BOTANICAL ANTI-MANGE SOAP

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I. RATIONALE

Mange is a skin disease of animals caused by mange mites and dogs are no exception. Mangy dogs abound everywhere, an indication that it is becoming a problem because mites can be transmitted through contact with infested dogs and contaminated beddings. Mange pose a great threat to the health and well being of our pet dogs. It is manifested by skin lesions which leads to scratching that will result to unsightly skin and coat condition and if left untreated it may lead to death of the animal.

Many brands of synthetic acaricides are available in the market. They may be oral or injectable medications, topically applied formulations, dips and shampoos. These formulations are chemical based hence, they may be toxic or leave chemical residue to our pets and pet handlers. In addition resistance of organisms to drugs is becoming an increasing problem. Moreover, due to high cost of these commercial medications pet owners leave their dogs untreated. In order to address this ever growing predicament of pet ownersresearchers of the Institute of Veterinary Medicine (IVM) of DMMMSU, Bacnotan, La Union formulated two(2) botanical soap the Natural Coco Oil Soap (NCOS) and *Kakawate* Leaf Extract Soap (KLES). NCOS and KLES arecheap yet effective non-chemical treatment for mange already incorporated in our weekly health care routine for our pets.

NCOS(70%) was proven highly effective(100%) against mange in dogsand other ectoparasites comparable to the commercial anti-mange. It was also found out that it hastens wound healing, it eliminates doggie odor and it adds luster to hair coat. Information drive, small scale production and marketing initially started after the efficacy studies. Comments and suggestions of users of the soap were incorporated in the standardization process and as basis in the general acceptability(85.20%) and efficacy awareness(93.02%) studies by users of the soap.The formulation and process were standardized and the stability was tested, dermal

irritation test was also conducted to confirm the safety of our patients and clients which further enhanced the quality of the soap. The effect of at most 2 years of storage on the efficacy of NCOS was compared with the freshly cured soap and results of this study increased further our confidence in moving on globally.

KLES(20%) was likewise found highly effective(98.99%) against mange in dogs which is comparable to the commercial antimange. It was claimed by previous authors that it has insecticidal property, relieves itching, hastens wound healing, checks growth of bacteria and considered the most effectivescabicide. Upon knowing its efficacy against ectoparasites, information drive, continuous production (small scale) and marketing was conducted. As a result comments, suggestions and recommendations from users of the soap were incorporated in the succeeding formulation to improve the quality of the soap. These were also the basis for the formulation of questions for the acceptability and efficacy awareness studies. The general acceptability of the soap by users is 86.72% and the efficacy awareness is 93.02%.

Information drive activities involving both NCOS and KLES included brochures, posters, exhibits, marketing, techno-demo, presentation in local, regional and international fora, the internet is leading our product close to the international market.

Natural Coco Oil (NCO) is a naturally processed product from fresh coconut meat or its derivative (coconut milk and milk residue). It is the purest form of oil, is water white in color and has not undergone any chemical processing during extraction. It contains natural vitamin E and very low free fatty acid with mild scent (Garcia (2003). Dayrit (2003) stressed that lauric acid, a major active component of the coconut oil, is a medium chain fatty acid which is converted into monolaurin when processed by the body.

Lauric acid is most potent particularly in its monoglyceride form, monolaurin which solubilizes or dissolves the lipids, causing the disintegration of the covering or envelope of the disease causing organism, with the cover torn down; the integrity of the pathogen is compromised resulting to death (Kabara, 2004).

Isaacs and Thorman (1991), Isaacs *et al* (1990) also emphasized that the lauric acid which is the active ingredient of Virgin Coconut Oil disrupts the lipid membranes of organisms and thus inactivates them.

The efficacy of *kakawate* (*Gliricidia sepium*)) leaf extract soap could be due to its major constituents, which were sulfur, tannin, glycosides and fats. Sulfur, which is abundant and one of the active ingredients when applied to the skin, does not only destroy the parasites, but also slightly checks the growth of bacteria. Tannin on the other hand acts as an astringent. It precipitates protein either externally or internally. Its action is on the surface cells to facilitate the formation of the protective layer, under which healing can proceed as revealed by Einstein (1994), Fats are another major constituent of *kakawate,* used as protective agent to prevent contact with irritating substance, act as lubricating agent, which aid in the removal of crusts, and prevent excessive dryness (Musser, 1969).

For the added ingredients like sodium hydroxide and palm oil during soap making, *kakawate* leaf extract soap became more efficient against mange mites of dogs than the other previous studies about *kakawate* leaf extract. This is because *kakawate* leaf extract soap does not only target the mites, but it also cleanses the lesion, exposing the deeper portion of the skin where mange mites hide. During this process, some of these mites could already be rinsed off. Because of this action of the soap, the major constituents of *kakawate* could easily penetrate onto the stratum corneum, acting directly to the elimination of the mites.

Because of the above studies conducted and other information presented this research project is now ready to be presented as a technology.

II. OBJECTIVES

1. To determine the species of mites which can be treated bynatural coco oil soap and*kakawate* leaf extract soap
2. To determine the efficacy/effect ofnatural coco oil soap and*kakawate* leaf extract soap (KLES) against mange in dogs
3. To determine the most effective concentration ofnatural coco oil soap and*kakawate* leaf extract soap to treat mange in dogs
4. Todetermine the efficacy awareness of the respondents tonatural coco oil soap and*kakawate* leaf extract soap as an alternative medicine in controlling mange in dogs

5.To determine the general acceptability of the respondents to naturalcoco oil soap and *kakawate* leaf extract soap against mange in dogs

6.To standardize thenatural coco oil soap

7. To testthe stability of natural coco oil soap

8. To produce cheaper/affordable and natural treatment for mange in dogs

9. To promote/disseminate the natural coco oil soap and *kakawate* leaf extract soap to the public and

10.Togenerate income from production and marketing of botanical soap.

III. EXPECTED OUTPUT

1. To create/produce botanical anti-mange soap for dogs from natural source .

2. Base-line data for policy makers regarding the use of botanicals in treating parasites in animals

3. Clients who are fully aware of the efficacy regarding the efficacy of the botanical anti-mange soap.

4. Highly acceptable product by the animal owners/clients.

5. A stereotype, stable product which is more competitive in the market.

6. A stable 2 year old NCO soap in terms of efficacy, color and odor similar to that of a freshly cured soap

7. Available NCO soap year round, well informed public about NCO soap

8. Commercialization of the product throughout the country

9. Generate additional income for the university.

IV. REVIEW OF RELATED LITERATURE

Mange is a parasitic skin disease caused by microscopic mites. This is caused by several species of mites, depending on the type may live in the superficial layers of the skin or deep in the hair follicles. The general characteristics of mange infection are hair loss, severe itchiness and the presence of scabs. This may also lead to secondary infection (Orion Educator's, 2006).

Urquhart *et. al.* (1996), described the Life cycle of the two most common mites affecting dogs (Sarcoptes and Demodex). The fertilized female sarcoptes creates a winding barrow or tunnel in the upper layer of the epidermis, feeding on liquid oozing from the damaged tissues. The eggs are laid in these tunnels, hatch in 3-5 days, and the 6-legged larvae crawl on the skin surface. These larvae, in turn, burrow into the superficial layers of the skin to create small "moulting pockets" in which the moults to nymph and adult are completed. The adult males then emerge and seek a female either on the skin surface or in a moulting pocket. After fertilization the females produce new tunnels, either de novo or by extension of the moulting pockets. The entire life cycle is completed in 17-21 days.

Demodex on the other hand live as a commensal of the skin of most mammals, and are exceptional in being selective for a particular skin site, namely the hair follicles and sebaceous glands. Most species spend their entire life cycle in the follicles or glands, in each of which they occur in large numbers in a characteristic head-down posture. The mites then move into the extended habitats, going much deeper into the dermis then the sarcoptes and hence being much less accessible to surface-acting acaricides. This form of mange is best documented in dogs, but the pathogenesis and epidemiology in other animals suggest that their infections may have much in common with canine demodicosis.

The mange most people think of (Aiello, 1998) is the *sarcoptic* mange, caused by the mite *Sarcoptesscabieivarcanis*. There are four developmental stages: egg, larva, nymph and adult. The eggs are oval and the body of the mite is almost circular with short legs. The nymphaland adult stages have four pair of legs, the larvae stage has three. In the adult, the third and fourth pairs do not extend beyond the margin of the body. The entire life span is spent on the host and requires 17-21 days for completion. Levine, (1978) described that the female parasite is larger than the male, which measures 330-600u by 250-400u comparing to male, which is about 200-240um by 150-200um.

According to Brownman (2003), the female *Sarcoptes* of the suborder *Astigmata* burrows through the epidermis producing tunnel, which is filled with eggs and feces. The pretarsusof this mite has a long unfermented stalk, the pedicel. Soulsby (1982) stated that activities produce a marked irritation, which causes intense itching and scratching witch

aggravates the condition. The resulting inflammation of the skin is accompanied by exudates, which coagulates and forms crusts on the surface and further characterized by excessive keratinization and proliferation of the connective tissue. As a result, skin becomes thickened, wrinkled and there is a concomitant loss of hair.

Another type of mite according to Soulsby (1982) causing mange in dogs is *Demodex canis* which causes demodectic mange. This is a very specialized group of parasitic mites which live in the hair follicles and sebaceous glands. The parasite is elongated, about 0.25 mm long and has a head, thorax which bears four pairs of stumpy legs and an elongate abdomen which is transversely, striated on the dorsal and ventral surface. According to Levine (1978), the mite is presumably spread by contact, however, it has been found in the blood, liver, spleen and lymph nodes. Griffin (1993) elaborated that it is transmitted to the pups from the dam during nursing within 72 hours afterbirth, but Fraser (1991) described that demodectic mites lose their capacity to invade hair follicles when off the host. The mites die briefly when outside the animal host.

Wall and Shearer (2001) added that *Demodex canis* lives as commensals, embedded heads down in hair follicles, sebaceous and meibomian gland of the skin where they spend their entire lives. Females lay 20-24 eggs in the hair follicle which give rise to hexapod larvae, in which each short leg ends in a single, three pronged claw. Octopod, protonymph, tritonymph and adult stages then follow. Immature stages are moved to the edge of the follicle by sebaceous flow, and it is here that they mature. One follicle harbours all life cycle stages concurrently. The life cycle is completed in 18-24 days.

The pathogenesis of canine demodicocis is thought to involve immunosuppression. Juvenile demodicocis which occurs between 3 and 15 months of age presents as non-pruritic areas of focal alopecia on head, forelimbs, and trunk. In puppies, the first lesions are frequently observed just above the eye. The disease is self-limiting and recurrences are rare. However, if immunosuppressive therapy with glucocoticoids is administered, the dermatoses deteriorate and may become generalized and pustular. Adult-onset demodicosis is often concurrent staphylococcal pyoderma and is a pustular form. It can be localized or generalized and the clinical features seen are erythema, pustules, crust and pruritus. The skin often may become

pigmented in chronic cases. The localized form often confined to the feet. If demodicosis occurs spontaneously in elder dogs, underlying debilitating diseases, including neoplasia, may be responsible. Immunosuppresive therapy for other diseases may also lead to canine demodicosis The female *Demodex* mite according to Belizario and de Leon (2004), favors places on the body where is wrinkled such as the wrists, elbow, feet, penis, scrotum, breasts, axillae and in between fingers. Using its short, stout, sharp pincer-like chelicerae, the mite digs and eats its way through the surface of the stratum corneum. It buries itself, excavates and creates tunnel then feed on liquids oozing from dermal cell. During the mites progress along the tunnel, it lays about 4-6 eggs and sometimes defecates while feeding.

Mateo as cited by Camalig (2005) mentioned that possible solution to problem of health care in rural areas would help develop the traditional system using herbal medicine with proven potentials. Plant species contain useful constituents found in the specific plant parts such as, roots, stem, leaves, flowers, fruits, barks or seeds or in the whole plant. These medicinal plants are sources of important pharmacological substances such as alkaloids, saponins, tannins, amino acids, phytosterol, mineral substances and other organic acids. Plants may act synthetically or antagonistically in producing the activity necessary in the treatment of diseases. The mode of preparation for most herbal medicines depends on the active ingredient extracted, route of administration, and medicinal intent, whether prophylaxis or therapy. The common preparations are infusion, decoction, powder, drops, ointment, juice, bark or fumes.

Gliricidia sepium, commonly known as madre cacaoor *kakawate*, is a leguminous tree that belongs to family Febaceae, which originated in Central America, is used in many tropical and subtropical countries as a live fencing. In the Philippines, *kakawate* is washed and pounded to extract the juice from the leaves. It is then applied to the area affected by the external parasites once or twice a day for one week. Finding also shows that it is effective in treating mange infection in dogs and other animals http://www.ansci.cornell.edu/plants/medicinal/gliricid.html).

The generic name Gliricidia refers to “mouse killer” in Latin, and the species eopithet is named from the Latin “saepes” meaning hedge*Gliricidia sepium* is a smooth, deciduous tree, 3-10 meters high. (http://www.worldagroforestrycenter/SEA.html).The leaves are 15-25 cm long

with pointed tip and rounded base. The flowers are 2 cm long and pink truncate calyx at the end of branches without leaves. The pods are narrowly oblong to oblanceolate, 10-14 cm long, about 2 cm wide and flat, and contain 6-8 seeds. *Kakawate* is often planted in the Philippines as an ornamental flowering tree because of its beautiful pink flowers; as a shade for cacao and to make living fence. The wood is hard and durable, being used locally for small house posts, agricultural implements, tool handles, etc. the branches are much used (crushed) externally to rid dogs ticks and fleas and cattle ticks.

The juice or decoction of leaves, bark or roots might be used on the skin with dermatitis or skin itching. Fresh leaves may apply to the skin as insect repellant. Crushed leaves are applied as poultice and works as a counterirritant to rheumatic pains, sprains and closed fractures. Saps of barks, leaves and roots have also been used for wound healing and treatment of scabies (Quisumbing, 1978).

The active medicinal compounds maybe tannins or other compounds such as afromosin, an isoflavan, which is reported to be an antitumor promoting agent; or the medicarpin, a pterocarpan that acts as antifungal or some isoflavins (http;//www.ansci.cornell.edu/plants/medicinal/gliricid.html). Another active compound to be present is the coumarin, which is usually found in the leaves (Baňez, 2006).

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In other research, the crude extracts of *kakawate* have been shown to have antifungal activity. *Madre de cacao* is a folk remedy for alopecia, boils, bruises, burns, colds, cough, debility, eruptions, erysipelas, fever, fractures, gangrene, and headache. Itch, prickly heat, skin tumors, ulcers, urticaria and wounds (http://www.worldagroforestrycentre/SEA.html).

Another research shows that *kakawate* leaves extract in any preparation is effective against *Demodex canis*(Caluscosin, 1991). Palpal (2001) found out in his study that *kakawate*

leaves infusion is efficient in controlling mange mites in dogs, particularly the *Sarcoptes scabieivarcanis*and *Demodex canis.*

In other research, tannins are the most abundant compound present in *Gliricidia sepium* which composes up to 40.7/ kg dry matter of gliricidia. Tannin is reported to have potential antidiarrheic, antidysnteric, antimutagenic, antinephritic, antioxidant, antiradicular, antiviral, bactericidal, cancer preventive, hepatoprotective, pesticide, psychotopic, viricidal and acaricides (http://www.ansci.cornell.edu/plants/medicinal/gliricid.html).

Another active constituent that *kakawate* has is the tannin. Tannins are complex principles found widely distributed in plants. It is used for topical administration in bedsores, weeping ulcers and eczematous dermatoses. It is also used as soothing lotion for edema of the eyelids, for skin irritation, for superficial bleeding, and to relieve itching and pain (Musser and O'Neill, 1969). In addition, tannin lotion is useful in extensive dermatoses since it remains in contact with the lesion (Kirk, 1997).

Sulfur, according to Musser and O'Neill (1969), is one of *kakawate's*active constituents and is said to be the most effective scabicides. Sulfurated topical solution is moderately alkaline, orange-colored liquid with an odor of hydrogen sulfide. When applied to the skin, it does not only destroy the parasites, but also slightly checks the growth of bacteria. The drug softens the keratin, thereby facilitating penetration to the stratum corneum, into which the mites have burrowed.

According to Brander, *et. al* (1982), sulfur is the best anti-mange dressing available for veterinarian use until the advent of cholorinated benzene derivatives, It is used extensively for routine baths for dogs. Sulfur has been the most inexpensive treatment for both bacterial and parasitic skin diseases. Booth and McDonald (1982) reported that it was also used as an aid in the control and treatment of nonspecific dermatoses, pruritus and fungus infections in the dog and cat. Further, Blood, *et. al* (1983) reported that elemental sulfur is often fed to livestock as tonic control external parasites.

Another active constituent found in the *kakawate* leaf is fats. Fats are needed in dermatology as protective agents to prevent contact with irritating substance and lubricating agents, which aid in the removal of crusts and to prevent excessive dryness and vehicles for the

incorporation of drugs in the treatment of skin diseases (Musser and O'Neill, 1969). Kirk (1979) found out that fat make for intimate and prolonged contact with the skin. Aside from the active chemical content in the *kakawate* leaves, they also reported that its leaves posses a fetid smell. When crushed, it is applied externally in order to rid dogs of ticks and fleas and ticks of cattle too.

Brander, *et. al* (1982) stated that tannic acid is considered as plants and metal astringent compound, the action is one of the precipitating sufficient proteins in the open end of the vessel to block it and encourage normal clotting mechanism.

According to Booth and McDonald (1982), astringent are drugs used locally to precipitate proteins either externally or internally. They do not penetrate deeply; their precipitant action is exerted only on the surface cells and relatively weak. After action of astringent drug upon surface tissue cell, permeability of the cell membrane is greatly reduced but the cell remains viable. Einstein, *et al,* (1994) reported that the action of astringent is on the surface cells, where they facilitate the formation of a protective layer, under which healing can proceed.

Rabena (1996) reported that *kakawate* leaves contains potent chemicals, which drives away insects and pests. Ethylene, a toxic gas is responsible in the said effect. Many other chemicals were also discovered and they all contributed to the synergistic effect as a botanopesticide and also as an antimange medicinal plant.

The tree of life has undoubtedly invaded the medical community. From its long time tag as an export winner crop, coconut, particularly its oil, is now regarded by medical experts world wide as a powerful tool against infectious diseases.

Ticzon (1996) described the coconut *(Cocos nucifera*) as high, unarmed, unbranched tree, reaching up to 25 meters high. The roots grow deep, and they are numerous. The leaves at the upper end of the tree form an apical crown, featherlike compound. The fruit is three-angled and one-seeded; the fruit is surrounded with fibers and a hard shell with three apical eyes. The inside is a white edible flesh supplied with sweetish water.

Garcia (2003) documented that Virgin Coconut Oil (VCO) is a naturally processed product from fresh coconut meat or its derivative (coconut milk and milk residue). It is the purest form of oil, is water white in color and has not undergone any chemical processing

during extraction. It contains natural vitamin E and very low free fatty acid with mild scent. It has been around for quite sometime but was not given much attention until some researchers found that it possesses the fatty acid known as lauric acid.

As cited by Brekel (2004), the fatty acids composition of VCO are the following: lauric acid-46%, myristic acid-19.9%, palmitic acid-9.8%, caprylic acid-6.8%, oleic acid-6%, capric acid-6%, stearic acid-3.4%, linoleic acid-1.3%, and caproic acid-.4%. Among them, the beneficial medium chain fatty acids are lauric, capric, caproic, caprylic and myristic acids.

Lauric acid or dodecanoic acid is an antimicrobial fatty acid with structural formula CH_3 $(CH_2)10$ COOH. Caprylic acid ($C_8H_{16}O_2$) is an antifungal short chain fatty acid and has unpleasant smell and taste used in making dyes. Capric acid ($C_{10}H_{20}O_2$) is used in artificial flavors. Caproic acid ($C_6H_2O_2$) is a liquid fatty acid that occurs in fats and oils or is made synthetically used in flavorings and in medicine. Myristic acid ($C_{14}H_{28}O_2$) is an acid found in the fats of plants and animals, and used in making soaps, flavoring, cosmetics, and perfumes. Oleic acid ($C_{18}H_{34}O_2$) is an unsaturated fatty acid used to make soap, ointments, cosmetics and lubricating oils. Linoleic acid ($C_8H_{32}O_2$) is an essential fatty acid, colorless liquid, essential to human nutrition, found in linseed and other natural oils and used in making soaps, emulsifiers, and quick drying oils. Stearic acid ($C_{18}H_{36}O_2$) is a colorless, odorless waxy crystalline fatty acid used in cosmetics, soap and lubricating medicine.

Dayrit (2003) stressed that lauric acid, a major active component of the coconut oil, is a medium chain fatty acid which is converted into monolaurin when processed by the body. Monolaurin is used by human or animal to kill lipid coated viruses such as HIV, herpes, cytomegalovirus, influenza, and other pathogenic bacteria like *Listeria monocytogenes* infecting dairy cattle and poultry, and *Helicobacter pylori* that causes ulcer. Other health wonders coconut oil could do include reduction of cardiovascular diseases, fungal skin diseases, weight loss stimulating, and boost the immune system.

The efficacy of VCO against parasites could start with the basic information claimed by Chandler and Read (1961) that the cuticle of insects which serves as the supporting skeleton is made up of very thin layer of lipid materials which is freely permeable by lipid substances.

Moreover, several authors described the mode of action of the active ingredient and were found to be complementary and are as follows.

Kabara (2004) noted that lauric acid is most potent particularly in its monoglyceride form, monolaurin. It is postulated that monolaurin solubilizes or dissolves the lipids, causing the disintegration of the covering or envelope of the disease causing organism, with the cover torn down; the integrity of the pathogen is compromised resulting to death.

Dayrit (2003) and Fife (2001) complemented that Medium Chain Fatty Acids (MCFA) are easily digested and absorbed by the body. They are not packaged into lipoproteins and do not circulate in the bloodstream like other fats, instead they are sent directly to the liver where they are immediately converted into energy.

The fact that monolaurin's activity is limited to lipid coated organisms suggests strongly that the relatively short C-12, C-10 or C-8 probably exert their action on the lipid-layered coat or plasma membrane to destabilize it or even to cause its rupture (Dayrit, 2003).

Several researches have been conducted regarding the properties and medicinal uses of Natural Coco Oil. A pre-trial experiment was made to prove that NCO is effective against these parasites prior to the formulation of NCO shampoo. Pico (2008) found out that 60-80 percent NCO shampoo is highly effective against ectoparasites such as lice, fleas, ticks and mites. The potential of NCO soap against parasites especially mites in dogs was proven by Mamauag (2007) where she concluded that 70 percent NCO soap is highly effective based on the standard criteria by Riek and Kieth (1975). In the same study, it was also observed that fleas, lice and ticks were eliminated. Likewise, the doggie odor of the experimental animals was eliminated and their hair coat became glossy and shiny as was similarly claimed by Pico, 2008 using NCO shampoo and Mamauag (2007) using NCO soap.

Coconut oil is known for its many uses such as massage oil and body lotion leaving the skin soft and smooth, relieving dry skin itchiness and other skin diseases, making the skin supple (Enig, 2003).

In the Philippines, coconut oil is much used as a vehicle for liniments in the skin medicines, external application for hair strengthening, made into cosmetics and remedy for alopecia. The lower and cheaper grades of the coconut oil which usually contain a considerable

portion of free fatty acids are used principally for making soap, lotion and hair beautifiers (Quisumbing, 1978).

When coconut oil, which is made of triglycerides, is put on the skin, it does not have any immediate antimicrobial action. However, bacteria which are always present on the skin turn these triglycerides into free fatty acids, just as it does with sebum; the result is an increase in the number of antimicrobial fatty acids on the skin and protection from the infection (Mercola, 2006).

According to Fife (2001) in the making of coconut as soaps, the soaps do not have a tallow smell or the smell of a vegetable oil. Instead, it has a nice fresh smell and yields a nice fluffy lather. Coconut oil is one of the most popular oils used in soap making. (Fife, 2001)

Soap is defined as the product of fats/oils with lye, and is not made of synthetic oils (petroleum). It is made up of a molecule that has a polar end and a non-polar end. When dirt and oil is washed from the skin's surface using soap, the dirt and oil are surrounded by the soap particles with non-polar 'tails' and the polar 'heads' point away from the dirt and oil into the water. Having hydrophilic and hydrophobic properties at opposite ends in one molecule is what makes soap such an effective cleanser. Soap allows dirt and oils to mix with water and let it all get rinsed away, thus leaving the skin surface dirt and oil free (Oostveen, 2005).

When bathing or showering, soap washes the protective layer of oil and acid off the skin. Often afterwards the skin becomes tight and dry. Adding moisturizer helps the skin feel better, but it does not replace the acid or the protective MCFA layer that was removed and the skin is vulnerable to infection at this time. Many germs survive washing by hiding in cracks and folds of the skin. Before long the skin is again teaming up with microorganisms, both good and bad. Until sweat and oil return to establish the body's chemical barrier the skin is vulnerable to infection. By using coconut oil, it can quickly help reestablish the skin's natural antimicrobial and acid barrier (Peat, 2006).

Sodium hydroxide (NaOH) is white, odorless, and non-volatile solution. It was produced mainly in three forms: 50% and 73% aqueous solution and anhydrous sodium hydroxide in the form of solid cakes, flakes or beads. Its uses include: chemical manufacturing, pulp and paper manufacturing, petroleum and gas industry, manufacture of soap and detergents, and other

cleaning products. NaOH is extremely corrosive and is capable of causing severe burns with deep ulceration and permanent scarring. It can penetrate to deeper layers of the skin and corrosion will continue until removed. The severity of injury depends on the concentration of the solution and the duration of exposure (http://www.msds.org/oshanswers/chemicals/chem_profiles.html).

VCO is ideal for skin care. Peat (2006) considered coconut oil to be an antioxidant due to its stability and resistance to oxidation and free radical formation. Coconut oil that is incorporated in the diet reduces the need for Vitamin E.

Pure VCO is the best natural ingredient for skin lotion, cream, ointment and soap. It prevents destructive free radical formation and protection against them. It can help to keep the skin from developing liver spots, and other skin blemishes caused by aging and over exposure to sunlight. It helps to keep connective tissue strong and supple so that the skin doesn't sag and wrinkle. It might even restore damaged and diseased skin. The oil is absorbed into the skin and into the cell structure of the connective tissues, limiting the damage excessive sun exposure can cause (Mercola, 2006).

Coconut oil will not only bring temporary relief to the skin, but it will aid in healing and repairing. It will aid in removing the outer layer of dead skin cells, making the skin smoother. The skin will become more evenly textured with a healthy shine.

Mercola (2006) concluded that the small molecular structure of the coconut oil allows it for easy absorption through the skin. The antiseptic fatty acids in the coconut oil help to prevent fungal and bacterial infections in the skin when it is consumed and to some extent, when it is applied directly to the skin. The only way to gain entry into the body other than the nose and mouth is by penetrating the skin.

VCO, when given to pet dogs, can also have the same multiple benefits as in human, such as: reduces bad odor and bad breath, clears up eczema, flea allergies, contact dermatitis, itchy skin etc., improves digestion, may help with arthritis and ligament problems, and many more (Puotinen, 2004).

Lawson (2009) recommended coconut oil for soap making because of its good lather, lively bubbles producing a hard bar. In addition to adding hardness, it adds a fluffy lather to the

soap. He also added that resistance to spoiling; help create a good hard soap that has a wonderful skin nourishing fluffy lather.

In the making of coconut as soaps, the soaps do not have a tallow smell or the smell of a vegetable oil. Instead, it has a nice fresh smell and yields a nice fluffy lather. Coconut oil is one of the most popular oils used in soap making (Fife, 2001).

Soaps are organic salts. Soaps are made by reacting fats or oils with sodium hydroxide or potassium hydroxide. The process of making soap is called saponification. Soaps increase the cleaning action of water (Smith *et al*., 1993).

Ordinary salt or sodium chloride is one of the most important mineral substances. Salt is used in the dairy industry, in the treatment of hides, the preservation of meat and fish, the control of ice on streets and highways, and the regeneration of ion exchange resins. In the chemical industry, salt is a source of sodium, chlorine, sodium hydroxide, hydrochloric acid, sodium carbonate, sodium sulfate and other sodium and chlorine compounds (Petrucci, 1982).

According to Tatum (2010), sodium hydroxide (NaOH) is a chemical compound, a white crystalline substance that readily absorbs carbon dioxide and moisture from the air. It is very soluble in water, alcohol, and glycerin. It iscaustica and a strong base. Commonly known as caustic soda, lye, or sodium hydrate, it is available commercially in various solid forms, e.g., pellets, sticks, or chips, and in water solutions of various concentrations; both solid and liquid forms vary in purity. The major use of sodium hydroxide is as a chemical and in the manufacture of other chemicals; because it is inexpensive, it is widely used wherever a strong base is needed. It is also used in producing rayon and other textiles, in making paper, in etching aluminum, in making soaps and detergents, and in a wide variety of other uses.

Fisher (2010) concluded that the hardness of the soap is directly related to the types and balance of oils used. Different oils make the soap harder or softer depending on their fatty acid makeup. But many soap makers add a bit of salt to their soap to help increase the hardness. This has been refuted as just a carryover from when lye was leached out of wood ashes, but his tests have shown that adding salt does indeed increase the hardness at first. It does not result in an overall harder finished product, but it does make the bar get harder quicker. The benefit of a harder bar is that it makes getting it out of the mold quicker and easier, pH is a measure of the concentration of hydronium ions in a solution. Solutions with pH

greater than 7 are basic while solutions with pH lower than 7 are acidic. A solution with a pH of exactly 7 is neutral – neither acidic nor basic. Pure water has a pH of 7 (Smith *et al*., 1993).

Petrucci (1982) stated that acid substances have a sour taste, produce a prickling sensation on the skin, and dissolve certain metals. On the other hand, the most striking feature of bases is their ability to neutralize the characteristic properties of acids. Other properties of basic substances are a bitter taste and a slippery feel.

Sodium chloride (NaCl), in pure water, would be seven or neutral in pH because the Cl is from a strong acid and the Na is from a strong base and when you mix a strong acid and a strong base together, it creates a neutral solution.

Based on chemistry, the pH level of soap can differ from the brand of soap. The balanced pH level of soap is 7. The truth is that the pH level of a bar of soap is not nearly as important as what the bar actually contains. This is because whatever is on the bar will transfer to the skin, and ultimately be absorbed by the body. All natural soap contains no chemical additives (**http://www.elmhurst.edu/~chm/vchembook/554soap.html**).

A dog's pH varies from 6.2 to 8.62 depending on the breed. The pH varies according to the breed. According to the Royal Canine Research Center measured alkaline pH skin levels and found a German shepherd to have pH values of 8.62, Golden Retriever of 7.57 and Labradors at 6.84. This is a large variation of skin pH values. With different pH values, you deal with different issues. The German shepherd's higher alkaline skin can be more conducive to bacterial proliferation than an acidic skin. To counter act this, research has shown that by limiting water loss in the skin and coat; can prevent the penetration of bacteria and any allergens. Using products that help retain moisture in the skin and coat can keep the coat healthier (Samodumskaya 2009).

Freeze-drying, or lyophilization, is, in simple terms, a dehydration technique. The aspect of the freeze- drying process that makes it different from other dehydration techniques is that dehydration takes place while the product is in a frozen state and under a vacuum. These conditions stabilize the product, minimizing the effects of oxidation and other degradation processes. Freeze-drying has become an accepted method of processing heat sensitive products that require long-term storage at temperatures above freezing (Pikal and Reiter, 2008).

Obermeyer and Obermeyer (2010) stated that natural soap will always work as soap; however, the oils do have a shelf life. The soap is best during its first six months if kept out of heat and direct sunlight. After that, it will still get you clean but may no longer have its fresh scent.

Coconut oil has a long shelf life compared to other oils, lasting up to two years due to its resilience to high temperatures. Coconut oil is best stored in solid form -i.e. at temperatures lower than 24.5 °C (76°F) in order to extend shelf life. However, unlike most oils, coconut oil will not be damaged by warmer temperatures. Among the most stable of all vegetable oils, coconut oil is slow to oxidize and thus resistant to rancidity.

As the soap cures, changes will start to happen. The colors will change slightly. Its shine will disappear, being replaced with a duller, matte surface. It will become harder. After three weeks, it can be used. But letting it cure for several months is much better. The longer the soap cures, up to about a year, the harder it will be and the longer it will last. If you set the bars on end or on side, it leaves more surface area for moisture to leave and speeds up the curing (Ward, 2007).

As cited in http://www.detergentsandsoaps.com/packaging.html, the final stage of the detergents and soap manufacturing process is the packaging. In this stage, the finished product is packed for final supply to consumer.Packaging is one of most important steps in soaps and detergents manufacturing and hence should be given prime consideration by soap and detergents manufacturers. Some of the important benefits of soap packaging are that it enhances the marketability of product, improves the appearance and attractiveness of product, increases the shelf appeal of product, increases the shelf life of product, reduces the waste during production, makes the product easy and is convenient to use.

V. METHODOLOGY

NATURAL COCO OIL SOAP

Methodology for Efficacy Trials

The Experimental Animals

Fifteen mongrel dogs positive of mange and regardless of sex and age, were used in the study. They were distributed into five treatments, based on degree of infestation and pre-treatment mite count, and replicated three times with one dog per replicate following the Complete Randomized Design (CRD). The animals were classified into light, moderate and severely infested. In light infestation, lesions are present on one region of the body only, either on the head, thorax, abdomen or limbs. In moderate infestation, lesions are present on two regions of the body, either on the head and thorax, head and abdomen, or abdomen and thoracic regions. In severe infestation, lesions are distributed all throughout the body. The animals were properly labeled based on the treatment applied.

The following treatments were used:

T_{0-}-60 % Palmoilsoap

T_{0+} - 0.3% Commercial anti-mange

T_1 - 50% NCO soap

T_2 - 60% NCO soap

T_3 - 70% NCO soap

Skin Scraping Technique

Three sampling sites measuring 4 x 4cm were prepared by clipping the hair to expose the skin lesions. Mineral oil was applied thinly over the sampling sites. Using a sterile surgical blade the sampling area was scraped by holding the blade vertically and with a gentle sweeping motion the area was scraped covering as much as the clipped area as possible, until a slight capillary hemorrhage was evident. The scraped sample was spread evenly on a clean glass slide where a drop of mineral oil was previously placed. A cover slip was placed over the sample and was examined under the microscope to identify the mites and determine the mite count.

Preparation of Natural Coco Oil

Only matured coconut was selected for the preparation of VCO. The coconut meat was grated and the cream was from the coconut meat. The coconut cream was filtered and was left

undisturbed to allow the oil to separate from the cream and fluid. The liquid portion was discarded and the cream and oil were filtered until the oil was clear.

Preparation of Palm Oil soap

Palm oil soap was made by pouring the desired amount of water into a beaker. Caustic soda was added and stirred gently; the resulting solution was called lye. The desired amount of palm oil was carefully added in small drops while stirring constantly in one direction until the mixture thickened like honey. The thickened mixture was poured into a molder, covered with cloth or wax paper and was allowed to stand undisturbed for 1 month.

Preparation of NCO soap

NCO soap was made by incorporating the desired concentrations of NCOto make 100 grams soap. The mixture was poured into a molder and was allowed to stand for a month of curing.

Preparation of Commercial Anti-mange Wash

One ml of Commercial anti-mange was added to bottle spray containing 375 ml tap water. The bottle spray was shaken to properly mix the solution.

Application of the NCO soap

The mangy dog was bathed with tap water to remove the scales and crust. The whole body was soaped liberally to form lather. The lather was allowed to remain in the coat for at least 15 minutes before it was rinsed. The bathing was done two times weekly for eight weeks.

Application of Commercial Anti-mange Wash

The mangy dog was bathed first with water to remove the dirt, scales and crust from the body. The bottle spray containing Commercial anti-mange solution was shaken and sprayed on the body giving particular attention to the site of lesions. The application was done two times weekly for eight weeks.

Standardization and Stability Test

Treatments

A total of 102 pieces of NCO soap weighing 100 grams were prepared and distributed to the different treatments for Experiment I to determine the right amount of hardener, Experiment II for the most effective freezing duration and Experiment III to determine the stability of NCO soap considering color, odor, weight and pH using the Complete Randomized Design (CRD). To determine the difference among treatment means, t - Test was used.

Amount of Hardener

Thirty (30) pieces of NCO soap were prepared based on the assigned treatments to test the right amount of hardener to be used in making NCO dog soap. The soap were distributed into six treatments replicated five times with one NCO soap per replicate. The following treatments were used:

T_1 - 1.0 gram per 100 gm of soap

T_2 - 1.25 grams per 100 gm of soap

T_3 - 1.50 grams per 100 gm of soap

T_4 - 1.75 grams per 100 gm of soap

T_5 - 2.0 grams per 100 gm of soap

Freezing Duration

After finding the right amount of hardener which is 2 grams per 100 grams of soap, 12 pieces of NCO soap were randomly assigned to the treatments to evaluate the most effective freezing time to be employed in making NCO soap. The soap were distributed into four treatments replicated three times with one NCO soap per replicate.

The treatments used were as follows:

T_0 – not subjected to freezing

T_1 – 3 hours freezing

T_2 – 6 hours freezing

T_3 – 9 hours freezing

Stability Test

Results of Experiment I which is 2 grams of hardener per 100 grams of soap and Experiment 2 which is 9 hours freezing duration were employed in Experiment 3.

Sixty (60) pieces of NCO soap were assigned randomly to the different treatments to evaluate the effect of freezing and curing duration on stability of NCO soap. The soap were distributed into two treatments replicated fourteen times with two soap per replicate. The number of soap per replicate was based on the number of weeks of observation.

Treatments used:

T_0 - not subjected to freezing

T_1 - 9 hours freezing

Procedures

Solubility Test Procedure. A small sample weighing 27.74 grams of each of the soap from the different treatments cured for two weeks and four weeks to test the right amount of hardener were separated and individually labeled. Each was soaked in 1 liter of water for 1 hour. After 1 hour, the remaining soap was weighed and the weights were properly recorded.

Freezing Duration Procedure. After the soap were placed in the molders, they were left undisturbed for 24 hours to allow saponification to complete. The properly labeled soap were placed in the freezer with a temperature of 0^0C for 3 hours, 6 hours and 9 hours. The control soap (T_0) were not subjected to freezing. The frozen soap were removed from the freezer based on the time required for each of the treatments.

Curing Procedure. After removal from molders, the soap were weighed, lined with cheese cloth and arranged in rows giving space in between rows to facilitate curing. After the withdrawal of moisture, they were transferred to racks to allow faster and even drying of the soap and were allowed to cure for 2 - 4 weeks.

Determination of Stability. The treatments assigned to be packed after 2 weeks and 4 weeks curing were weighed and packaged using plastic including the product label. The initial color and odor were recorded. Presence of moisture was observed daily for one week after unmolding and changes in color and odor were noted every week and weekly thereafter. The pH of the different treatments was tested once a week by dissolving 20grams of finely scraped NCO soap in 20 ml of sterile tap water. The pH meter was dipped into the solution and the readings were properly recorded. The weights of the soap were also taken weekly. Likewise, room temperature and relative humidity were also recorded three times daily at a given time.

Dermal Irritation Test (Draize Method). The test result was based on the laboratory test conducted by Philippine Institute of Traditional and Alternative Health Care, Cagayan Valley Herbal Processing Plant, Carig, Tuguegarao City. Ten mature guinea pigs were held captive in individual cages with food and pellet feeds without restriction. After three days of acclimatization in cage condition each were hair clipped of their back alongside the thoracic region with about 1 square inch without abrasion. Accordingly, about 0.5 g of the moistened test material swabbed in gauze pack was secured topically to the clipped area with packing tape. After 4 hours of contact time, test animals were unwrapped and the skin washed with water and dried using sterile gauze. Skin reaction was evaluated in 30 min, 24 hours, 3 days, and 7 days for reactions of erythema and edema, (Hayes, 1989).

Acceptability and Efficacy Awareness

Research Design

The study used the descriptive survey research in collection, organization, presentation, and interpretation of data to describe the samples under the study.

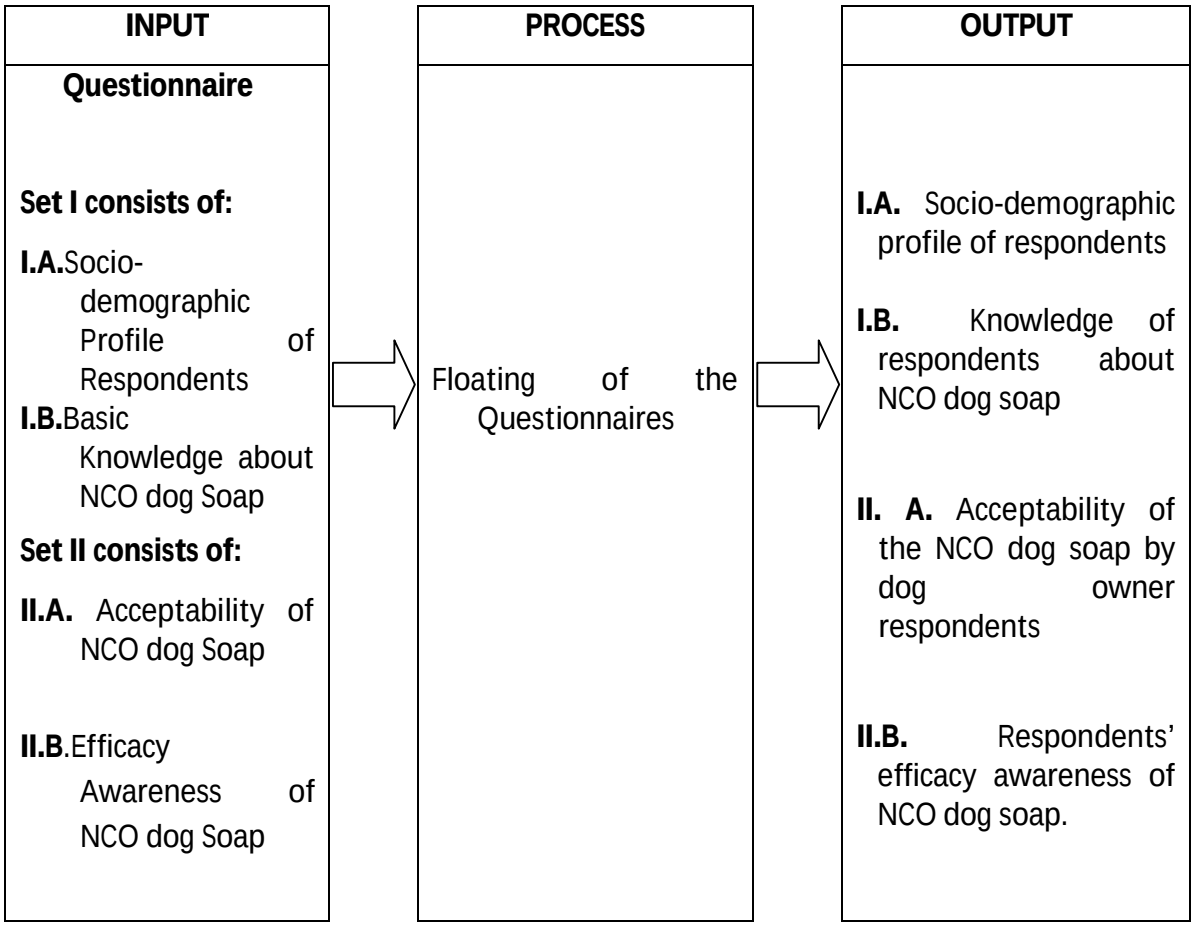

Figure 1. Research Paradigm

Figure 1 represents the interplay of variables. The paradigm relates the assessment and determination of the acceptability, efficacy awareness and the relationship of selected variables.

Instrumentation

Questionnaires were the main tool used to gather data and information. The questionnaires were composed of two sets. Set one dealt with I.A. Demographic profile of the respondents and I.B. Basic knowledge about NCO dog soap. Set two dealt with II.A. Acceptability of NCO dog soap and II.B.Efficacy awareness of dog owners to NCO dog soap.

Population and Sample

The study was conducted in barangay Sapilang, Casiaman, Salincob, Say-oan, and San Martin of Bacnotan and in barangay Carlatan, Lingsat, Pagudpud, Pagdalagan, and Dalangayan Este of the City of San Fernando from November 2010 to February 2011. Twenty percent of the total numbers of dog owners were the respondents of the study. The number of dog raisers as respondents in Bacnotan and City of San Fernando were 70 and 134, respectively with a total of 204 respondents.

Two hundred four dog owners who have mangy dogs were randomly selected for the interview in Questionnaires Set I.A. Demographic Profile and I.B. Basic Knowledge about NCO dog Soap.

During the first interview, Questionnaire Set II.A. (Acceptability of NCO soap by Dog Owners) was answered by 13 respondents who were found to be users of NCO dog soap before and during the conduct of the study.

Among the 191 respondents who were non-users of NCO dog soap, 50 percent of which is equivalent to 95 respondents were given NCO dog soap which they used in bathing their mangy dogs. The researcher monitored the dogs every week and made sure that the owners bathed the dogs as instructed by the researcher when she gave the soap. After a month of using NCO dog Soap, they answered Questionnaire Set II.A. (Acceptability of NCO dog soap) and II.B. (Efficacy Awareness of NCO dog Soap).

Pre-test of Questionnaires

Both sets of questionnaires were pre-tested in chosen barangays in the town of Bacnotan and City of San Fernando to 20 respondents on the month of November to December, 2010.

Reliability of Questionnaires

The correlation coefficient is equal to 0.8587. This indicates high reliability of the questionnaire. Thus, revision in the questionnaire is not necessary and the interview was conducted using the same questionnaire.

Interview of Dog Owners

The respondents were interviewed by the researcher based on the prepared questionnaires. Questionnaires Set I.A. (Demographic Profile) and I.B. (Basic Knowledge about NCO dog Soap) were asked first.

Respondents who were identified to be users of NCO dog soap (old formulation) before and during the conduct of the first interview were asked to answer the first questionnaire of Questionnaire Set II.A. (Acceptability of NCO dog Soap).

Fifty percent of the respondents who were identified to be non users of NCO dog soap were given NCO dog soap (new formulation) which they used for bathing their dogs. The researcher explained to the dog owners the frequency and steps on how to use the NCO soap. Aside from that, the researcher left a copy of instructions on how to use the NCO soap in case that the owner will forget it.

The researcher monitored the dogs weekly. After one month, the researcher interviewed the respondents regarding the Questionnaire Set II.A. (Acceptability of NCO dog Soap and II.B. Efficacy Awareness of NCO dog Soap).

Preparation of Natural Coco Oil - same procedure was used as specified in Study 1

Soap Making

Natural Coco Oil Soap. Natural Coco Oil soap was made by dissolving 2 g of hardener to the desired amount of caustic soda and water. The Natural Coco Oil was added to the lye solution slowly and mixed carefully until thick like honey. The thickened mixture was placed in the molder and was cured for 2 weeks.

Effect of Storage duration

Experimental Animals

Fifteen mongrel dogs positive of mange and regardless of sex and age, were used in the study. They were distributed into five treatments, replicated three times with one dog per replicate considering degree of infestation following the Complete Randomized Design (CRD).

The animals were classified into light, moderate and severely infested. In light infestation, lesions are present on one region of the body only, either on the head, thorax, abdomen or limbs. In moderate infestation, lesions are present on two regions of the body, either on the head and thorax, head and abdomen, or abdomen and thoracic regions. In severe infestation, lesions are distributed all throughout the body. The animals were properly labeled based on the treatment applied.

The following treatments were used:

T_0 - 1 month old NCO soap

T_1 - 6 month old NCO soap

T_2- 12 month old NCO soap

T_3 -18 month old NCO soap

T4 - 24 month old NCO soap

The procedure used in study 1 was adopted in the succeeding procedures

Skin Scraping Technique

Species of lice, ticks and fleas present

Preparation of Natural Coco Oil

Preparation of NCO soap - the soap that were tested were taken from previous formulations that were set aside for the purpose that matched the needed storage duration.

Application of the NCO soap

Data Gathered

Efficacy of NCO Soap

Species of mites present. The types of mites were identified based on anatomical features by examining the samples under the microscope before treatment with NCO soap.

Pre-treatment Mite Count. This was taken by counting the number of mites prior to first application of NCO soap divided by 3, the number of sites of skin scraping.

Post-treatment Mite Count. This was taken by counting the number of mites at 2^{nd}, 4^{th}, 6^{th}, 8th weeks of treatment, and two weeks after the last treatment with NCO soap. The total number of mites was divided by 3, the number of sites of skin scraping.

Percent Efficacy of NCO soap. This was measured by subtracting the mean post-treatment mite count from the mean pre-treatment mite count divided by mean pre-treatment count multiplied by 100.

The efficacy NCO soap was based on the standard criteria by Riek and Kieth (1975) which are the following:

a. 81 – 100% reduction of the count is highly effective.

b. 60 – 80% reduction of the count is effective.

c. less than 60 percent reduction of the count is ineffective.

Standardization and Stability Test

Experiment 1.

Percent Weight Reduction during Solubility Test. The right amount of hardener was computed by subtracting the final weight (Fw) from the initial weight (Iw) of the soap after soaking in 1 liter of water for 1 hour divided by the initial weight (Iw) multiplied by 100 using the formula:

$$\textbf{Percent Weight Reduction} = \frac{Iw - Fw}{Iw} \times 100$$

Experiment 2.

Freezing Time. The most effective freezing duration was determined by the length of time for moisture withdrawal observed on the surface of the soap and ease of unmolding. The rating scale used in evaluating the most effective freezing duration is shown in Table 2.

Table 1. Rating scale for freezing duration of NCO soap

Rating Scale	Descriptive Interpretation
0 - 1.0 - no freezing	Totally difficult to unmold
1.1 - 2 hours freezing	Fairly difficult to unmold
2.1 - 3.0 - 6 hours freezing	Moderately easy to unmold
3.01- 4.0 - 9 hours freezing	Very easy to unmold

Interpretation

Totally difficult to unmold - the entire sides and bottom of the soap were tightly sticking into the molder.

Fairly difficult to unmold - the whole bottom of the soap were still sticking into the molder.

Moderately easy to unmold - only the center of the bottom of the soap were sticking into the molder.

Very easy to unmold - presence of spaces between sides of molder and soap as well as on the bottom of the soap.

Experiment 3.

Stability Test. The stability of NCO soap was determined by counting the number of days that the soap remained stable in color, odor, pH and weight from the time they were packaged after two weeks and four weeks of curing.

Color of NCO soap. Rating scale for color of NCO soap was formulated based on the observed color of the soap that was compared to the color scheme of white (Appendix figure 1) downloaded from http://en.wikipedia.org/wiki/white is as follows:

Table 2. Rating scale for color of NCO soap

Rating Scale	Descriptive Interpretation
0 – 1.0	White smoke
1.01 – 2.0	Floral white
2.01 – 3.0	Old lace
3.01 – 4.0	Ivory/ Antique white
4.01 – 5.0	Linen

Odor of NCO soap. Rating scale assigned to evaluate the odor of NCO soap is shown in Table 4. The rating scale was based on the normal odor of oil as described by Petrucci (1982) and the odor of stale oil.

Table 3. Rating scale for odor of NCO soap

Rating Scale	Descriptive Interpretation
0 – 1.0	Fresh oil scent
1.01 – 2.0	Off oil scent (sour)
2.01 – 3.0	Rancid

pH of NCO soap. The pH of NCO soap was taken by adding the pH taken weekly from the different treatments divided by the total number of readings.

Weight of NCO soap. The weight of soap was taken by adding the weight noted weekly divided by the total number of readings.

Temperature and Relative Humidity. The room temperature was determined by adding the temperature taken daily divided by the total number of readings. The humidity of the room was determined by adding the humidity taken daily divided by the total number of readings.

Presence of Dermal Irritation. The presence of dermal irritation was evaluated based on the laboratory test result using the Draize Method conducted by Philippine Institute of Traditional and Alternative Health Care (PITAHC), Cagayan Valley Herbal Processing Plant, Carig, Tuguegarao City.

Acceptability and Efficacy Awareness of NCO Soap

Socio-Demographic Profile of the Respondents. This was obtained by computing the percentage of the respondents' socio-demographic profile.

Respondents' Acceptability to Natural Coco Oil Soap. This was obtained by computing the percentage of acceptability of the NCO dog soap to the respondents.

Respondents' Awareness to Natural Coco Oil Soap. This was obtained by computing the frequency of respondents' awareness on the efficacy of the NCO dog soap.

Relationship of Socio Demographic Profile of Respondents and their Response to Questionnaires. This was obtained by using the Spearman Rho analysis.

Effect of Storage Duration on the Efficacy of NCO Soap

Types of mites present. The types of mites were identified based on anatomical features by examining the samples under the microscope before treatment with NCO soap.

Species of lice, ticks and fleas present. The types of ectoparasites were identified with the aid of magnifying lens based on their morphological features before treatment with NCOshampoo.

Ectoparasite Infestation Rate. This was taken by counting the number of dogs infested with ectoparasites (lice, ticks and fleas) divided by the total number of experimental dogs multiplied by 100.

Pre-treatment Mite Count. This was taken by counting the number of mites per skin scraping site prior to first application of NCO soap divided by 3, which is the number of sites of skin scraping.

Post-treatment Mite Count. This was taken by counting the number of mites after the2nd, 4th, 6thweeks of treatment with NCO soap ,until the pos- treatment count becomes zero. The total number of mites was divided by 3, the number of sites of skin scraping.

Percent Efficacy of NCO soap. This was measured by subtracting the mean post-treatment mite count from the mean pre-treatment mite count divided by mean pre-treatment count multiplied by 100.

The efficacy NCO soap was based on the standard criteria by Riek and Kieth (1975) which are the following:

a. 81 – 100% reduction of the count is highly effective.

b. 60 – 80% reduction of the count is effective.

c. less than 60 percent reduction of the count is ineffective.

Percent Efficacy of NCO soap against fleas, lice and ticks. This was measured by subtracting from the total number of animals with ectoparasites pre-treatment the total

number of animals without ectoparasites post-treatment divided by the total number of animals treated multiplied by 100.

Information Drive on the Efficacy of NCO Soap

Available NCO soap year round, well informed public about NCO soap and additional income for the University, Campus and IVM in particular.

Other Observations

The experimental dogs were kept for observation after bathing with NCO soap to determine any reactions of the dogs to the NCO soap. Changes as a result of healing during the course of the observation period were noted. Information not included in the questionnaires which are related to the study were properly documented.

Data Treatment and Statistical Analysis

The gathered data were tabulated using means and percentages. One-way ANOVA (Analysis of Variance) was used to determine the differences among treatments for efficacy test, standardization and stability test and effect of storage duration on efficacy. To further test significant differences among factors, the Tukey, HSD method was used for efficacy test, t - Test was used for standardization and stability test .Data gathered for acceptability and efficacy awareness were arranged, tabulated and analyzed using frequency counts and percentages. For the demographic profile, basic knowledge about NCO dog soap, general acceptability as well as level of awareness of NCO dog soap, frequency count and percentage means were likewise used. Spearman Rho analysis was used to determine the relationship of the socio demographic profile of the respondents and their response to questionnaires.

KAKAWATE LEAF EXTRACT SOAP

Efficacy of *kakawate* Leaf Extract Soap

Experimental Animals

Thirty (30) mixed breed dogs, regardless of age and sex, positive of mange were used in the study. They were classified into light, moderate and severely infested and were distributed into five treatments, replicated three times, with two animals per treatment.

The treatments used were as follows:

T0- = Plain soap

T0+ = Commercial anti-mange drug

T1 = 10% *Kakawate* leaf extract soap

T2 = 15% *Kakawate* leaf extract soap

T3 = 20% *Kakawate* leaf extract soap

Preparation of the *Kakawate* Leaf Extract

Mature, fresh, clean and healthy leaves of *kakawate* plants were collected, finely chopped and ground in a blender to facilitate the collection of the leaf extract. With the use of mortar and pestle, the leaves were pounded until extract was evident. Handful of the pounded leaves was placed on cheesecloth and was wringed until all extract was collected. The procedure was repeated for the rest of the leaves.

Soap Making

Plain Soap.A lye solution was prepared by mixing cold water and caustic soda with a ratio of (29 ml water: 11 grams caustic soda). 40 % lye solution was mixed into a 60 % vegetable oil. The percentage of lye and oil was based from the total volume of the soap made. The mixture was stirred slowly in one direction until it thickened like honey. The mixture was poured into a molder, covered with wax paper and was left undisturbed for a week of curing.

***Kakawate* Leaf Extract Soap.**

Kakawate leaf extract soap was made by incorporating the different concentrations of kakawate extract corresponding to the treatments into the plain soap. The thickened mixture was poured into a molder, covered with wax paper and was allowed to stand undisturbed for a month of curing.

Skin Scraping Technique

Skin scrapings were performed prior to the application of the different treatments to identify and count the mites infesting the experimental animals. Hairs were clipped to expose the skin lesions; typically, four by four centimeters was prepared. Mineral oil was applied thinly over the sampling sites. Using a sterile surgical blade, the sampling area was scraped by holding the blade vertically and with a gentle sweeping motion, the area was scraped covering

as much as the clipped area as possible until slight capillary hemorrhage was evident. The scraped sample was spread evenly on a clean glass slide where a drop of mineral oil was previously placed. A cover slip was placed over the sample and was labeled according to treatment. The samples were examined under the microscope for positive identification of mange mites and were counted for pre-treatment mite count.Skin scrapings were also taken at two, four, six, and eight weeks post-treatment for mite counts.

Application of the *Kakawate* Leaf Extract Soap

Prior to treatment, the experimental animals were bathed with a plain soap to remove the surface debris, scales and crusts. They were rinsed thoroughly for better absorption of the medicated soap.

The *kakawate* leaf extract soap was lathered throughout the body of the animal for ten minutes, giving emphasis on the affected area. The lather was allowed to remain in the coat for five minutes before it was rinsed. The animals were bathed once a week for six weeks.

Acceptability of the kakawate leaf extract soap

Instrumentation

Questionnaire was the main tool used to gather data and information. The questionnaire consists of two sets; Set A, deals on the efficacy awareness of respondents to *kakawate* leaf extract soap; Set B, deals on the general acceptability of the *kakawate* leaf extract soap to the respondents.

Population and Sample

The study was conducted in two towns of La Union, namely; Bacnotan (Cabaroan, Casiaman, Salincob, Sapilang and San Martin) and San Fernando City (Biday, Carlatan, Lingsat, Pagdaraoan and Tanqui). One hundred thirty two (132) dog raisers were randomly selected for the survey in questionnaire A which deals with the efficacy awareness of KLES, and from the one hundred thirty two (132) dog raisers thirty two respondents after using KLES answered questionnaire B which deals with the general acceptability of KLES.

Data Gathered

Species of Mites Present. The species of mites were identified by examining the samples under a compound microscope based on the morphological characteristics before and after treatment with *kakawate* leaf extract soap.

Mean Pre-treatment Mite Count. This was taken by counting the number of mites present prior to the first application of *kakawate* leaf extract soap, divided by three, the number of sites of skin scraping.

Mean Post-treatment Mite Count. This was taken by counting the number of mites two weeks after each application with *kakawate* leaf extract soap. The total number of mites was divided by three, the number of sites of skin scrapings.

Mean Post-Treatment Mite Count Difference. This was taken by subtracting the current mite count from the previous mite count.

Percent Efficacy of *Kakawate* Leaf Extract Soap. This was taken by subtracting the mean post-treatment mite count from the mean pre-treatment mite count divided by mean pre-treatment mite count multiplied by 100. The efficacy of the *kakawate* leaf extract soap was based on the standard criteria by Riek and Keith (1975), which were as follows:

81-100% reduction of the count is highly effective

60-80% reduction of the count is effective

Less than 60% reduction of the count is ineffective.

Efficacy Awareness to *Kakawate* Leaf Extracts Soap.This was obtained by computing the frequency of awareness of the respondents on the efficacy of the kakawate leaf extract soap.

Acceptability of *Kakawate* Leaf Extract Soap.This was obtained by computing the percentage of acceptability of the kakawate leaf extract soap to the respondents

Statistical Method Used

The data gathered were tabulated using frequency counts, means and percentages. In the determination of differences and efficacy of the soap, One-Way Analysis of Variance (ANOVA) and t-Test were used. To further test significant differences of factors, the Tukey, HSD method was used. Descriptive survey was used in the determination of acceptability of the soap

VI. DISCUSSION OF RESULTS

NATURAL COCO OIL SOAP

Efficacy Trials of NCO Soap

Species of Mites Affected by NCO soap

There are 2 species of mites identified during the pre-treatment and post-treatment mite count as shown in Table 4, the *Sarcoptes* and *Demodex* species. All treatments using NCO soap (T_1T_2 and T_3) were found highly effective against mites in dogs as seen in Table 1 on the percent efficacy based on the standard criteria as interpreted by Riek and Kieth (1975). Having attained 100% efficacy and eliminated both *Sarcoptes* and *Demodex* species after 6 weeks of treatment (12 applications) Treatment 3 is the most effective concentration.

Results indicate that NCO is effective in killing mites of dogs. Monolaurin is the main active composition that makes the NCO soap an effective agent in killing the ectoparasites of dogs by penetrating deeply into the skin and into the cell structure of the connective tissues, and solubilizes or dissolves the lipids causing the disintegration of the covering or envelope of the disease causing organisms (Kabara,2004). With the cover torn down; the integrity of the pathogen is compromised resulting to death. The myristic acid in VCO works by dissolving the wax that covers the exoskeleton of the parasites, killing them by dehydration (http://www.answers.com/topic/myristic-parasites). It acts hand in hand with caprylic acid by dissolving the cell membrane causing changes in fluidity and permeability that lead to disaggregation (Fife,2001). The 70% NCO soap offers 6 weeks relief from mites earlier than the other NCO preparations. Moreover, it was also able to totally eliminate *Demodex,* the species of mite difficult to eliminate which was not eliminated by the lower concentrations of NCO soap. This implies that 70% NCO soap is the most effective concentration that can kill both *Demodex* and *Sarcoptes* species. The ectoparasiticidal effect of NCO was validated by a pet owner who tried using NCO to eliminate tick and flea and even mange of his dog (Poutinen,2004).

Table 4.Species of mites affected, level of efficacy and most effective concentration of NCO soap

Treatment	Species affected	Level of efficacy	%Efficacy/ # of application	Most effective concentration
1	*Sarcoptes* and *Demodex*	Highly Effective	100%/16	
2			98.91%/20	
3			100%/12	70% NCO soap

A couple of spoonful of NCO with each meal was introduced daily that kills lice and other parasites (Fife, 2001). The synergistic action of lauric acid and other medium chain fatty acids (MCFA) particularly caproic acid present in NCO could be the reason why it is an effective ectoparasiticide (Fife,2001)

Table 5. Cost To Totally Eliminate Mites

	Cost (Php)	No. of applications (2/wk)	Percent efficacy
T0+-Commercial anti-mange	180.00	12 applications (6 wks)	100
T3 -70% VCO	40.00	12 applications (6 wks)	100

NCO(70%) soap is more economical considering both cost and efficacy as shown in Table 5, it only costs Php 40.00 to totally eliminate mites as compared to Php180.00 for the commercial anti-mange. NCO soap is affordable, safe to use, effective, easy to prepare, readily available treatment for mange.

Standardization and Stability Test of NCO Soap

Effect of Varying Amounts of Hardener Used on NCO soap Making

Based on the result of the solubility test 2gms (T_5) of hardener per 100gms soap is the standard amount to use for making NCO soap. The harder the soap, the more economical it is and easier to unmold. NCO soap contains 70% NCO which is higher than the standard ratio of oil and other

ingredients for regular bath soap, hence it is softer. The hardness of the soap is directly related to the types and balance of oils used. Different oils make the soap harder or softer depending on their fatty acid makeup. Adding a bit of salt to soap will increase their hardness and the benefit of harder bar is that it makes getting it out of the mold quicker and easier(Fisher, 2010).

Effect of Freezing and the Most Effective Freezing Duration

Results show that soap subjected to 9 hours freezing (T_3) were very easy to unmold. In general freezing the soap for 9 hours will not only ease unmolding and shorten moisture withdrawal but also shortens curing time by 2wks from the conventional method of 4 wks. This implies shorter processing (curing period) less labor intensive resulting in decreased labor/production cost hence, increasing profit. Dehydration in the soap that were frozen was manifested by the space in between the molder and the soap that resulted in easier unmolding. The shorter the duration of moisture withdrawal by the soap subjected to freezing could be due to the fact that oils freeze faster than water whereas water thaws faster than oil allowing withdrawal of water for a shorter period of time as compared to the soap with less freezing time and more so for the unfrozen soap. These observations demonstrate the principle of dehydration by freeze-drying or lyophylization, (Picar& Reiter, 2008) dehydration takes place while a product is in the frozen state. These conditions stabilize the product, minimizing the effects of oxidation and other degradation processes. Freeze-drying has become an accepted method of processing heat sensitive products that require long-term storage at temperatures above freezing.

Effect of Freezing and Curing Time on Stability of NCO soap

Results indicate that the soap that have undergone freezing observed for a period of 12 months at the prevailing temperature of 27.84ºC and relative humidity of 73.95%, retained a lighter color longer than the unfrozen soap, as seen in Table 6 . Observations on color, (Ward, 2007) that as soap cure, there will be slight change in color. Its shine will disappear being replaced with duller, matte surface. The characteristic white color of NCO soap is due to the white color of sodium hydroxide. Change in odor of the NCO soap, for the whole

duration of 12 months observation period, was not observed. It retained its fresh oil scent, among the most stable of all vegetable oils, coconut oil is slow to oxidize and thus resistant to rancidity (Ward, 2007). That pure, fats and oils are colorless, odorless and tasteless (Petrucci, 1982)

The frozen soap recorded a pH of 7.22 for soap cured for two weeks while soap cured for four weeks had a pH of 7.10. The pH of NCO soap conforms with the standard pH of soap (Petrucci, 1982) where sodium chloride (NaCl), in pure water would be seven or neutral in pH because the Cl is from a strong acid and the Na is from a strong base and when you mix a strong acid and a strong base together, it creates a neutral solution. This pH 7 is neutral which is within the range of dog's skin pH which is from pH of between 6.2 and 7.2, (Melman, 1994). There was a slight change in the pH of the soap from an initial pH of 7.0 which is neutral. This is due to hemoconcentration as a result of the loss of moisture as manifested by decrease in weight of the soap.

Effect of Temperature and Relative Humidity

For a period of one year the room had a mean temperature and mean relative humidity of 27.94 ^{0}C and 73.95percent respectively as presented in Table 6.

Results indicate that the NCO soap remained stable for 12 months at the prevailing temperature and relative humidity for twenty-three weeks considering the stability parameters of odor, pH and a slight decrease in weight although in terms of color, soap cured for four weeks for both frozen and unfrozen soap turned more stable within the prevailing temperature and humidity than for soap cured for two weeks which manifested faster change in color. Coconut oil has a long shelf- life compared to other oils lasting up to 2 years due to its resilience to high temperatures (Ward, 2007). The change in color could have been due to the hardener added to the mixture.

Table 6.Standard procedure and stability of NCO soap

Parameter	**Standard**		**Prevailing Temperature and Relative Humidity (12mo)**
Amount of Hardener	2 grams		
Freezing Duration	9 hours		
Curing Time	2 weeks		
Stability	Initial	Final	
Weight (12 mo)	72.04	70.90	
Color (12mo)	White smoke	Old lace	
Odor (12mo)	Fresh oil scent	Fresh oil scent	
pH (12mo)	7.0	7.22	Temperature **27. 4°C**
Dermal Irritation test	Result		
Erythema	Negative		Relative Humidity **73.95%**
Edema	Negative		

Dermal Irritation Test of NCO in guinea pig

Based on the laboratory test result conducted at PITACH, occlusive patch application of NCO samples did not cause erythema and or edema reaction in guinea pig as seen in Table 6. This test confirms that NCO dog soap is safe to use.

Acceptability and Efficacy Awareness of NCO Soap

Basic Knowledge About NCO dog Soap

Results indicate that veterinary students are trusted by the people with regards to the problems of their dogs. These students help in one way or another in promoting an effective, affordable and satisfying product for the treatment of mange and elimination of external parasites of dogs resulting to worry free dog owners and happy and healthy dogs in the community. Respondents (61.54%) used NCO dog soap in their dogs for the elimination or

treatment of mites or mange while others (38.46%) used it for the elimination of other external parasites such as fleas, lice and ticks. This shows that the main reasons of the dog owners in using NCO dog soap is to treat mange and kill external parasites.

Acceptability of NCO dog Soap

Table 7 shows that majority 86.32% accepted that NCO dog soap is effective in treating mange or killing mites of dogs as manifested by the observed signs of healing of mange, like drying of wounds, growth of hair and disappearance of redness, itchiness and dead skin, after one to two weeks of using NCO dog soap. VCO when processed into soap is very effective in eliminating mites and treat mange in dogs which is comparable to the commercial preparation, (Mamauag, 2007). Aside from mites, the soap is also claimed to be effective in killing other ectoparasites of dogs. Majority of the respondents, 76.92% accepted that NCO dog soap is effective in killing fleas, 69.23% claimed that it can kill lice and 76.92% claimed that ticks can also be killed by VCO dog soap. Seventy percent (70%) NCO dog shampoo is effective against fleas, lice, mites and ticks, (Pico, 2008). Dogs were really benefited from the use of VCO on their fur and skin (http://www.ehow.com/how_5260202_use-sores-coat-problems-pet.html., it will add shine and help resolve sores and skin problems due to fleas, ticks, bites and other issues. It also acts as a repellant of these ectoparasites. Majority of the user respondents (69.23%) confirmed that NCO dog soap is effective in treating wounds of dogs. VCO is effective in treating surgical wounds of dogs, (Cueva, 2007). Coconut oil will not only bring temporary relief to the skin, but it will aid in healing and repairing of wounds, (http://www.coconut-onnections.com/skincare.htm). The coconut oil will aid in removing the outer layer of dead skin cells, making the skin smoother. The skin will become more evenly textured with a healthy shine. As to the elimination of doggie odor, almost all (92.31%) accepted that the soap is effective in eliminating doggie odor. Result reveals that VCO is really effective to minimize and eliminate bad or doggie odor of the, (Mamauag, 2007, Pico, 2008).

Table 7.Acceptability of NCO dog soap

Soap Attribute	Percent Acceptability
Effective in treating mange/killing mites	86.32
Effective in killing fleas	76.92
Effective in killing lice	69.23
Effective in killing ticks	76.92
Effective in treating wounds of dogs	69.23
Effective in eliminating doggie odor	92.31
Effective in adding luster to hair coat	92.31
Produces lather/soap suds	98.95
Easy to rinse off and is not sticky	92.31
Leaves a pleasant smell	100
Does not cause dryness to skin of handler	82.10
Does not cause dryness to skin of dog	96.84
Dissolves slowly, hence it is economical	54.74
Price is affordable	100
Color of soap is attractive	100
Texture of soap is smooth	100
Shape of NCO dog soap makes it easy to handle	100
Packaging of NCO dog soap is attractive	100
Label of NCO dog soap is attractive and informative	100
General Acceptability of NCO soap	85.20%

Majority (92.31%) claimed that NCO dog soap is effective in adding luster to hair coat, coconut oil can improve dog's skin and coat, and makes coats sleek and glossy, (Fife, 2001). The soap produces plenty of lather or soap suds as confirmed by to 98.95% of the users of NCO soap. It is easy to rinse off and is not sticky as claimed by 92.31%. It was confirmed by 100% of the respondents that the soap leaves a pleasant smell when used in bathing their dogs. As to the reaction of the soap to the skin, 82.10% accepted that NCO dog soap does not cause dryness to their skin after bathing their dogs. Majority (96.84%) accepted that the soap does

not cause dryness to the skin of their dogs. Result shows that NCO dog soap is safe to use because it does not irritate the skin of the dog and the owner. According to 54.74% of the respondents NCO dog soap dissolves slowly, hence it is economical. Other users (53.85%) who claimed otherwise is probably because some of the respondents scrubbed the soap very hard on the coat of the dog and some soaked the soap directly to the water for a long period of time while bathing their dogs which makes the soap easy to dissolve. NCO dog soap contains high concentration of oil which makes it softer than other soap. As to the price of the soap, 100% of the respondents claimed that it is affordable. All respondents accepted that the color is attractive. All (100%) of the respondents accepted that the texture of the soap is smooth, its shape makes it easy to handle, the packaging of the soap is attractive, and the label is attractive and informative. Result shows that NCO dog soap users are satisfied with regard to the color, texture, shape, packaging and label of the NCO dog soap. The general acceptability of NCO dog soap by dog owners is 85.20%.

Dog Owners' Efficacy Awareness to:

NCO dog Soap

Results as shown in Table 8, that majority of the respondents were aware of the different aspects of the soap. Most of the respondents (98.95%) were aware that the soap is effective against skin problems in dogs, 94.74% were aware that it is effective in elimination and prevention of ectoparasites of dogs like fleas, lice and ticks and 96.84% were aware that it is effective in the treatment of mange of dogs as they observed the different signs of healing of mange. As to the efficacy of the soap, 77.89% of the respondents were aware that it is noticed within 1-2 weeks of use. Majority (95.79%) were aware that it is effective against fleas, lice and ticks, 92.63% claimed that they were aware that the soap can treat wounds of dogs and 95.79% said that they were aware that the doggie odor of the dog can be eliminated by NCO dog soap, it adds luster to hair coat, and produces plenty of lather or soap suds. Respondents (85.26%) know that the soap is easy to rinse off and is not sticky, and it does not cause dryness to the skin of dog handler. Majority of the respondents (95.79%) were aware that the soap does not also cause dryness to the skin of the dog and more than sixty% (65.26%) were aware that the

soap is economical because it dissolves slowly. Regarding the price of the soap, 95.76% were aware that it is affordable. All respondents (100%) were aware that the texture of the soap is smooth. Almost all of the respondents (97.89%) were aware that the soap is easy to handle due its shape.

Table 8. Efficacy awareness of NCO soap by dog owners

Soap Attributes	Percent Awareness
Effective against skin problems in dogs	98.95
Effective in the elimination and prevention of ectoparasites	94.74
Effective in the treatment of mange of dogs	96.84
The efficacy of NCO soap is noticed within 1-2 weeks of use	77.89
Effective against fleas, lice and ticks	95.79
Effective in treating wounds	92.63
Effective in eliminating doggie odor	95.79
Effective in adding luster to hair coat	95.79
Produces lather or soap suds	95.79
Easy to rinse off and is not stick	85.26
Does not cause dryness to the skin of dog handler	85.26
Does not cause dryness to the skin of dog	95.79
Dissolves slowly, hence it is economical	65.26
Price is affordable	95.79
Texture is smooth	100
Shape of the soap makes it easy to handle	97.89
Packaging is attractive	98.95
Label is attractive and informative	98.95
Color is attractive	100
Efficacy Awareness of NCO soap	93.02%

Majority (98.95%) of them claimed that they were aware that the color of the soap, the packaging and label of NCO soap is attractive, and informative. Result shows that awareness of the respondents regarding the efficacy and the different aspects of NCO soap is high (93.02%). This information proves that the product is really effective and it is ready for commercialization.

Duration of Using NCO dog Soap When its Efficacy was First Noticed/Observed

Result shows that majority of the respondents (42.10%) noticed the efficacy of NCO dog soap within 2 weeks of use, 22.10% noticed its effect after 3 weeks, 15.79% noticed it after 1week and 11.58% noticed it after 1 month as presented in Table 9. Other respondents (8.42%) said that the NCO soap is not effective to their dogs.

Table 9. Acceptance and awareness of respondents on NCO soap

Parameter	Duration	Percentage
Healing Time	1 week	15.79
	2 weeks	42.10
	3 weeks	22.10
	1 month	11.58

This could be due to the species of mite affecting their dogs. The *Demodex* is harder to eliminate than *Sarcoptes,* (Pico, 2008, Cueva, 2007, Mamauag, 2007).Hence, it requires longer use of NCO dog soap. It could also be due to the extent of severity of mange lesions and the up keep of the dogs. Nutrition is one of the important reasons why the response is delayed and perhaps the direction on the use of the soap was not strictly followed.

Effect of Storage on Efficacy

Species of Ectoparasites Identified

There were 2 species each of the ectoparasites identified, for mites *Sarcoptes* and *Demodex* species, for fleas *Ctenocephalidesfelis* and *Ctenocephalidescanis*¸ in the case of lice *Trichodectus*and *Linognathus*species and ticks *Rhipicephalus*and *Argas*species were identified.

Pre-treatment Mite Count

The mean pre-treatment mite count of the experimental dogs per treatment are shown in Table 10. Mean pre-treatment count of the different treatments are 24.78, 14.67, 12.33, 15.33 and 14.67 for T0, T1, T2, T3 and T4 respectively. Numerical differences exist however, Analysis of Variance (ANOVA) revealed no significant difference among the treatment means which indicates that the severity of the lesions of the different treatments are comparable.

Post-treatment Mite Count

The post-treatment count of the various treatments after 6 wks of application are presented in Table 10. After six weeks of application of , all the post-treatment mite count went down to zero for all the treatments. Comparison among treatment means in all the 3 scrapings revealed no significant difference which means that all the treatments are comparable.

Table 10. Species of mite identified, mean pre-treatment and post-treatment mite count of experimental dogs in the different treatments

Treatment	Species of mite Identified	Pre-treatment mite count	Post-treatment mite count
T_0 – freshly cured	*Demodex spp.*	24.78 ns	0.0
T_1 - 6 mos storage	*Demodex spp.*	14.67ns	0.00
T_2 - 12 mos storage	*Demodex spp.*	12.33ns	0.00
T_3 - 18 mos storage	*Sarcoptes and Demodex spp.*	15.33ns	0.00
T_4 - 24 mos storage	*Demodex spp.*	16.33ns	0.00

ns – no significant difference

Percent Efficacy of NCO soap Stored at Different Durations

Table 11 shows the mean percent efficacy of NCO soap stored at different durations. After 6 wks of application all the treatments totally eliminated all the mites in all the treatments which is considered highly effective based on the standard criteria by Riek and Kieth. Results imply that the freshly cured NCO soap is as effective in eliminating mites as the

older soap stored for 6, 12, 18 and 24 moths. In other words storing the soap for 24mos does not significantly affect the efficacy of NCO soap in eliminating mites.

Table 11.Mean percent efficacy of NCO soap against mange at varying storage durations

Treatment	Mean percent efficacy
T_0 – freshly cured	100
T_1 - 6 mos storage	100
T_2 - 12 mos storage	100
T_3 - 18 mos storage	100
T_4 - 24 mos storage	100

Efficacy of NCO soap Against Fleas, Lice and Ticks

Table 12 shows that all the animals were infested with fleas, lice and ticks before the application of the different treatments of NCO soap. Results show that all (100%) the experimental dogs in all the treatments were free of fleas and lice one hour after the application of the different treatments. The ticks slowly became weak, constricted and dehydrated and after 24 hours from application of the different treatments the ticks were completely immobile. All the NCO soap stored at different durations eliminated all (100%) the fleas, lice and ticks which according to the standard criteria set by Riek and Kieth it is considered highly effective.

Table 12.Efficacy of NCO soap against fleas, lice and ticks

Treatment	Percent efficacy of VCO		
	Fleas (after 1 hr)	Lice (after 1hr)	Ticks (after 24hrs)
T_0 – freshly cured	100	100	100
T_1 - 6 mos storage	100	100	100
T_2 - 12 mos storage	100	100	100
T_3 - 18 mos storage	100	100	100
T_4 - 24 mos storage	100	100	100

ns – not significant

Observation on the action of the active ingredient of NCO which is lauric acid in the form of *monolaurin* to the ectoparasites conforms with the claim of the following authors: Kabara *et al.,* 2004 described that it solubilizes or dissolves the lipids, causing the disintegration of the covering or envelope resulting to death, Dayrit, 2000 postulated that relatively short C-12, C-10 or C-8 probably exert their action on the lipid-layered coat or plasma membrane to destabilize it or even to cause its rupture while Isaacs and Thorman (1991), and Isaacs *et al* (1990) emphasized that the lauric acid which is the active ingredient of Natural Coco Oil disrupts the lipid membranes of organisms and thus inactivates them.

The comparative efficacy of NCO soap in the different treatments be it freshly cured or stored for 2 years could be due to the observation of Ward (2007) when he concluded that coconut oil has a long shelf- life compared to other oils lasting up to 2 years due to its resilience to high temperatures.

Color and Scent of NCO soap at Different Storage Durations

Changes in color and scent of NCO soap observed during the course of the study considering storage duration are presented in Table 13.

Freshly cured NCO soap are floral white in color and changes into old lace before they reach 6months and maintains the same color till 12 months of storage. After the NCO soap were stored for 18 months they become antique white in color until 24 months of storage.

The fresh oil scent of freshly cured NCO soap which was maintained up to 12months of storage onwards, however, on the 18th month the scent of the soap changed to pleasant oil scent until the end of 24 months of storage.

These observations substantiated the claim of Ward (2007) when he concluded that among the most stable of all vegetable oils, coconut oil is slow to oxidize and thus resistant to rancidity and also to the statement of Petrucci (1982) that pure, fats and oils are colorless, odorless and tasteless.

Table 13. Color and scent of NCO soap at different storage durations

Treatments	Color	Scent
T_0 – freshly cured	Floral white	Fresh Oil Scent
T_1 - 6 mos storage	Old lace	Fresh Oil Scent
T_2 - 12 mos storage	Old Lace	Fresh Oil Scent
T_3 - 18 mos storage	Antique White	Pleasant Oil Scent
T_4 - 24 mos storage	Antique White	Pleasant Oil Scent

KAKAWATE LEAF EXTRACT SOAP

Efficacy of Kakawate (*Gliricidia sepium*) Leaf Extract Soap

Species of Mites Affected by *Kakawate* Leaf Extract Soap

The types of mite infesting the experimental animals from the different treatments were *Sarcoptes spp.* and *Demodex spp.* Sixty three percent (63%) of the experimental animals were infested with *Sarcoptes* mites and thirty seven percent (37%) with *Demodex* mites.

Table 1.Percentage reduction of mites affected by *kakawate* leaf extracts soap.

Mite	Pre-treatment	Post-treatment	Reduction (%)
Sarcoptes spp.	30.61	0.00	100.00
Demodex spp.	28.24	3.17	88.77

Table 1 show that the count of *Sarcoptes spp*. decreased from 30.61 (pre-treatment) to 0.00 (post-treatment) with a reduction of 100 %. On the other hand, the count of *Demodexspp*. decreased from 28.24 to 3.17 with a reduction of 88.77 %. This implies that mange mites were sensitive to the effect of *kakawate* leaf extract soap as revealed by the decrease in the post-treatment mite count. This finding substantiates the claim of Rabena (2006) where he reported that *kakawate* leaves contains potent chemicals, which drives away insects and pests. Ethylene, a toxic gas is responsible in the said effect. Many other chemicals were also discovered and they all contributed to the synergistic effect as a botanopesticide and also as an antimange medicinal plant.

It could also be noted that after the treatment with *kakawate* leaf extract soap, the only mite present was the *Demodex spp.* This finding was similar with the result of the previous studies about *kakawate* leaves in different preparations against mange in dogs by Palpal in 2001 and Caluscosin in 1991. Both authors observed that *Demodex spp*. was the common mite present during the post-treatment mite count. This indicates that *Demodex canis* was more difficult to eliminate as compared to *Sarcoptes scabieivarcanis*. As what Fraser (1991) reported that canine Demodectic mange, particularly the generalized form, may be persistent and often responds poorly to treatment. Pinney (1992) supported that finding wherein he found that demodecosis often requires a long-term therapy unlike the Sarcoptic mange. This is due to the characteristic of *Demodex spp*. wherein they live in the hair follicle and deeper than where *Sarcoptes spp*. could be found.

The t-test analysis revealed that the effects of *kakawate* leaf extract soap on *Sarcoptes spp* and *Demodex spp* was not significant.

Efficacy of *Kakawate* Leaf Extract Soap AgainstMange in Dogs

Table 2 shows the efficacy of *kakawate* leaf extract soap against mange mites of dogs. Dogs treated with commercial anti-mange drug (To+) showed a decrease in the number of mites from 44.11 to 0.11 registering the highest reduction which is 99.79 %, followed closely by 20% *kakawate* leaf extract soap (T3) reducing the mite count from 51.94 to 1.0 obtaining the second highest reduction percentage of 98.99 %. Likewise, 15% *kakawate* leaf extract soap (T2) had a reduction of 88.22 % while 10% *kakawate* leaf extract soap (T1) reduced the mite count to 72.30 %. On the other hand dogs treated with plain soap (T0-) had a reduction of 6.56 % was the least.

Results imply that all animals in the different treatments responded favorably, however, they vary in degree and pace. The result coincides with the findings of Palpal (2001) and Caloscusin (1991), that *kakawate* in any form of preparation was generally effective for the treatment of mange in dogs.

Table 2.Percentage efficacy of *kakawate* leaf extract soap.

Treatment		Reduction (%)
T0-	Plain soap	6.56
T0+	Commercial anti-mange	99.79
T1	10% *Kakawate* leaf extract soap	72.30
T2	15% *Kakawate* leaf extract soap	88.22
T3	20% *Kakawate* leaf extract soap	98.99

The efficacy of *kakawate* leaf extract soap might be due to its major constituents, which were sulfur, tannin, glycosides and fats. Sulfur, which is abundant and one of the active ingredients when applied to the skin, does not only destroy the parasites, but also slightly checks the growth of bacteria. The drug softens the keratin, thereby facilitating penetration to the stratum corneum into which the mites burrowed (Musser and O'Neill, 1969). Tannin on the other hand acts as an astringent. It precipitates protein either externally or internally. Its action is on the surface cells to facilitate the formation of the protective layer, under which

healing can proceed (Einstein, *et. al, 1994*). Fats are another major constituent of *kakawate,* used as protective agent to prevent contact with irritating substance, act as lubricating agent, which aid in the removal of crusts, and prevent excessive dryness (Musser and O'Neill, 1969).

For the added ingredients like sodium hydroxide and palm oil during soap making, *kakawate* leaf extract soap became more efficient against mange mites of dogs than the other previous studies about *kakawate* leaf extract. This is because *kakawate* leaf extract soap does not only target the mites, but it also cleanses the lesion, exposing the deeper portion of the skin where mange mites hide. During this process, some of these mites could already be rinsed off. Because of this action of the soap, the major constituents of *kakawate* could easily penetrate onto the stratum corneum, acting directly to the elimination of the mites.

Another factor that contributes to the efficacy of the *kakawate* leaf extract soap is its emollient action wherein it softens and moisturizes the skin. This action helps to correct the dryness and scaling of skin, and thus gives protection to irritants caused by the environment and the action of the mite itself. This also helps in the progression of healing.

Effective Concentration of *Kakawate* Leaf Extract Soap

The most effective concentration of *kakawate* leaf is shown in Table 3. Result shows that all of the concentrations of *kakawate* leaf extract soap made were effective against mange mites of dogs. This was based on the standard criteria of Riek and Keith (1975). *Kakawate* leaf extract soap at T2 (15%) and T3 (20%) were highly effective with an efficacy of 88.22% and 98.99% respectively, while T1 (10%) was rated effective with an efficacy of 72.30%. Among the three *kakawate* leaf extract soap, T3 (20%) was the most effective against mange mites of dogs. Analysis of Variance and Tukey HSD showed insignificant difference between commercial anti-mange and the soap prepared from 20% *kakawate* leaf extract (T3) indicating that *kakawate* leaf extract soap at 20% concentration was as effective as the commercial anti-mange against Sarcoptic and Demodectic mange.

Table 3.Most effective concentration of *kakawate* leaf extract.

Treatment	Percent Efficacy	Interpretation
T0- Plain soap	6.56 a	Ineffective
To+ Commercial anti-mange	99.79 b	Highly effective
T3 20% *Kakawate* leaf extract soap	98.99 b	Highly effective
T2 15% *Kakawate* leaf extract soap	88.22 c	Highly effective
T1 10% *Kakawate* leaf extract soap	72.30 c	Effective

* Means followed by the same letter are not significantly different at 5% level, Tukey HSD.

Table 4. Cost to totally eliminate mites

	Cost	Remarks
Commercial Anti-mange Preparation	P 240.00 12 ml at P 20.00 /ml	a standard size dog needs at least 3 ml per treatment.
Kakawate Leaf Extract Soap	P40.00	1 pc KLE soap was used for 6 treatments/ applications

Table 4 , shows the cost analysis of treatment in dogs, results revealed that it is much cheaper to use *kakawate* leaf extract soap compared to commercial anti-mange preparation.

Acceptability of Kakawate (*Gliricidia sepium*) Leaf Extract Soap

Efficacy Awareness to *Kakawate* Leaf Extract Soap (KLES)

Table 5, shows efficacy awareness of the respondents to *Kakawate* Leaf Extract Soap

Table 5.Awareness to KLES as an effective means in treating mange in dogs.

	f	Percentage
Aware	23	17.40
Not Aware	109	82.60
Total	**132**	**100**

The table shows that 82.6% of the respondents are unaware of *Kakawate* Leaf Extract Soap as an effective means in treating mange in dogs, while very few of the respondents, (17.4%) claimed that they are aware that KLES as an effective means in treating mange in dogs. This result reveals the absence of publicity and media campaign of the products which makes the product not popular to the community.

Table 6.Awareness on the availability of *Kakawate* Leaf Extract Soap in the market

	f	**Percentage**
Aware	12	9.09
Not Aware	120	90.91
Total	**132**	**100**

Table 6 shows that 90.91% of the respondents are not aware that *kakawate* Leaf Extract Soap is available in the market and 9.09% were aware. Few respondents claimed that they are aware that KLES are available in the market.

Results revealed in table 7 that 89.4 % claimed that they haven't tried using KLES to their dogs, while very few of the respondents who claimed to be aware regarding the efficacy of KLES tried using it to their dogs (10.6 %).

Table 7.KLES trial used in dog/s

	f	**Percentage**
Tried	14	10.6
Not tried	118	89.4
Total	**132**	**100**

Table 8 shows that 92.85 % of the respondents who claimed to be users of KLES mentioned that it is effective in treating mange of their dogs, while 7.14% mentioned that it is not effective. This was observed probably because the respondents/user did not follow the

directions in using KLES and could be due to the severity of the mange lesions of the animals which requires a longer period of medication.

Table 8.Efficacy in treating mange of dogs

	f	Percentage
Effective	13	92.85
Not Effective	1	7.14
Total	**14**	**100**

As to the source of the KLES shown in table 9, it was introduced by their neighbors and relatives, classmates, officemates and friends with 37.71, 28.57, 21.23 and 7.14 percent respectively. In order to increase awareness of dog owners to the product, KLES should be introduced to agrivet supplies and veterinary clinics.

Table 9.Source of *Kakawate* Leaf Extract Soap

	F	Percentage
Classmate	4	28.57
Officemate	3	21.32
Friend	2	14.28
Veterinary clinic	0	0
Agrivet supply	0	0
Neighbors and relatives	5	37.71
Total		**100**

Table 10 revealed that more than 1/3 (37.14%) claimed that *Kakawate* Leaf Extract Soap eliminates external parasites (kuto, pulgas, garapata), 25.71% claimed that *Kakawate*Leaf Extract Soap prevents external parasites, 20% used for prevention of mange and 17.14% used as treatment to mange in dogs. Kirk (1979) mentioned that aside from the active chemical content in the *kakawate* leaves, they also reported that its leaves posses a fetid smell, that

when crushed and applied externally can get rid of ticks and fleas of the animals. Also Rabena (1996) reported that *kakawate* leaves contains potent chemicals which drives away insects and pests. Ethylene, a toxic gas is responsible to that action of *kakawate* leaves.

Table 10.**Reasons in using *Kakawate* Leaf Extract Soap to their dogs**

	F	Percentage
For elimination/treatment of external parasites (kuto, pulgas, garapata)	13	37.14
For prevention of external parasites (kuto, pulgas, garapata)	9	25.71
For elimination/treatment of mange mite (galisaso)	6	14.0
For prevention of mange mites (galisaso)	7	20.0
Total	35	**100**

Table 11 shows the duration of the efficacy of KLES. Results revealed that 50 percent of the in respondents/users of KLES claimed that it is effective starting after 1 week of administration and only 14.29 percent answered that it is effective only after 3 weeks of administration. Respondents further confirm that healing and drying of the wounds starts during the first week of using KLES, then followed by growing of hairs. Findings of this study coincide with the results of Palpal (2001) and Caluscosin (1991) that *kakawate* in any form of preparation was generally effective for the treatment of mange in dogs; however they vary in degree and pace.

Table 11.Duration of the efficacy

	f	Percentage
1 week	7	50
2 weeks	5	35.71
3 weeks	2	14.28
1 month	0	0
Others	0	0
Total	**14**	**100**

General Acceptability of *Kakawate* Leaf Extract Soap

Table 12 shows the general acceptability of *Kakawate* Leaf Extract Soap to respondents in treating mange in dogs.

Majority of the respondents 86.72 percent claimed that *Kakawate* Leaf Extract Soap is acceptable in treating mange of dogs and only 13.28 percent of the respondents claimed otherwise. Respondents further confirmed that *Kakawate* Leaf Extract Soap is effective in treating mange in dogs, produce lather or soap suds, easy to rinse off, and does not stick to the skin of the handler. According to the respondents the shape the shape of the *Kakawate* leaf Extract Soap is a problem because it can easily slipped off from the hand of the user, with regards to the packaging 56.25% of the respondents claimed that it has a good appearance and 43.75 claimed otherwise. The efficacy of KLES might be due to its major constituents, which were sulfur, tannin, glycosides and fats. Sulfur, which is abundant when applied to the skin does not only destroy the parasites but also slightly checks the growth of bacteria (Musser and O'Neil, 1969). Tannin acts as an astringent and it acts on the surface cells to facilitate the formation of the protective layer, under which healing can proceed (Einstein, *et al*, 1994).

Table 12.General acceptability of KLES

	f	Acceptable (%)	f	Not Acceptable (%)
Is KLES effective in treating mange in dog/s?	32	100	0	0
Does it produce lather or soap suds?	32	100	0	0
Is it easy to rinse and does not stick to the skin of the handler?	32	100	0	0
Does it leave a pleasant scent?	31	96.87	1	3.13
Does it leave no irritation to the skin of the handler?	28	87.50	4	12.50
Does the coat become soft and smooth?	31	96.87	1	3.13
Does it melts slowly and water soluble?	30	93.75	2	6.25
Is it affordable?	28	87.50	4	12.50
Is the color attractive?	24	75	8	25.00
Does KLES have smooth texture?	29	90.62	3	9.38
Does the KLES have a good shape?	18	56.25	14	43.75
Does the packaging have a good appearance?	18	56.25	14	43.75
General acceptability		**86.72**		**13.28**

Information Drive on the Use of Botanical Anti-Mange Soap

Results of the efficacy awareness and general acceptability of the Natural coco oil soap and *kakawate* leaf extract soap were outstanding, thus there is a great potential for these products to be commercialized throughout the country or even abroad . Also due to the increasing number of mange cases in dogs, information drive has been intensified to inform the dog owners regarding the products.

A. DISTRIBUTORS/OUTLETS

Name	Address
1. Sulit.com	internet
2.RC Animal Land and Veterinary Clinic	San Fernando, La Union
3.Animalville Veterinary Clinic	Agoo, LU
4.Songcuan Agrivet Trading	San Fernando, LU
5.LeoPan General Merchandising	Sudipen, LU
6.Bacnotan Ice Cream House	DMMMSU, Bacnotan, LU
7.SS Songcuan Marketing	San Fernando, LU
8.DMMMSU-Research Office	Bacnotan, LU
9.TJVon Enterprises	San Fernando, LU
10.DMMMSU-IGP	DMMMSU, Bacnotan
11.Sevilla – Songcuan Trading	San Fernando, LU
12.DMMMSU-IVM Teaching Hospital	DMMMSU,Bacnotan, LU
13.Institute of Veterinary Medicine	DMMMSU, Bacnotan, LU

B. EXHIBIT/MARKET

AFFAIR/OCCASION	VENUE	DATE
TECHNO DEMO " RIMAT"	City of San Fernando Plaza	Oct. 2008
TECHNO DEMO DMMMSU Foundation Anniversary	MLUC, DMMMSU City of San Fernando	Jan. 17, 2009
ABM Entrepreneurship	DMMMSU NLUC	Yearly since 2008
Regional Symposium on R & D Highlights	Regional Symposium Venue	Yearly since 2008
DMMMSU Foundation Anniversary	Venue of Program	Yearly since 2008
Campus Foundation Anniversary	NLUC, Bacnotan, La Union	Yearly since 2008
Agriculture and Fisheries Technology Forum and Product Exhibition	SM Megamall Pasig City	Yearly since 2010
REDTI (International Conference) April 18-20,2013	VMUF, San Carlos Pangasinan	April 18-20,2013
9. 2013 North Luzon Cluster Science and Technology Fair Exhibit	Benguet State Univeristy La Trinidad, Benguet	Sept. 30-October 4, 2013
10. 19th Regional Symposium on R & D Highlights	RELC City of San Fernando, LU	August6-7,2007
11.22nd Regional Symposium on R & D Highlights	UNP, Vigan City Ilocos Sur	August 11-12,2010

C. INFORMATION CAMPAIGNS CONDUCTED

1.) DMMMSU Foundation (January 9-18, 2013)
Venue: Bacnotan and Agoo, La Union

2.) Walk-in Clients
Venue: IVM Teaching Hospital, IVM Building

3.) REDTI (International Conference) April 18-20, 2013
Venue: VMUF, San Carlos City, Pangasinan

VII. CONCLUSIONS AND RECOMMENDATIONS

Conclusions:

Based on the results of the study, the following conclusions were derived:

1. Botanical anti-mage soap such as natural coco oil soap and *kakawate* leaf extract soap can reduce or kill *Sarcoptes* and *Demodex* species of mites.

2.Botanical anti-mange soap such as natural coco oil soap and *kakawate* leaf extract soap are effective against mange mites of dogs.

3.Seventy percent (70%) natural coco oil soap (100 % efficacy) and 20 % *kakawate* leaf extract soap (98.99 % efficacy) are highly effective in treating mange of dogs and comparable to the effect of commercial anti-mange preparations.

4. Efficacy awareness of the respondents/dog owners to natural coco oil soap is 93.02 % , while 82.6 % in *kakawate* leaf extract soap.

5. General acceptability of the respondents/dog ownersto natural coco oil soap is 85.20% while 86.72 % in *kakawate* leaf extract soap.

6.Two (2) grams of hardener, and 9 hours freezing time are the best combination to standardize natural coco oil soap.

7.Natural coco oil soap cured for 2 weeks are the most stable for color, odor, pH and weight.They are stable for 1 year at room temperature of 27.94 degrees centigrade and 73.95 % humidity, does not cause erythema or edema in dogs.

8. Cost analysis revealed that using natural coco oil soap and *kakawate* leaf extract soap is much cheapercompared to commercial anti-mange drug.

9.Activities were conducted to disseminate/promote the use of natural coco oil soap and *kakawate* leaf extract soap.

10Botanical anti-mange soap was already considered as income generating project of the university

Recommendations:

Based on the findings of the study, the following recommendations were formulated:

1. Botanical anti-mage soap is recommended for the treatment of mange in dogs.
2. Botanical anti-mage soap should be used regularly especially for dogs with demodectic mange.
3. Information dissemination should be conducted to inform dog owners of the efficacy of botanical mange soap against mange.
4. Further study shall be conducted to verify the efficacy of botanical anti-mange soap against other ectoparasites of dogs.
5. The efficacy of botanical anti-mange soap should be evaluated after 1 year and 2 years of storage;
6. Botanical anti-mage soap must be commercialized and promoted in the market, and that vigorous advertisement should be done.
7. Outlets of botanical anti-mage soap like agri-vet supplies should be increased to establish high awareness and usage of the soap.
8. In using the soap, users should follow the instruction properly to attain the best.

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