COCONUT OIL AS AN ALTERNATIVE FOR IMMERSION OIL

Balba, Dolce Monica U. Balba, Karla Anika A. Quiobe, Casey Lyn Z. Valbuena

ABSTRACT

Immersion oil is the one used for oil immersion microscopy and it is responsible for the definite image viewing of what is present in the slide or sample being observed. There are many types of oil used as immersion oil, and the most common example is the xylene oil and synthetic immersion oil. Researches have been utilizing coconut oil as an effective alternative for commonly used immersion oil. This study aimed to have an image comparison between samples using two different oils—coconut and synthetic oil—in terms of sharpness and contrast of the image. Experimental research design was used in this study. The experiment was done in Lyceum of the Philippines – Laguna. The researchers used eight (8) samples as one of their instruments. The study used t-test to compare the results. After series of experiments, the comparison between the sharpness of coconut oil and synthetic immersion oil has a t-test result of p=0.06 which indicates the characteristics of coconut oil being not significantly different in terms of sharpness. For the contrast of the two oils, with a t-test result of p=0.11, the test reveals that there is no significant difference between coconut oil and immersion oil in terms of their contrast. Based on the results of this study, coconut oil can be used as a potential alternative for immersion oil since the results are comparable in terms of the sharpness and contrast of image. Coconut oil is also more cost effective and can benefit the laboratory.

***Keywords**: Immersion oil, oil immersion microscopy, synthetic, coconut oil*

INTRODUCTION

In the field of medicine, macroscopic or physical examination alone is insufficient to diagnose a patient. Laboratory testing will always be the primary tool in identifying underlying diseases by examining

different body fluids, such as blood or urine, and certain tissues to determine for abnormalities. Different testing methods and procedures can be used. It can be by rapid test kits, machines, such as ELISA, and the most commonly used, microscopy.

Microscope is a daily need in a laboratory setting. It is used as an equipment to see parts and components of different substances that cannot be seen by the naked eye. It can be a single lens microscope or a compound microscope wherein the latter is compose of four (4) different types of objectives with different magnifications: scanner objective (4x), low power objective (10x), high power objective (40x) and oil immersion objective (100x) (Davidson, 2017). An oil immersion objective is specially designed to be used differently compare to other objective lenses. This technique is used to increase the resolution of the image viewed in microscope. According to Das (2017), the process is done by dropping an oil on the specimen and immerse the objective lens giving increase numerical aperture of the objective lens. The lens is moved down toward the slide and into the oil, thus there is no air gap between the slide and the lens. The oil is meant to have the refractive index of glass so as not to refract the beams of light being focused on the item being examined. The said oil is responsible for the definitive image viewing of what is present in the slide or sample. There are many types of oil used as an immersion oil, and the most common example is the xylene oil, and synthetic immersion oil.

Xylene oil gives a high quality and clear result but on the other side, it is known for its toxicity and carcinogenicity (Kandyala et al., 2010). The researchers conducted an experiment on the possible and suitable alternative or substitute. A study conducted with a xylene-free method versus the xylene and as outcome adequate results produced from Xylene-free method that nearly equaled the xylene method. Added advantages included better working atmosphere, cost effectiveness and decreased toxicity (Metgud et al., 2013). Synthetic immersion oil from Cargille is widely used in most laboratories. Synthetic oil does not yellow with age, has no tendency of hardening on lenses due to uneven volatility, acidity, and changing viscosity (Cargille, 2008). However, it is usually expensive and it may be a disadvantage for the laboratories in rural areas. On other related studies, it states that vegetable oil, coconut oil particularly, is an effective form. It can be a substitute to highly hazardous xylene oil without altering the microscopic details. It

shows the highest resistance to rancidity, non-toxic, heat stable and is slow to oxidize. It is endemic in tropical countries and has a cheaper value compared to synthetic immersion oils (Adeniyi et al., 2015).

The purpose of this study is to establish image comparison using synthetic immersion oil and coconut oil. The researchers differentiated the results they obtained using two different oils in terms of the images' sharpness and contrast. The findings of the study will redound to the benefit in the laboratory and school based laboratory considering that immersion oil is a need and most used in all laboratory. Nowadays, expensive immersion oils are used in the laboratory considering the fact that most of them are toxic and carcinogenic. The researchers acknowledged the alternative, coconut oil, which is non-toxic and inexpensive. The medical technology students will acquire the knowledge of using coconut oil as an alternative to synthetic immersion oil. The clinical instructors will study the outcome and can lead them to the introduction of the alternative coconut oil as immersion oil during the laboratory experiments. The clinical laboratory can help the registered medical technologists in the laboratory to the use of coconut oil as alternative to synthetic immersion oil considering that coconut is endemic in the Philippines.

Objectives of the Study

The purpose of this study is to identify the probability of using coconut oil as a substitute for synthetic oil for the benefit of laboratories to have a non-toxic and inexpensive oil that can be used routinely in the laboratory, and at the same time can give the same quality of results. . Further, the study sought to aim the following objectives: 1. to determine the quality of images according to sharpness and contrast; 2. to determine if there is a significant difference between the images produced using synthetic oil and coconut oil; and 3. to compare the cost efficacy between synthetic oil and coconut oil.

METHODOLOGY

In this study, experimental research design was applied. The experimental method of research was used to determine the difference of the two (2) results—one with coconut oil and other with synthetic immersion oil—in terms of sharpness and contrast of the image. It is a continuous process of gathering, analyzing, and testing data or knowledge on how coconut oil can be used as an alternative for synthetic oil. Experimental design was suitable for this study since the two variables—synthetic and coconut oil were correlated but were compared on how it will affect the results of this study (Blakstad, 2008).

Coconut was purchased from the local public market of Sto. Tomas, Batangas. Coconut oil extraction was performed in Malvar, Batangas, while the observation of specimens using coconut oil and synthetic oil with oil immersion microscopy was performed in Lyceum of the Philippines - Laguna, Brgy. Makiling, Calamba City Laguna. Fixed slides containing human skin, mammal lung, mammary gland, liver, blood*, Trichinella spiralis, Hymenolepis diminuta, and Staphylococcus aureus* were purchased from RTC Laboratory Services and Supply House, Quezon City. These are the types of specimens tested in the different sections in the laboratory such as Histopathology, Hematology, Parasitology, and Bacteriology.

Cargille Immersion Oil was used as the control in this study. The refractive index of the said oil is 1.51, identical to the refractive index of the glass that was used so the beam of light will not refract on the item being examined (Cargille, 2008).

According to Tamil Nadu Agricultural University (2017), the main source of coconut oil is from the dried coconut meat, known as the copra. Young coconuts have less meat and more water, in contrast with the dried or mature coconut that has more meat and less water, making the latter produce more coconut oil. Therefore, the researchers used matured coconut for the extraction of coconut oil. Coconut, being a tropical crop, grows well in a hot climate making it endemic in the Philippines. Upon extraction, coconut oil is heat stable and will be able to have a longer shelf life when kept in room temperature. Coconut oil has a refractive index of 1.43 - 1.46 (Intenational Gem Society, 2017).

According to FEDOIL (2017), coconut oil is mainly extracted from copra. With a fat content of 63-70%, 650 mL (650 grams) of coconut oil can be obtained from one kilogram of copra. The researchers used 0.5 kilograms of coconut and produced approximately 320mL of coconut oil and 15 mL of which is used in the microscopic observation of specimens.

The procedure used in making coconut oil was according to the procedure of Handayani et al. (2008). Dried coconut was grated upon purchase and the grated coconut is mixed with hot tap water. After squeezing and filtering using an old cloth, the coconut milk is placed in the refrigerator for five hours. After refrigeration, the cream separated from water. The water rich in protein settled below, and was drained off. The solidified cream rich in oil, went to the upper portion and was transferred into a pan for heating. While heating, constant stirring is done until remnants became brown and clear oil has exposed. The researchers used this freshly extracted coconut oil in the observation of specimens. This oil was unfractionated therefore this is limited to store only at room temperature or above to maintain its liquid form which is suitable for use in the laboratory (Wallace, 2017).

With the use of the low power objective, the area of interest in the slide is focused within the field of view. After focusing on the area of interest, a drop of coconut oil or synthetic oil is added to the specimen. The nosepiece is rotated to bring the oil immersion objective into position. Using the coarse adjustment knob, the stage is adjusted until the drop of oil on the slide barely comes in contact with the front of the objective lens. While looking through the eyepiece, the fine adjustment knob is used to move the lens up and down in tiny increments, to put the specimen into focus. When observation is done, the oil is removed from the objective lens and any other parts of the microscope that will come in contact with the oil (Oliver, 2016). The prepared slides with synthetic oil was compared with the slides with coconut oil to observe the difference of the immersion oil quality.

Data were analyzed to compare the synthetic and coconut oil in microscopic examination and to determine if coconut oil can be a potential alternative to immersion oil used in the laboratory. In the study, there were eight specimens viewed under the oil immersion objective using the two (2) oils. The researches and their mentor

graded the sharpness of the image as follows: 3 - image is well-enhanced, clear details and edges; 2 - poorly enhanced and slightly blurry; and 1 - blurred image. Sharpness is defined as a property of image inversely related to blur which is typically determined by the spread of edges in the spatial domain, and accordingly the attenuation of high frequency components (Prajapati et. al., 2015). On the other hand, the contrast of image was graded as follows: 3 - colors are fully pigmented and clearly differentiated; 2 - colors are slightly pigmented and differentiated; and 1 - no colors seen. Contrast refers to difference in grayscale or color that exists between image features. It is related with discrimination of objects or content within an image (Prajapati et. al., 2015). The average grading of the sharpness and contrast of the images were based from the score of each of the researchers including their mentor who is a registered medical technologist. This was computed using the t-test to determine if there is a significant difference between the images produced by Cargille synthetic oil and coconut oil. Results of their grading were analyzed and compared using t-test and average results were tabulated.

RESULTS AND DISCUSSION

The specimens like mammary gland, *Staphylococcus aureus*, liver and blood showed well-enhanced images according to sharpness using both Cargille synthetic oil and coconut oil. However specimen like human skin, mammal lung, *Trichinella spiralis* and *Hymenolepis diminuta* showed well-enhanced images only in Cargille synthetic oil and poorly-enhanced images using coconut oil in terms of sharpness. After summing up the values, the researchers garnered an average of 2.84 for synthetic oil and 2.59 for coconut oil leaving it with a p-value of 0.06. This p-value shows that there are no significant difference between the two oils.

The specimens showed fully pigmented and clearly differentiated images according to contrast in both Cargille synthetic oil and coconut oil, only *Hymenolepis diminuta* showed fully pigmented images using Cargille synthetic oil and slightly pigmented images using coconut oil. However, there was no significant difference between the

two variables which showed a T-test value of p = 0.011 according to contrast.

The cost between the two variables as for 15 ml showed that coconut is cheaper compared to coconut oil; the researchers spent Php130 for 320mL of coconut oil and the control, which is the synthetic oil, is more expensive amounting to Php2, 100 for 100mL.

CONCLUSION

Coconut oil can be a potential alternative to immersion oil. Synthetic immersion oil produced more sharpness in its image than coconut oil, however there is no significant difference in the p value; and synthetic oil produced a more pigmented image than coconut oil, however there is no significant difference in its p value as well. Therefore coconut oil can be a potential alternative because it is comparable to the control in terms of the sharpness and contrast of image. The price difference of coconut oil and synthetic oil is Php1970, thus indicating that coconut oil is more cost efficient than synthetic oil; which make it suitable to small laboratories particularly in rural areas or in free-standing laboratories.

RECOMMENDATIONS

The researchers suggest the continuous testing and manipulating of coconut oil while ensuring the standardization of materials used and calibration of it, to obtain same average results of the two variables. The researchers also encourage future researchers to use multiple samples using coconut oil and synthetic immersion oil to obtain data that could be more reliable and complete for the better comparison and understanding of the two oils. The use of other techniques and methods suitable in the study is also encouraged for the better outcome and understanding of results; such as fractionating the coconut oil to maintain a liquid form, and also to identify the factors that may affect the efficacy or composition of the oil such as coconut species, age of coconut and processing and storage of oil. The researchers also recommend future researchers to use freshly prepared smears such as peripheral blood smears; and to further study

the component of coconut oil that may affect the objective lenses of the microscope.

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