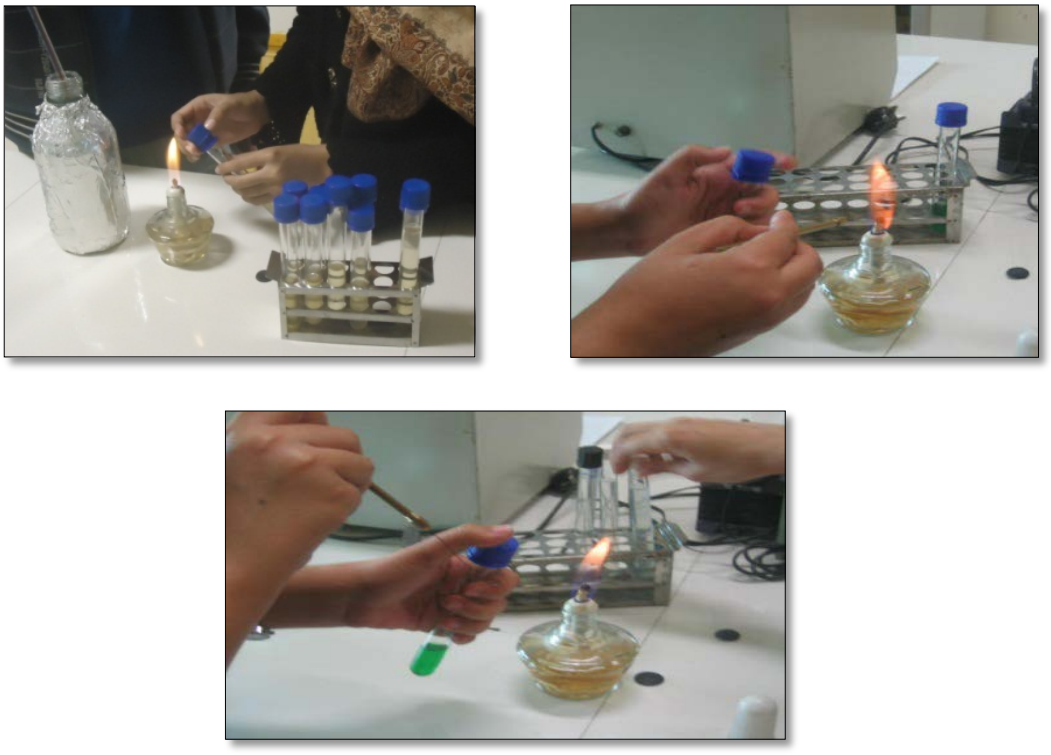


Figure 6-Transfer of Positive Test Tubes of LTB to BGLB Test Tubes

3.3.3 Spread Plate Count (SPC) Method

The purpose of the spread-plate technique [4] is to grow and isolate colonies of bacteria. A sample of bacteria is transferred to the agar plate, an environment that provides nourishment for the bacteria to grow. This is done by serial diluting the samples, placing 0.1ml of the diluted sample in the middle of an agar plate and spreading the sample over the surface with a help of an L-rod. After the incubation the colonies can be counted.

This method yields colonies that form on the surface of the agar. Plate counts assume that every colony is founded by a single cell.

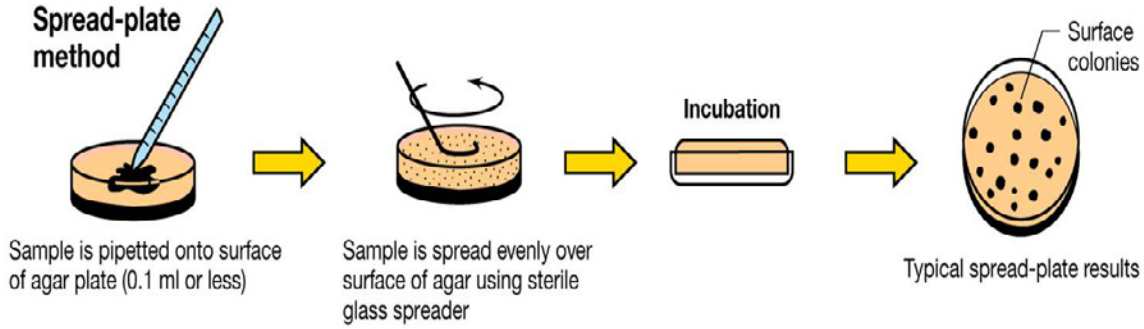


Figure 7-Flow Diagram for procedure of Spread Plate Count

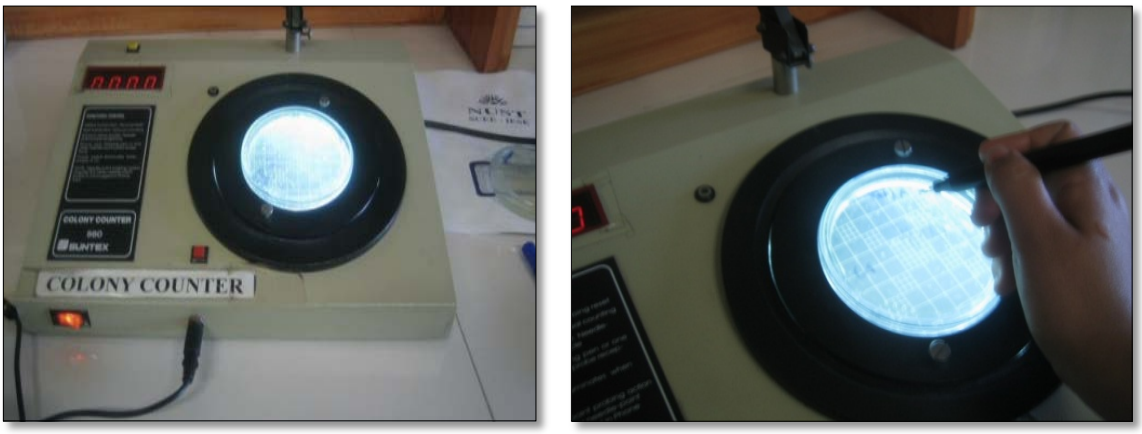


Figure 8-Colonies Counting After Incubation of Sample

(3) http://filebox.vt.edu/users/chagedor/biol_4684/Methods/platecounts.html

3.4 Summary

This chapter presented the materials and methods used to synthesize thermochromic strip. Two different methods were presented from which method one was unsuccessful while method two was found to be successful for the synthesis of the strip. The thermochromic strip was successfully fabricated by pasting the correct composition of material on a filter paper. Bacterial load of tap water was determined by MPN, MF and SPC technique.

Chapter 4

4 Discussions and Analysis of Results

4.1 Introduction

This chapter details the result obtained from microbial analysis of water performed before and after heating up to 65^0C

4.2 Membrane Filtration (MF)

Initially a contaminated water sample was taken and filtered through the membrane; however, the bacteria were Too Numerous To (TNT) count. Hence, MPN and SPC techniques were selected for further microbial analysis of the tap water.

Figure 4-1: Filtration Assembly

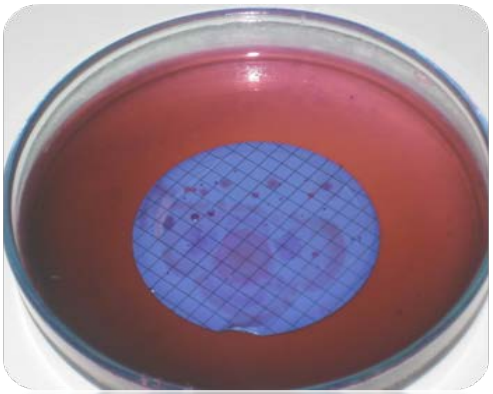


Figure 4-2: Membrane Filter with EMB Agar

4.3 Most Probable Number method (MPN)

4.3.1 Before Heating Up To 65^0C

Gas bubbles and muddy color indicates the presence of *coliforms* in water sample (Figure9).

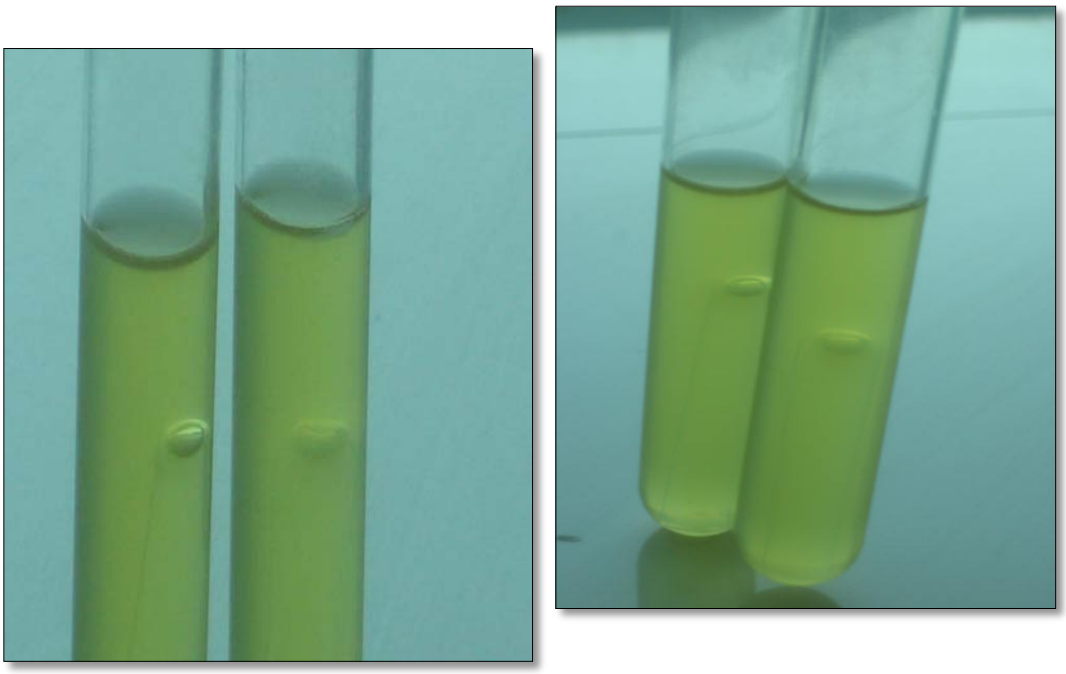


Figure 9-Water Samples with Coliforms

Table 2- Results of MPN before Heating Up To 65^0C

Sample ID	Total Coliform	Fecal Coliform	95% Probability Range
	MPN Index/100mL	MPN Index/100mL	

Sample 1	5.1	5.1	1.6-13

4.3.2 After Heating Up To 65˚C

No gas bubbles and transparent medium indicate absence of coliforms in water sample. (Figure10).

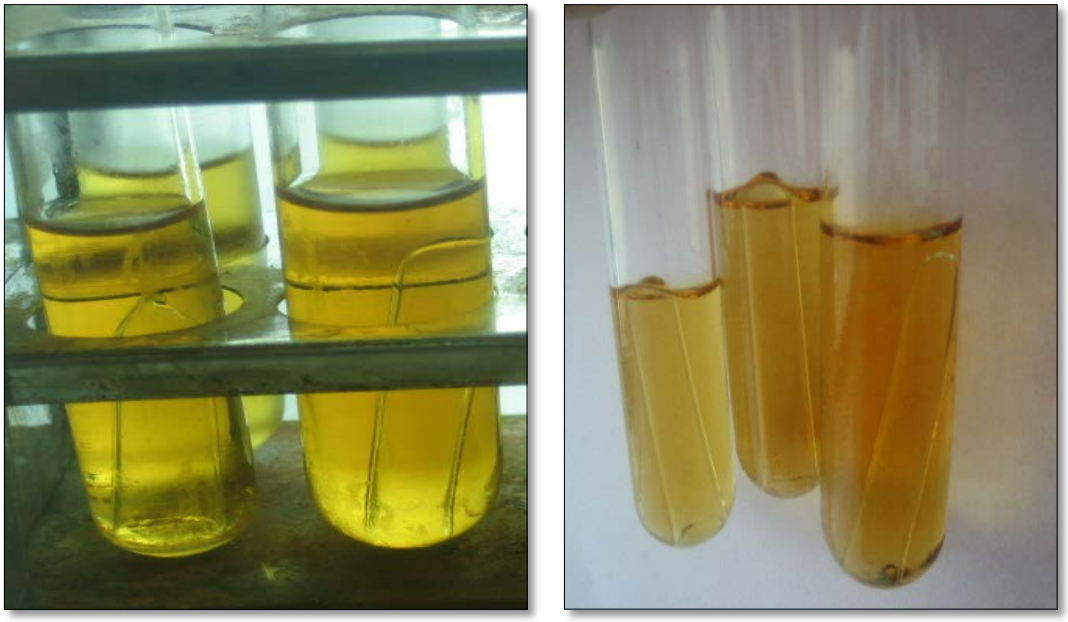


Figure 10-Water Samples without Coliforms

Table 3-Results of MPN after Heating 65^{o}c

Sample ID	Total Coliforms	Fecal Coliforms	95% Probability Range
	MPN Index/100mL	MPN Index/100mL	
Sample 1	<1.1	<1.1	0 - 3.0

4.4 Spread Plate Count (SPC) Method

4.4.1 Before Heating Up To 65˚C

Light yellow colonies formed on agar indicate the presence of coliforms in water sample before heating up to 65˚C.

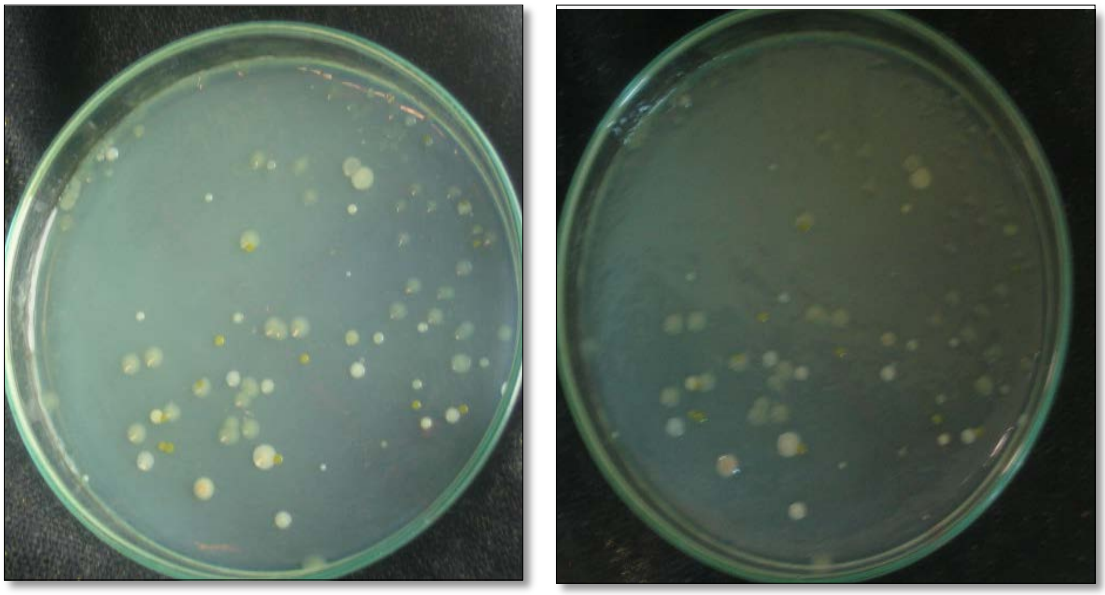


Figure 11-Light Yellow Colonies of Coliforms

Table 4-Results of SPC before Heating Up To 65^{o}c

Sample ID	SPC (CFU/ml)
Sample 1	98

4.4.2 After Heating Up To 65^0C

No colonies of coliforms on agar after heating water sample up to 65˚C.

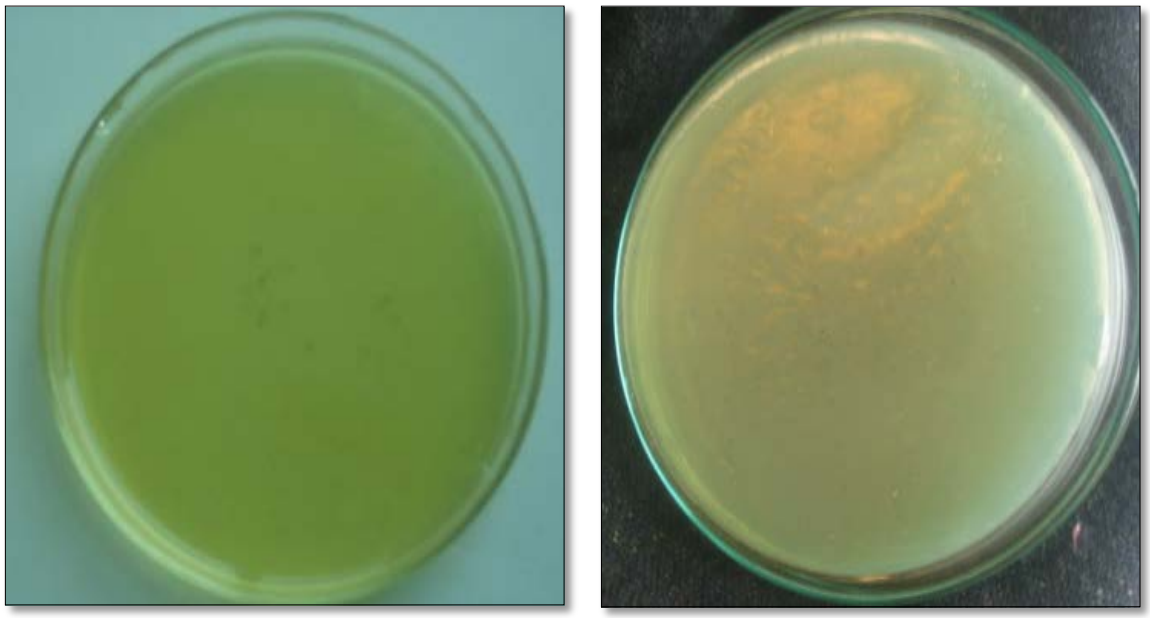


Figure 12- No Colonies of Coliforms

Table 5-Results of SPC after Heating Up To 65^oc

Sample ID	SPC (CFU/ml)
Sample 1	0

4.5 Summary

Results obtained from microbial analysis proved that heating tap water upto 65^0C is effective in killing of pathogenic microbes in water

Chapter 5

5 Design, Manufacturing and Testing of Water Disinfection Container

5.1 Design of container

A container incorporating the Thermochromic strip was designed and fabricated with the assistance of School of Mechanical and Manufacturing Engineering (SMME), in the Manufacturing Research Center (MRC), headed by Lt Col (R) M. Naweed Hassan (DD MRC & CAC Coordinator).

5.2 Materials Used

Following are the different materials used in manufacturing of the container.

- Aluminum
- Copper
- Glass
- Bakelite
- Super glue

5.3 Dimensions

These are dimensions of the container body:

Width: 182mm Length: 182mm Height: 218mm.

Figure 17-18 describe all the dimensions and design of container body and strip assembly. And also show their sketch.

5.4 Manufacturing Drawings of the Container

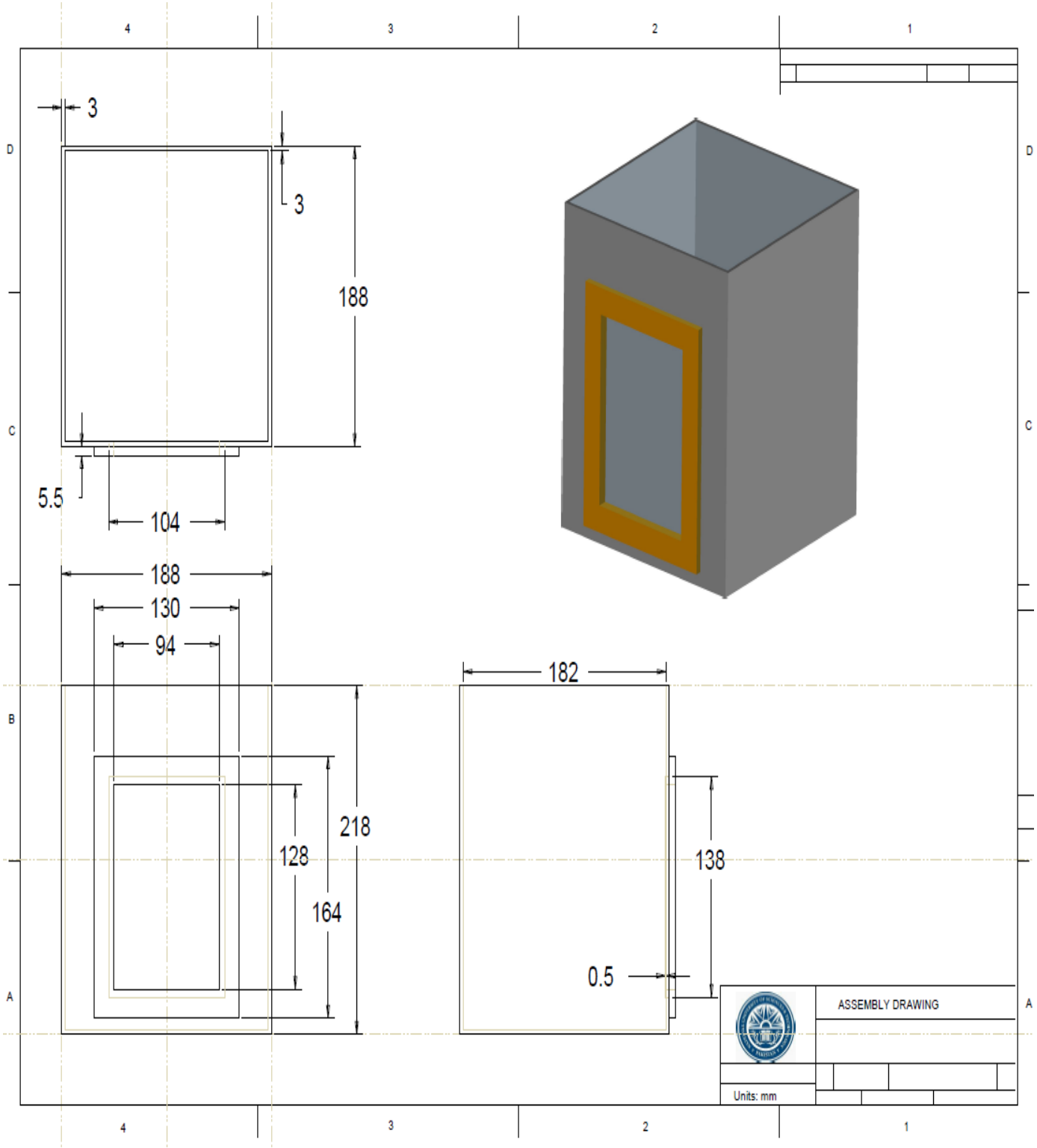


Figure 13-Manufacturing drawing of container

5.5 Drawing of the strip assembly

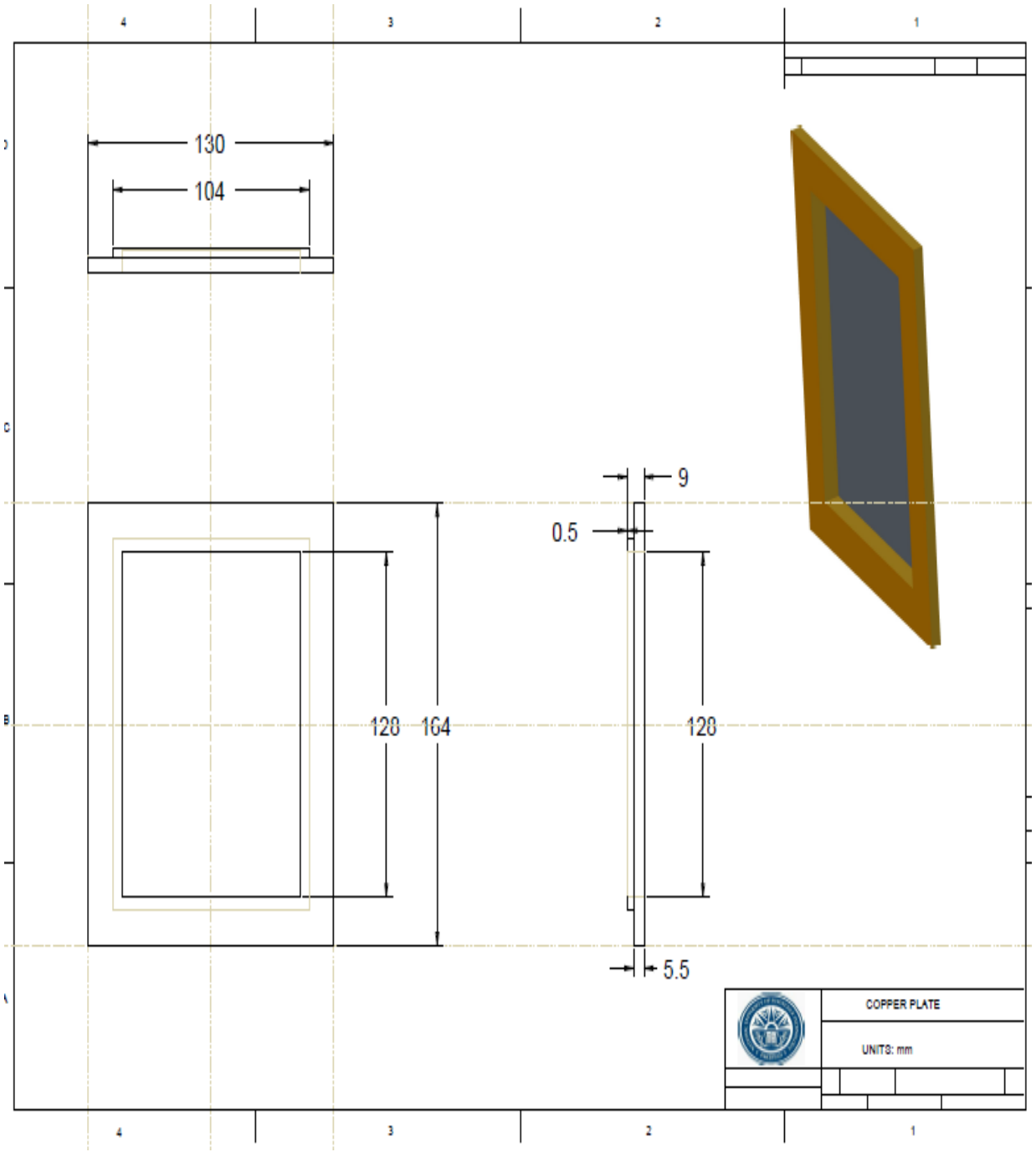


Figure 14-Drawing of strip assembly

5.6 Manufacturing

Initially the proposed design was created using AutoCAD 2007 and all the dimensions were specified. Later on we made specific additions to the design.

The container was made with aluminum sheets. A rectangular hole was drilled into the centre of the front wall. The strip was then pasted on a rectangular copper plate with the super glue. This prepared “strip assembly” was then insulated from all sides with an insulating material “Bakelite”, so that the strip receives heat only from the back side of the copper plate that is in contact with water. This assembly was covered with a glass frame for visual purposes so that color change in strip can be easily observed. This copper plate was then incorporated into the rectangular hole of the container.

One advantage of this design is that we can replace the strip instead of buying the whole container again.

Container was covered with a lid attached through hinges. Dispensing water was facilitated with a tap near the bottom.

To make the container more portable, two handles of Bakelite were attached to either side.

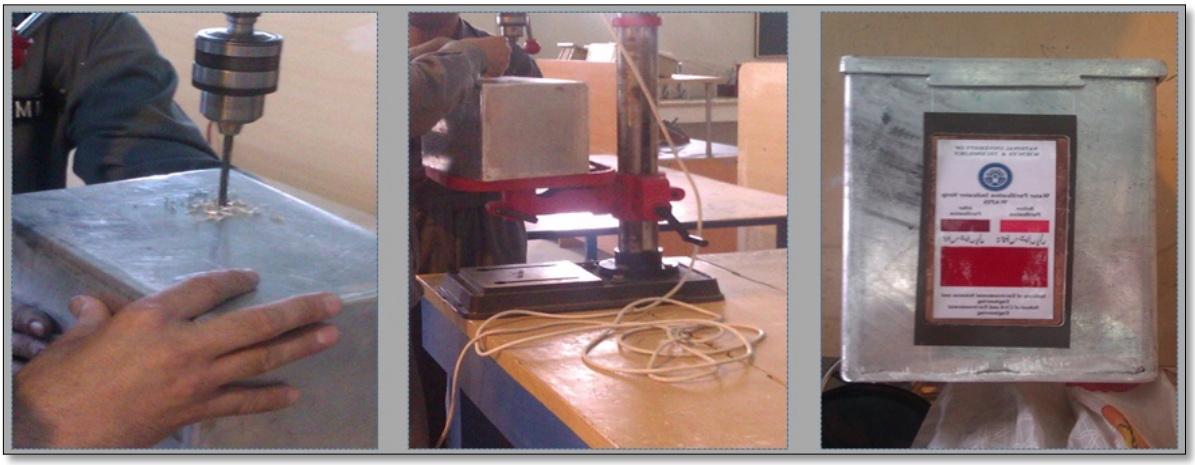


Figure 15-Container Manufacturing

5.7 Testing of the Container

Finally the container was put to test. Water in the container was heated on a hot plate up to 65-70^{O}C for purification.

The strip changed color sharply from red to dark brown at this temperature. This color was intense and the strip regained its original color after cooling. Thus water was purified and the container incorporating the strip proved to be a significant indicator of water purification, with considerable energy saving and limited scaling.

5.8 Summary

This chapter presented the design, manufacturing and testing of the container. A container incorporating the Thermochromic strip was designed with the assistance of SMME (School of Mechanical and Manufacturing Engineering). Basically the design was converted into a container in the MRC (MANUFACTURING RESEARCH CENTER) headed by Lt Col (R) M. Naweed Hassan (DD MRC & CAC Coordinator).Designed container incorporates a thermochromic strip; an efficient indicator of temperature change ,provides potable and safe drinking water and purifies drinking water by means of efficient energy consumption

Chapter 6

6 Conclusions and Recommendations

6.1 Conclusions

The following conclusions were drawn from this research work:

- Heating tap water up to 65-70^{0}C range is effective in killing pathogenic microbes.
- It prevents scaling in boilers, blandness of taste and results in considerable energy savings
- Designed container incorporates a thermochromic strip; an efficient indicator of temperature change.
- The designed container can be used for water purification purposes in homes
- Ethanol can be used to make a uniform paste of thermochromic pigment.

6.2 Recommendations

- To make the container design more economical we can eliminate the copper plate because it is more expensive and Thermochromic strip can be pasted directly onto Aluminum frame.
- Further methods need to be devised for applying paste onto the paper so that it can be more uniform.

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