UNIVERSITI MALAYSIA KELANTAN

Manipulation Of The Pineapple Leaves For The Development Of Silage Suitable For The Cattle Feed

Mohamad Nabil Bin Razali

F18A0059

A report submitted in fulfilment of the requirement for the degree of Bachelor of Applied Science (Animal Husbandry Science) With Honours

FACULTY OF AGRO-BASED INDUSTRY
UNIVERSITI MALAYSIA KELANTAN

2022

DECLARATION

I hereby declare that the work embodied in here is the result of my own research except for the excerpt as cited in the references.

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ACKNOWLEDGEMENT

Bismillahirrahmanirrahim. All praise be to Allah, the Lord of the world, the Master of the Day After, who has given unlimited mercy to His creations which show Allah's love is spread around.

This experiment was partially supported by faculty of Agro-based Industry of Universiti Malaysia Kelantan. A special gratitude is presented by the writer to Madam Hazreen Nita Binti Mohd Khalid which is writer's coordinator of final year project and for the Husbandry Sciences courses. Then, the writer expressed biggest gratitude to Dr. Raimi Binti Mohamed Redwan as writer's supervisor who guiding writer in completing this study and not forget to the writer academic advisor, Prof. Madya Dr. Seri Intan Binti Mokhtar who supporting the writer all this time.

The writer also would like to thank beloved parent, Mr. Razali & Mrs. Mazlina for all their support and prayer for the writer in completing this project. All writer's friends from the same course and even different courses also have contribute for various aspect during this study were carried out. Without them, will be more difficulties for the writer in completing this study.

Finally, the writer wishes gratitude to all lab assistants that has help the writer when carried the study on the laboratories. They have made the way ease by providing a high standard of service all students. Without all these people, the writer cannot reach this stage and moment. May Allah bless all of us. Thank you.

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LIST OF SYMBOLS

ha	Hectare
NaOH	Sodium Hydroxide
cm	Centimetre
℃	degree Celsius
H_2SO_4	Sulfuric Acid
H_3BO_3	Boric acid
HCl	Hydrochloric acid
g	Gram
KOH	Potassium hydroxide
ml	Millilitre
UV	Ultraviolet

LIST OF ABBREVIATIONS

EM	Effective Microorganisms
PM	Pineapple leaves silage with molasses only
PEM	Pineapple leaves silage with molasses and EM
UMK	Universiti Malaysia Kelantan, Jeli
MPIB	Malaysian Pineapple Industrial Board
WSC	Water-soluble carbohydrate
DM	Dry Matter
MA	Moisture analyser
CP	Crude Protein
CF	Crude Fat

ABSTRACT

Pineapple leaves has been identified as potential alternative feed especially to solve the issue of limited cattle feed supply. Abundant amount of pineapple leaf waste from plantation introduced a new cattle feed source and make it easier to obtain. However, the high fibrous level in the leaves need to be reduced prior to the cattle consumption to prevent digestive problems on the cattle. The purpose of this study was to investigate the suitability of pineapple (*Ananas comasus*) leaf silage as alternative feed for the cattle by using two treatments, one with the additive of molasses and another one is with the addition of molasses and effective microorganisms (EM). Two groups of silage with 5% of molasses only (PM) and 5% of molasses with addition of 5% of EM (PEM) were prepared. Nutrition composition (dry matter, moisture content, ash content, crude protein, crude fibre and crude fat) of both silages were determined by proximate analysis while reducing of sugar was determined by the reaction of alkaline cooper reagent with phosphomolybdic acid in Folin-Wu method. Proximate analysis showed that the PM silage has high amount of moisture content, ash content and crude fat while PEM silage was higher in dry matter, crude protein and crude fibre. Addition of EM on pineapple leaf proved that energy in silage can be improved by the increased of dry matter and crude fibre along with crude protein. The result showed that the reaction of lactic acid bacteria with yeast from EM solution may have facilitated the fermentation of the pineapple leaves as indicated by the increased of reducing sugar concentration than PM. For reducing sugar, both silages have undergone successful fermentation process as indicated by low concentration of sugar with PM has lower concentration than PEM. However, there is no statistically significant different between PM and PEM silage on all elements analysed (DM, moisture content, ash content, crude protein, crude fibre, crude fat and reducing sugar). Nonetheless, but the result suggests that the addition of EM increased the protein and sugar content while PM has higher value of moisture content, ash content and crude fat.

Keywords: pineapple leaf, silage, EM, feed, fibre

ABSTRAK

Daun nanas telah dikenal pasti sebagai makanan alternatif yang berpotensi terutamanya untuk menyelesaikan isu bekalan makanan lembu yang terhad. Lebihan sisa daun nanas dari ladang menghasilkan sumber makanan lembu yang baru dan memudahkan untuk diperoleh. Walau bagaimanapun, tahap serat yang tinggi dalam daun perlu dikurangkan sebelum penggunaan lembu untuk mengelakkan masalah pencernaan pada lembu. Tujuan kajian ini adalah untuk menyiasat kesesuaian silaj daun nanas (*Ananas comasus*) sebagai makanan alternatif untuk lembu dengan menggunakan dua rawatan, satu dengan tambahan molases (sirap hitam) dan satu lagi adalah dengan penambahan molases dan mikroorganisma efektif (EM). Dua kumpulan silaj dengan 5% molases sahaja (PM) dan 5% molases dengan penambahan 5% daripada EM (PEM) telah disediakan. Komposisi pemakanan (jisim kering, kandungan kelembapan, kandungan abu, protein mentah, serat mentah dan lemak mentah) kedua-dua silaj ditentukan oleh analisis proksimat manakala pengurangan gula ditentukan oleh tindak balas reagen alkali tembaga dengan asid fosfomolibdik melalui kaedah Folin-Wu. Analisis proksimat menunjukkan bahawa silaj PM mempunyai jumlah kandungan kelembapan, kandungan abu dan lemak mentah yang tinggi manakala silaj PEM lebih tinggi nilainya pada jisim kering, protein mentah dan serat mentah. Penambahan EM pada daun nanas membuktikan bahawa tenaga dalam silaj boleh diperbaiki dengan peningkatan jisim kering dan serat mentah bersama-sama dengan protein mentah. Hasil kajian menunjukkan bahawa tindak balas bakteria asid laktik dengan yis daripada larutan EM mungkin telah memudahkan penapaian daun nanas seperti yang ditunjukkan oleh peningkatan kepekatan gula PM. Untuk pegurangkan gula, kedua-dua silaj telah menjalani proses penapaian yang berjaya seperti yang ditunjukkan oleh kepekatan gula yang rendah dengan PM mempunyai kepekatan yang lebih rendah daripada PEM. Walau bagaimanapun, tidak ada perbezaan statistik yang signifikan antara silaj PM dan PEM pada semua elemen yang dianalisis (DM, kandungan kelembapan, kandungan abu, protein mentah, serat mentah, lemak mentah dan pengurangan gula). Walau bagaimanapun, hasilnya menunjukkan bahawa penambahan EM meningkatkan kandungan protein dan gula manakala PM mempunyai nilai kandungan kelembapan, kandungan abu dan lemak mentah yang lebih tinggi dari PEM.

Kata kunci: daun nanas, silaj, EM, makanan haiwan, serat

CHAPTER 1

INTRODUCTION

1.1 Research Background

Pineapple industry has been established in Malaysia for more than 100 years. Pineapple (*Ananas comosus*) not a native crop species in this country. It was introduced by the Spanish back in the 1600s and become the oldest agro-based export activity in this country. States that are mostly covered with peat soil area like Johor, Sarawak, Kedah, Perak, Kelantan and Terengganu are heavily involved in pineapple planting for domestic purpose due to the soil's marginality with any other crop farming. Planting of pineapple on peat soil are mostly in Johor not in other states (Anem, 2014). Continuity to the large-scale farming activity has result to the abundant amount of pineapple by-product waste especially the pineapple leaves. Pineapple leaves has a high potential in becoming an alternative fibre source in various industry.

Malaysia ruminant industry are still developing but in slower rate compared to other livestock industry. More than 60% of people in Malaysia consume beef in their diet (Ariff, Sharifah & Hafidz, 2015). In 2020, each person consumed around 5.42 kilograms of beef (OECD, 2021). It is estimated that this number will keep increasing for the coming years. Reflected to this expansion, cattle meat demand also keeps increasing. Despite of importing beef from other countries, local farmers always search for solution to improve the country beef production. One of the methods is supplying nutritious feed to the cattle in order to boost their cattle meat production. Unfortunately, formulated feed price in the country seems to be unpredictable. In certain seasons, the price for cattle feed keeps increasing and left the farmer to the cast down condition. As an alternatives way for the farmer to fulfil their herd diet, they chose several crops to be fed to the cattle as a silage, hay and haylage. It is one of recommended ways in reducing the feed cost.

In silage making industry, there are few popular grasses, crop and forages that casually were chosen such as Napier grass (*Pennisetum purpureum*), Guinea grass (*Panicum maximum*), sugar graze and sweet corn stover (Chin & Idris, 2000). Alternatively, high fibre content in pineapple leaves may be used in silage making. Pineapple could be used as a low-cost alternative source in aiding the cattle breeder in maintaining their livestock's feed supply while keeping their feed cost still under control despite of its high fibre content. This application could also become a beneficial solution for the pineapple farmer in managing the leaves waste with an eco-friendly and profitable way.

1.2 Problem Statement

Cattle cannot digest fibre by themselves because they do not produce fibre digestive enzyme. They will rely on the microorganisms in their stomach to produce digestive fibre enzymes (cellulase) to break down the fibre. Cattle will aid the microorganism to ease the fibre break down by rumination process which will physically break the fibre to smaller size. As the pineapple leaves are fibrous, it can be harmful to the ruminant cattle when consuming them in uncontrolled amount. There also possibility of less effective fibre for the digestion even the fibre level in pineapple leaves is high. The same problem also occurred to the soybean hull that is known for its high fibre level (Chee *et al.*, 2005).

Subsequent to those issues, pineapple leaves need to undergo some process of manipulation towards achieving the suitable condition such as silage making with additive of molasses and EM for cow. Theoretically, pineapple leaves could produce a much better fibre level and effective for the ruminant digestion. It also could provide choice for the cattle breeders. However, there also worries when the fermentation complete, pineapple leaf silage could not provide enough other nutrients required for the cattle. It is important for us to make sure the chemical composition of the pineapple leaves after fermentation process in the silage making procedure are suitable for cattle intake. Combination of lactic acid bacteria and yeast in EM can stabilize the fermentation and produce high quality silage besides of improving feed value on the silage. Therefore, investigating on the best manipulation either using molasses only or together with EM towards making pineapple leaves in silage making could produce a bright path for various parties such as animal feed producers, pineapple farmers and cattle breeders.

1.3 Hypothesis

H^0 = There is no significant difference between the chemical composition of the pineapple leaves between pineapple leaves silage with molasses only (PM) and pineapple leaves silage with molasses and EM (PEM).

H^1 = There are significant differences between the chemical composition of the pineapple leaves between pineapple leaves silage with molasses only (PM) and pineapple leaves silage with molasses and EM (PEM).

1.4 Scope of The Study

The study was conducted in UMK Jeli Campus using the pineapple leaves collected from the local pineapple farms. The leaves were mixed with other ingredients which are EM with molasses or molasses only to form optimal fermentation condition required for silage making. The mixture was kept under an aerobic condition by sealing it in a container. Fermentation process was done in room temperature, and this process took 4 weeks to complete. When complete, the chemical properties of the pineapple leaves silage were analysed to compare the nutrient content between EM treatment and molasses only.

1.5 Significance of The Study

Pineapple waste has been managed by a few innovation ways to increase the sustainability of pineapple farming. It was used as textiles material for making fabric and for the green-energy resources which is biomass. Despite that only small amount of pineapple leaves were channelled to those sectors. Most of the waste were disposed by burning as a quick solution to dispose the huge amount of residue.

On the other hand, cattle breeders are facing the increasing of feed cost due the global issues. They tried to rely to the alternative feed to reduce their operational cost. Unfortunately, alternative feeds could also run short sometimes and this may be due that some of the feeds were not commercially grown or available at all seasons throughout the year. Some of them must be imported from other countries.

Certainly, this study could offer the solution for those problems. The connection between pineapple farmers and cattle breeders could be pioneered to find way of the solution for their problems. Pineapple farmers could manage their waste with efficient way while cattle breeders could have local ingredients silage for their herd consumption.

1.6 Objective

1. To develop suitable silage for cow using pineapple leaves as an alternative main ingredient.
2. To compare the chemical composition of pineapple leaves between EM treatment with molasses and molasses only.

CHAPTER 2

LITERATURE REVIEW

2.1 Pineapple Farming in Malaysia

Malaysia has started pineapple farming back in the 1888 by applying intercropping system. The fruit has become one of the most promising demands in the country (Nazri & Pebrian, 2017). In 2019, Malaysia has produced RM502mil worth of pineapple nationwide. Smallholders farm also contributed to that value alongside with the commercial farm. According to statistic, a total 16,653ha of plantation nationwide were recorded by Malaysian Pineapple Industrial Board (MPIB) in 2020 (Ahmad, 2021). Johor leads other states in producing pineapple with 58% of the country production (Anem, 2014). Until now, 434,811-ton metric of pineapple production has been recorded by Malaysian Pineapple Industrial Board (MPIB, 2020). As a result, from the expansion in the farming, the pineapple residue has also increased to the excessive amount. In pineapple farming, only the fruit is harvested and the plants are then pruned off in order to encourage suckers' development. Consequently, many of the fibre were left littered after harvesting process.

Figure 2.1: Pineapple leaf waste (Source: Reuters, 2021).

2.2 Pineapple Leaves Fibres Chemical Composition

Pineapple leaves fibre were made up from several components. Those components are cellulose, hemicellulose, holocellulose, lignin and extractives, such as gum and resin, are examples of extraneous content. From chemical analysis done by Zawawi, Mohd, Angzass and Ashuvilla (2014) indicate that pineapple leaves fibre has a high holocellulose's percentage with 85.7% more comparing to corn stalk and Napier grass. Holocellulose is a combination of cellulose content and hemicellulose. Holocellulose content for pineapple leaves is higher than other popular roughage or silage for cattle such as corn stalk, Napier grass, and banana stem with their percentage are 82.1%, 80.4% and 87.6% respectively (Khalil, Alwani & Omar, 2006). High holocellulose content in pineapple leaf are corresponding to its high cellulose content (66.2%) but its hemicellulose content (19.5%) is low. Comparing to corn stalk and Napier grass, they have far lower cellulose content, 39.0% and 12.4% respectively even their hemicellulose is higher than pineapple leaf with 42.0% and 68.2% for each. Meanwhile, hemicellulose content and lignin content in pineapple leaf are lower than corn stalk (7.3%) and Napier grass (10.8%). Lignin acts as an adhesive to hold the cellulose in the fibre together. Lower lignin content increases fibre strength and makes it more difficult for break down (Tran 2006) Therefore, pineapple leaves with 4.2% of lignin content are harder to break than corn stalk and Napier grass.

Zawawi *et al.* (2014) also showed that the ash content in pineapple leaf is around 4.5%. The ash content in pineapple leaf is lower than corn stalk and Napier grass with 24.9% and 14.6% respectively. Ash was described as total amount of mineral content either in diet or forage. Ash is important for the cattle especially in producing milk by supplying supplement of minerals. If the ash content of a forage is high, there could be a high probability that it was contaminated to the soil and it is not a desirable condition to feed cattle (Hoffman, 2005). Thus, comparing to corn stalk and Napier grass ash content, pineapple leaf may be the best choice for cattle consumption.

In term of 1% of NaOH solubility composition in pineapple leaf, Zawawi *et al.* (2014) found it was lower percentage (39.8%) than corn stalk that has highest percentage (69.6%) followed by Napier grass with 52.0 %. NaOH is efficient in improving the digestion of some fibre fractions of all silages except for corn silage (Staples, 1985). The solubility of 1% NaOH will show the fibre degradation proportions. Pineapple leaf's low percentage of 1% NaOH solubility which may ease the fibre degradation in rumen digestion.

The last chemical composition in pineapple leaf according to Zawawi *et al.* (2014) is moisture content (81.6%). Moisture content in pineapple is high and could cause a problem in storing. In the early spring harvested silage crops, there are also a high moisture content around 75% to 85% and causing problems in storing without reducing its natural moisture content (Gordon, 1967). High moisture in pineapple leaf content could results in storage losses for the silage.

2.3 Cattle Farmer's Cost Saving

Animal feed cost has arisen to an enormous amount around the world affecting every scale of farming. In the late of December 2020, United States expected that feed price could increase up to 12% (Reus, 2020). Increasing of feed ingredients prices such as soybean meal and corn has squeezed farming to use alternatives feed supply for their herd. Many efforts can be seen as a way in reducing the feed cost. The use of agro-industry by-products as ruminant feed is one of the techniques for increasing feed supply while keeping the animal to receive required nutrient and perform well (Mirzaei & Maheri, 2008). Pineapple leaf as an agro-industry by-product is seen as a potential resource to become alternative feed for cattle in Malaysia. In addition, pineapple leaf could be found in a huge amount due to high farming activity around the country. Suitable development in silage making will produce a good result to realize the idea in reducing cattle farmer cost (Suphalucksana, Sangsoponjit and Srikijkasemwat, 2017).

2.4 Cattle Nutrient Requirement

Cattle usually were reared for two main purposes but nowadays they are mostly reared for either meat or milk sources. Those cattle development heavily depend on their diet. Diet that contains fully required nutrient will influence cattle in performing better. Nutrient requirements are different according to the cattle life stage, age, weight, rate of growth, environmental conditions, breed, gender and other factors. Nutrient requirements are including water, protein, energy, vitamins and minerals. Therefore, nutrient required for different purpose of cattle obviously are different. Silage that was made from pineapple leaves must be in line with the ruminant's diet requirements or at least for the minimal amount in supporting the cattle main feed.

2.5 Fibre Digestion in Ruminant

Fibre is important for cow in digesting feed. It is not only to fulfil the nutrient requirement for them but also maintaining their health. Fibre will be digested at two sites which are rumen and large intestine of the cow. This is due to the reliance of cattle on the microorganisms that can produce fibre-breaking enzymes only available at those sites. Effectiveness digestibility of fibres are affected by some factors. Fibres could be digested effectively when low pH value reached at those sites. At that pH, those microorganisms could inhibit and perform better in fibres digesting process while maximizing the forage intake. Other than that, size of the fibres particle also affects the effectiveness. If the fibres were chop too long, it could damage the rumen wall. Meanwhile, if the fibres were cut too short, it could not increase the digestibility effectiveness and cannot promote the rumen health. Thus, fibres must be chopped at the optimum size to increase its digestibility effectiveness.

Fibre’s content such as cellulose, hemicellulose and lignin will form a clump that could result in energy losses for the cattle consumption. In cattle or ruminant digestive system, rumen is responsible in digesting the cellulose and hemicellulose from 60% and up to 90% in the gastrointestinal tract at the digestion site. However, the digesting process are depending on the presence of the lignin. Lignin will associate with the carbohydrate fraction and form a supramolecular net. The net will prevent disruption of the chemical bond between the carbohydrate fraction by microorganism enzymatic complexes in ruminants. Lignin can be analyse qualitatively and commonly using

Klason and AcBr methods (Lin & Dence, 2012). In summary, high fibre content in pineapple leaf may cause difficulties for ruminant to digest its fibre. Thus, the leaves need to undergo fermentation as a pre-treatment to reduce its lignin content to facilitate digestion for ruminant.

2.6 Silage in Animal Feed Industry

Silage can be made by forages, crop residues or any agricultural or industrial by-products that was conserved by natural or artificial acidification. Silage is used as animal feed for periods when insufficient of feed supply occurs. The process of making silage is known as ensilation (Mannetje,2016). This process has been applied by farmers from ancient times. Some of the common forages and crops of the silage around the world are, maize, grasses, legumes and sorghum.

Basically, silage is more popular in the temperate regions due to their seasons and well relevant concept of the ensilation. As the time flows, it can be seeming tropical country such as Malaysia has exploited into the silage making as a long period plan in sustaining the country feed supply at any scales of cattle farmer. One of the main factors for silage making in this country is that the process is less dependent to the weather conditions compared to the haymaking that always piloting on the weather at the temperate areas (Wong, 2000). Currently, there a few silages making development in Malaysia such as corn silage and Napier grass. More venture is needed to explore their sustainability and potential to the country animal feed industry.

2.7 Ensilation Process

In making silage there are some primary components that need to be taken carefully to produce a good silage. According to Mannetje (2016), the components are the suitable levels of water-soluble carbohydrate (WSC), oxygen exclusion, rapid pH decrease, crop buffering capability, withering of natural fibres, optimum temperatures, and the high of nutritional value of the material to be ensiled.

Commonly, the pH value for a crop's buffering capability is its ability to withstand a pH transition resulting from the addition of an acid or base. As for the ensiling, buffering capacity can be described as milliequivalents of acid required per kg of DM to reduce the pH from 6 to 4.

Mannetje (2016) also stated that ensiling process consist of five main phases to form a suitable silage for animal consumption. Those five phases are harvesting and storage phase, followed by the aerobic phases, fermentation phase, suitable phase and finally is the feed-out phase. Unfortunately, feed-out phase will not be run in this research because our main objective is developing a stable silage. Harvesting phase should be done by one or two days to prevent the leaves from damp. Other phases could take for 10 days to 3 weeks to established stable silage. The longer it takes in storing silage will changes the nutritive value as an example, cows that were fed with silage that were stored for more than 3 months will have more milk production (Amaral, 2013). It is advisable to feed the silage to animal after 3 or 4 weeks from harvesting.

2.8 Molasses as Incentive on Silage

Silage's nutritive value can be seemed to be low due to its natural composition of the ensiled material itself even there are no error in the silage making process according to the find from Hamilton, Catchpole, Lambourne, and Korr (1978). This attribute can be seemed to the silage made from tropical grass. Silage also has its own odour characteristics that may displease animal from consume it without addition of fresh forage or molasses.

The uses of additives on silage by farmers in twentieth century with one purpose which is to produce a quality silage (McCullough, 1977). Molasses is one of them and has shown to be a good additive to the silage. This has been proved by Watson and Nash (1960) when the uses of molasses in silage additives or supplement were advised by country such as United States, Netherlands, United Kingdom and Germany. Molasses is important for the efficiency of the silage preservation. The main factor affecting the quality of fermentation process is protein concentration in crops itself (Wilkinson, Bolsen & Lin, 2003). If the crop s were low in protein, addition of molasses could improve the quality fermentation process. It is important to know the suitable amount of molasses to be used for an efficient preservation to occur.

Based on the study conducted by Arbabi and Ghoorchi (2008), all silages produced with addition of molasses has better appearance characters which are smells, colour and structure compared to the control group. The study also found that silage produce with 2.5%, 5% and 7.5% of molasses has lower pH value compare than control groups which are a very good characteristic for silage . Besides, silage produce with 5% of molasses content the highest crude protein than other treatment including control group. In the *in situ* ruminal DM digestibility coefficient, treatment with 5% of molasses has the highest coefficient while treatment with 7.5% of molasses the lowest. Low coefficient on the 7.5% molasses resulted from the higher nutrient to be utilized and it will take longer time in the rumen to digest.

Finding from Arbabi and Ghoorchi (2008) confirm again Wilkinson, Bolsen & Lin, (2003) that amount of molasses used as additive to silage are important in order to produce quality silage too much or too little amount of molasses does not guarantee to produce a good quality. A good quality silage can be produced by the optimum amount of additional molasses and those amounts vary based on other factors.

2.9 Effective Microorganism (EM)

Despite the used of molasses as additive to the silage, Higa in 1991 found the concept of using effective microorganism (EM) on the silage. The mixed of naturally existed bacteria will be found in the EM. Those bacteria were developed scientifically to improve the plant and soil microbials diversity. Some of the species found in the EM are lactic acid bacteria and yeast (Higa, 1991). Other bacteria are *L. plantarum* that are majorly used in the South Korea farming by locally as traditionally developed EM. (Amanullah, Kim, Lee, Joo, Kim, & Kim 2014).

EM could increase the quality fermentation process through bacteria inoculants. Amanullah *et al*. (2014) found that barley silage with EM has the highest DM percentage compared to no bacteria inoculant (CON) and *L. plantarum* silage (LP) with 46.2%, 43.1% and 43.5% respectively. High DM percentage indicate there are good nutrient preservation and fermentation process on the silage. Their study also found EM silage has lowest yeast and mould found on the silage. This because high amount of acetate and propionate concentration in EM silage can reduce occurrence of yeast mould. Both EM silage and LP silage has lower pH value and ammonia compared to the CON silage. Further study may assist to explore more bacteria inoculant effect to improve silage quality especially bacteria inoculant by EM which are still have a vast area to be discovered.

2.10 Pineapple Waste as Animal Feed

Pineapple wastes has been well practiced as animal feed in tropical and sub-tropical regions. As one of the by-products in agro-industry, pineapple leaves are among the major crop residue from pineapple plantation that can be benefited for animal feed. During harvest season, only 25%-30% of the pineapple plant were extract for further processing. A huge amount of pineapple farming leftovers in form of by-product which are stem and leaf could be found at the farming or harvesting site. (Suksathit, Wachirapakorn & Opatpatanakit, 2011). In order to prevent pollution, those by-products can be preserved and developed into animal feeds. High fibre content in pineapple waste makes it suitable for sourcing energy and providing a good digestive feed for various types of animals such as poultry, broiler and cattle (Sumerhayari & Setyawati, 2005; Mandey, Tulung, Leke & Sondakh, 2018; Gowda *et al.*, 2015). Pineapple leaf waste as pellet is suitable for dairy cow due its high fibre content (Buliah Jamek, Ajit & Abu, 2019).

CHAPTER 3

METHODOLOGY

3.1 Silage Preparing and Making

Pineapple leaf waste were from farm nearby University Malaysia Kelantan, Jeli Campus for around 7 kg. Those leaves were chopped into small pieces (1-3 cm) by using a chopper at Animal Laboratory. Some of the leaves were chopped for more than one due to the leaves texture that hard and thick compared to the grass leaves. The leaves were wilted under the sun first before storing. Wilting or drying were done to achieve 30% to 50% Dry Matter (DM). Squeezing technique were used to determine whether 30% DM of the leave was achieved or not after wilting. The 1-2 cm cut samples were squeezed in hand tightly for 30 to 60 seconds. Then, quickly open the hand and observe the condition of hand and samples. If no water drips from hand, the palm of the hand begins to moist rather than wet and the sample does not return to its original form quickly, 30% DM approximately achieved after wilting.

The wilted leaves were divided into two parts with weight around 3-3.5 kg each. 5% of molasses were added to both parts and only one part were added with diluted EM as stated in the Table 3.1. Next, the sample undergo storing process where it was stored in airtight container compactly and seal tightly to prevent air flowing through the bag (Moran, 2005).

Storing was done as soon as possible to prevent the leaves from become wet again and disturb the ensiling process and produce unstable silage. The containers were stored at safe place that far from sunlight, children, animals and other factors that can cause punctures to the bag. During night, a sheet of plastic was used to cover the bag as a safety step from letting warm air flowing through it. After 4 weeks of storing, proximate analysis was carried out.

Table 3.1: Pineapple leaves silages composition

Group	**Composition (%)**	
	Molasses	**EM**
PM	5	-
PEM	5	5

3.2 Proximate Analysis

The quantitative study of macromolecules in food is referred to as proximate analysis. In this study, several techniques will be combined to analyse and study the pineapple leaf silage dry matter, moisture content, ash content, crude protein, fat, fibre, nitrogen. This analysis was carried out at Animal Laboratory in Universiti Malaysia Kelantan, Jeli Campus.

3.2.1 Determination of Dry Matter (DM)

The term "dry matter" refers to the substance that remains after the water has been removed. Forced air oven was used to drying the sample for 24 hours at 105℃ (Ahn, Kil, Kong and Kim, 2014). To calculate DM, formula 3.2.1 was used where weight of sample before drying, W1 must be obtain. The empty container, W0 was weighed first. Later, container that containing sample was weighed too. Total weight of container with samples were subtracted with the weight of empty container and W1 was obtained. Sample undergo drying process. After that, dried sample in container were weighted right after drying process and subtracted to W0 and weight of dried sample, W2 were obtained. W1 or known as moisture weight were divided to the W2 and the answer will be multiplied to 100. Final answer is percentage of DM.

$$\% \, DM = \frac{W1}{W2} \times 100 \tag{3.2.1}$$

When,

% DM = Percentage of Dry Matter

W1 = Weight of moisture Sample

W2= Weight of dry sample

3.2.2 Determination of Moisture Content

Moisture content is the amount of water consist in the feed. The optimum silage moisture depends on its silos storing site. For upright oxygen-limiting solos, the optimal silage moisture varies from 55-60%, 60-65% for upright stave silos, 60-70% for bags, and 65-70% for bunkers (Barnhart, 2009). Silage moisture content could be determining by using moisture analyser (MA).

At first, MA pan was put on the MA and "tare" button was pushed. Later, 1 g of the silage sample was taken and placed on the MA. Set the temperature for 200℃ for 12 minutes and the moisture content reading were gained.

3.2.3 Determination of Ash Content

The overall mineral content of a forage or diet is referred to as ash. It is important to determine ash content in silage because an abnormally high amount of it could be results from contamination the silage to the soils.

Ash content was determined using stated method by Shareef (2015). An empty pot was weighed and recorded as P1. Then, 1.0 g of the sample was putted in the pot and the pot was weighed and recorded as P2. The pot contained 1.0 g sample was put in the oven for 4 hours at 550℃ of temperature. Then, the pot was removed from the oven and left it to cool before putting it in a desiccator for twenty-minutes. The post-heated pot weight was recorded as P3. By using recorded data, ash content from the sample were calculate according with the formula 3.2.3.

$$\% Ash = \frac{P3 - P1}{P2} \times 100$$

(3.2.3)

When,

% Ash = Percentage of Ash Content

P1 = Weight of empty pot

P2= Weight of pot contained 1.0 g of sample

P23= Weight of pot after heating

3.2.4 Determination of Protein (Crude) & Nitrogen

Crude protein is referred to the protein content in an animal feed. The nitrogen content of food proteins affects crude protein. Crude protein was calculated by analysing nitrogen via Kjeldahl method and it was described as 6.25 × nitrogen (N) (Kyawt, 2020). Kjeldahl method consist of 3 steps in determining the crude protein for the sample which are digestion, distillation and titration.

In digesting the sample, it was heat for one hour at 400℃. After that, 1.0 g of the sample need was weighed using analytical balance. Caution step were follows to reduce error when measure the weight. Breakdown of sample in digestion step was used a catalyst to fasten the rate of digestion. As for kjeldahl method, a piece of kjeldahl tablet was acted as catalyst was putted in digestion tube together with 10 ml of distilled water and 12 ml of sulphuric acid.

Later, the Gerhardt Kjeldatherm was activated and heat it up to 400℃ for pre-heating step. Until fully turning the H_2SO_4 solution, vaporization sometimes stopped by attaching it tightly between the digestion tube and fume manifold. The digestion block was reset back for half an hour around 40℃-250℃ and reset it back to 400℃. After one hour, digestion tube was removed into rack holder to undergo cooling process.

Distillation steps was redone for three time for the purpose of cleaning. Afterwards, 40% of Sodium Hydroxide (NaOH) was putted into Gerhardt Vapodest distillation unit. Then, 50 ml of 45% NaOH was added to the digestion sample. 30 ml of the digestion sample was taking out and put into a receiver flask. A 250 ml of Erlenmeyer titration flask was putted on the table with receiving flask. Receiving flask was filled with boric acid (H_3BO_3). Following that, the samples was left for five minutes to prepare it for the distillation parts. Titration was started by titrating H_3BO_3 with standard 0.1 M of HCl and the pink colorization occur was observed. Value amount of HCL used was recorded and the percentage of Nitrogen was calculated using formula 3.2.4.

$$\% N = (T - B) \times N \times 14.007 \times 100 \times Weight\ of\ sample\ (mg)$$

$$\% CP = \%N \times 6.25$$

(3.2.4)

When,

% N = Percentage of nitrogen in sample

T =Volume HCl used for feed sample

B = Volume used for blank sample

N = Normality of titrant

% CP = Percentage of Crude Protein

3.2.5 Determination of Fat (Crude)

Crude fat refers to the crude mixture of fat-soluble material contained in a sample. The typical measure of fat in food products is crude fat, also known as ether extract or free lipid content. Other examples of crude fat are vitamins A, D, E, K, free fatty acids, resins and volatile oils. Determination of crude fat was done through 3 primary components using Soxhlet apparatus. The components are extractor to holds samples, a condenser for cooling and condensing the water vapour, and a round-bottom flask.

First, aluminium cups were heated for 30 minutes at 103℃ then left to cool it for 20 minutes in desiccators and their weight were measured and roundoff the value in for decimal places. Around 1g of sample was weight and putted into extraction thimble before putting the thimble into Soxhlet apparatus. De-fatted cotton was placed at top of the sample and moved the thimbles to its support. The thimbles were insert and make sure the magnet was connected to every thimble in the extraction unit. All the aluminium cups were filled with 80ml of petroleum ether each. The extraction with cup holder was used to insert by the aluminium cups. Press the “Mains” button and wait for the analysis happen. After that, the cups were removed and heated in furnace for 30 minutes at 103℃. Finally, the cups were left to cool for 20 minutes in the desiccators.

Their weight was recorded and the total of crude fat sample was calculated using the formula 3.2.5.

$$\% \, Crude \, Fat = \frac{W3 - W2}{W1} \times 100 \tag{3.2.5}$$

When,

W1= Weight of sample

W2= Initial weight of aluminium cup

W3= Final weight of aluminium cup

3.2.6 Determination of Fibre (Crude)

Fibre is a content in fruits and vegetables that indigestible and or cannot be absorbed by the human at gastrointestinal tract. Examples of fibres are lignin, cellulose, hemicellulose and pectin. Crude fibre in feed was determined by using principle of double process of boiling. Its purpose is to stimulate the conditions similar to digestive tracts pH value, acidic and alkaline in the stomach and small intestine.

The sample moisture content was determined first by heating at 150℃ of temperature in the oven. Then, the sample was be left to cool in the desiccator. The weight of the sample was measured approximately 1.0 g and recorded as W1. After preheating, a 150 ml notch was added with 1.25% of sulfuric acid by using hot plate to reduce boiling time. Antifoam agent, n-octanol was added for 3 to 4 drops and start boiling for 30 minutes. After that, sulfuric acid was dry up by using vacuum. The crucible was wash for three times with 30ml of hot deionized water and filled up the crucible for each time of washing. Air was compressed by stirring the content of crucible. After finishing the third wash, 150 ml of preheated potassium hydroxide (KOH) 1.25% was added along with antifoam for 3 to 4 drops. The crucible was boiled second time for 30 minutes. The crucible was filtered and wash it as the previous ways. At the final wash, cold deionized was used to cool the crucibles content then wash it three times with 25ml of acetone and stirred it every time. Then, the crucibles were removed and dried it on the oven for one hour at 105℃. The dry weight after heating was measured and recorded as W2.

$$\% \, Crude \, Fibre = \frac{W2 - W1}{W1} \times 100 \tag{3.2.6}$$

When,

W1= Initial weight

W2= Weight of crude fibre with ash

3.3 Determination of Reducing Sugar

The sugar content in silage is important for silage. If the sugar content exceeds 3%, it is suitable for silage. Reducing sugar in silage was determined by using Folin-Wu method. This method required alkaline copper reagent and phosphomolybdic acid that react together and the absorbance value of samples were recorded. By this method, concentration of sugar in the sample could be measured by reading the absorbance value based on the standard curve developed using concentration of glucose standard solution (Sudarmaji, Haryono, & Suhardi, 1984).

This method was carried out at Biology Laboratory in Universiti Malaysia Kelantan. At first, glucose standard solution was pipetted for 0.0, 0.25, 0.5, 0.75, 1.0 and 1.5 ml by using micropipette and transferred to different test tube. Each test tube was added with distilled water until become 2ml. Later, all test tube were added with 2 ml alkaline copper reagent. All test tube were kept in the boiling water bath for 10 minutes. After that, test tubes were taken out from the water bath and left to be cool until room temperature. Then, each test tube was added with 2 ml of phosphomolybdic acid and those test tubes were left for 1-2 minutes. All test tubes were added with distilled water until 10 ml and make sure to be mixed well. Each solution from those test tubes was pipetted and transferred to cuvette. Their absorbance was read by using UV spectrophotometer and a graph of concentration against optical density was sketch. All those steps were repeated for the sample solution.

3.4 Data Analysis

Independent samples T-Test was carried out using IBM® SPSS® Statistics (SPSS) version 26.0 software to compare between PM and PEM after the fermentation process. Independent samples test was used to test the significant difference between those two treatments (PM & PEM). The comparison was used to identify if ensilation of pineapple leaves could help in improving the nutrient availability for cattle feed.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Completed Ensiling Process

Ensiling process was supposed to be completed after fermentation occurred when stored in airtight condition for minimums of 21 days. Fermentation process in ensiling can continues from 7 days and can last until 45 days (Pahlow, Muck, Driehuis, Elferink & Spoelstra, 2003). In this study, PM and PEM were stored for one month or more than 4 weeks. This is to make sure that fermentation process has been completed for both silages. It is also to make sure proximate analysis were carried out at the optimize silage condition and nutrient where it is suitable for cattle consumption which are silage at the age of 3 to 4 weeks (Amaral, 2013).

Physical qualities of both PM and PEM silage after completed fermentation process were recorded as shown in the Table 4.1. When the silage containers were opened, both PM and PEM produced such an unpleasant odour like vinegar but not too sharp. According to Kung, Shaver, Grant, and Schmidt (2018), silages that has undergo good fermentation process was not supposed to have a strong smell. That is because the lactic acid which are responsible for the fermenting as the prime component was almost did not smelly. However, most silages produced regularly to have a vinegar like odour which is come from acetic acid. Acetic acid comes after lactic acid in term of concentration and it also appear to be labile.

Usually, the appearance of silages with vinegar smell and high acetic acid are in yellow colour (Kung *et al.*, 2018). The appearance of PM and PEM silage showed as stated. PM has a brighter yellow colour compared to PEM. The yellow colour also corresponds to the compaction of the silage in container which lead to high moisture content to the silage. From the colour of both silages, we may conclude that PM has higher moisture content than PEM.

There also presence of the mould for both silages. The mould appeared on the top of silage but not covering the silages. They were patched between the inner surface of the container and silage. The presence of mould and maybe along with mouldy smell may because of the anaerobic unstable developed silage (Kung *et al.*, 2018). The presence of mould on PM silage is higher than on PEM. Thus, addition of EM may prevent the excessive appearance of mould. Before undergoing proximate analysis, all the mould was removed. Contamination between unmould silage with mould silage can become mycotoxins which can cause critical problems on health, fertility and production of cattle (Korosteleva, Smith, & Boermans, 2009). Mould silage also not suitable for proximate analysis because of its low nutrient value due to the yeast existence earlier (Santos, Lock, Mechor & Kung, 2015).

Figure 4.1.1 : Completed PM silage

Figure 4.1.2 : Completed PEM Silage

Earlier, both PM and PEM was weight approximately 3.25 kg each before the fermentation begin. After one month, when PM and PEM were weighted again, there are about 20% weight different from before for both silages. The shrink is normal to well-developed silage as long in the amount of 10 to 20%. If the loss occurred more than 50%, there are serious problem in the post-harvest or during ensiling process. Borreani, Tabacco, Schmidt, Holmes & Muck (2018) reviewed that high percentage of DM losses occur because of not good enough management during field and pre-ensiling rather than respiration and temperature during ensiling. Based on the PM and PEM loss, we can assume that both silages are well managed because the loss rate still in the acceptable rate.

Table 4.1: Physical qualities of both silages

Silages	Physical Qualities	
	Colour	Smell
PM	Bright Yellow	Vinegar like
PEM	Brownish Yellow	Vinegar like

4.2 Proximate Analysis

The data for every analysis for PM and PEM nutrition composition (dry matter, moisture content, crude fat, crude protein, crude fibre and ash content) were recorded, determined and analysed. Table 4.2 showed the proximate composition of both silages. Results indicated that PM silage is statistically not significant with PEM silage although PM was analysed high for moisture content, ash, and crude fat while PEM silage is high for DM, crude protein and crude fibre. PEM is high in DM content (39.93%) as compared to PM (36.13%). Meanwhile, PM has high moisture content than PEM with 71.15% and 66.4% respectively. PM silage has higher ash content than PEM silage with value of 0.083% and 0.081% respectively. PEM silage (4.12%) is high in crude protein as compared to PM silage (3.0%). However, PM silage's crude fat is higher (3.48%) than PEM silage (2.64%). There are also different of crude fibre content between both silages where PEM (11.9%) is higher than PM (9.3%).

Table 4.2: Nutritive value of both pineapple leaves silage

Silages	Nutritive Value of the pineapple leaves silage					
	DM	**Moisture**	**Ash**	**Crude Protein**	**Crude Fat**	**Crude Fibre**
PM	36.13 ± 1.11^{a}	71.15 ± 2.11^{a}	0.083 ± 0.03^{a}	3.0 ± 0.11^{a}	3.48 ± 1.88^{a}	9.30 ± 0.25^{a}
PEM	39.93 ± 1.81^{a}	66.40 ± 1.70^{a}	0.081 ± 0.01^{a}	4.12 ± 0.68^{a}	2.64 ± 1.34^{a}	11.90 ± 1.03^{a}

Means followed by ± standard error mean
Same letters indicate statistically no significant differences between treatments (P > 0.05)

Suphalucksana, Sangsoponjit and Srikijkasemwat (2017) stated that if the silage produced has higher moisture content while lower in DM during rainy season, it may increase the risk for silage to be spoiled. In other case, low moisture content but high DM in silage may result in difficulty in compacting the silage for storing because of more fibre (Saranya and Jantakan, 1997). In this study, both PM and PEM silage were harvested and prepared during rainy seasons and there also mould were found on the silage as agreed by Suphalucksana *et al.* (2017). By preparing silage during summer season, it may reduce the risk for the silage to be spoiled and the chance for the mould to appear may be reduced too. Unfortunately, there will be a chance for the silage to be difficult for cattle consumption when the fibre content is high. PEM silage has value of DM (39.93%) than PM silage (36.13%). Generally, high percentage of DM in starting material would lead to silage with high protein and energy. Therefore, PEM silage have higher energy and protein content than PM silage. According to Hanafi, Selamat, Husni, and Adzemi (2009), pineapple leave has an accumulated DM of 48.5%. After the fermentation, those amounts were reduced but still in at the ideal level. The uses of EM can increase DM for PEM even there are statistically no significance difference (P > 0.05).

Correspond to the DM value, moisture content will become high if DM is low. When high moisture content forages undergo fermentation, there will be presence of *clostridia* bacteria which will consume carbohydrate, protein and lactic acid from the forages and butyric acid will be produced (Amaral-Phillips, 1996). Higher moisture content of the silage (more than 72%) with low DM (less than 28%) may result in ketosis when fed to the cattle. In this study, PM has lower DM but higher moisture content (71.15%) than PEM (66.4%). PM silage moisture content was still acceptable for the cattle consumption but PEM silage is more than suitable due to the higher DM content. Butyric acid analyses were not carried in this study to know whether its concentration was lower than 0.1% or not. Butyric acid analysis is beneficial to get evidence whether the silage has undergone clostridia fermentation which is harmful for cattle consumption. Besides, clostridial silage can be prevented by avoiding contamination during harvesting, harvest with suitable DM value and seal tightly during storage. Despite all the lacks during handling which may occur due to the of weather condition, the uses of EM showed that it can help to lower the moisture content while increasing the DM value for pineapple silage although there is no significance difference with PM moisture content ($P > 0.05$).

Ash content in other silages is different based on the ingredients used. Corn silage has approximately 5.0% and legumes-grass forages silage with 9.0% (Hoffman, 2005). Based on table 3, PM silage has a slightly higher mean for ash content (0.083%) than PEM silage (0.081%). However, there is no significant different for ash content between both silages ($P > 0.05$). There was a big difference of value compared to the corn silage and legumes-grass silages. For different varieties pineapple leaves that have not undergo fermentation process, the amount almost similar for MD2, Josapine and Moris with 2.35%, 2.11% and 2.08% respectively (Zainuddin *et al.*, 2014). There are still big differences where after fermentation occurred the ash content was reduced drastically. Compared to Suphalucksana *et al.* (2017) finding, the ash content for both PM and PEM in this study has a great different value with the ash content of 100% pineapple waste silage (9.38%). This indicate both PM and PEM silages were lacking in minerals which is not conducive for the dairy cows that require a high amount of ash from forages and grass. Although, EM were added on PEM it not heavily affected the ash content compared to PM. Amount of EM used could also be the reason of low ash content in PEM whereas the amount of EM added (5%) were not optimum for the pineapple leaf silage from having higher ash content.

Pineapple leaf waste silage with the mixed of pineapple leaves, crown and stem had 6.13% crude protein (CP) (Suphalucksana *et al.*, 2017). Meanwhile, PEM and PM has lower crude protein amount with 4.12% and 3.0%. The different between Suphalucksana *et al.* (2017) findings with PEM and PM may be different due to the different part that were taken from pineapple plant which in the sample only the leaf was used. Comparing PEM and PM crude protein, the addition of EM on pineapple leaves silage increase the CP even the different is statistically not significant ($P > 0.05$). Crude protein of MD2 pineapple leaves without ensiling process is 5.82% (Zainuddin *et al.*, 2014). Based on the finding, after fermentation occurred in PM and PEM silages the amount of crude protein in both silages were reduced. However, addition of EM in PEM is useful to prevent major loss of crude protein as compared in PM silage. The result suggested that the addition of EM can provide more CP in making silage derived from pineapple leaves.

Crude fat (CF) in other silages is different based on the crops such as maize, wheat and barley silages with 8%, 12% and 19% respectively (Udén, 2018). Both PM and PEM silage crude fat are lower than those three popular silages. A dairy cow weighed 635 kg need at least 3.6% fat per day (Jenkins, 1997). PM silages almost reach the required amount with 3.48% but PEM silages far from enough with 2.64%. Compared to the crude fat in pineapple leaf without ensilation process (3.05%) from Zainuddin *et al.*, in 2014, there are slight increase of amount for pineapple leaves silage with 5% molasses (PM). On the contrary, crude fat in PEM silages was lower than Zainuddin *et al.* (2014) findings. Improvement trough the composition might need to be work along with strict compliance to the precautions during preparing silages.

According to Zainuddin *et al.* (2014), crude fibre in different variety of pineapple leaves without ensiling is 30.93% for MD2, 31.03% for Moris and 31.04% for Josapine. In this study, after fermentation, total crude fibre for PM silage is 9.30% while PEM is 11.90%. There are crucial differences of crude fibre compared to pineapple leaves without ensiling where the amount of crude fibre of PM and PEM are lower. This reduction is good for the cattle consumption. Crude fibre in a feed for dairy cow should not exceed 16% (Nordfeldt, Iwanaga, Morita, Henke, & Tom, 1950). If the amount was exceeded, milk produced can drop. The study showed that the fermentation process has reduced the lignocellulose bioavailability on pineapple leaf silages and made it suitable for cattle consumption.

4.3 Reducing Sugar

A standard curve graph was constructed as Figure 4.3 to determine the concentration of PM and PEM from the absorbance reading of both silages. From those graphs, concentration of reducing sugars was determined for both silages using standard curve linear ($y = 0.1343x - 0.0289$) and the mean as stated in the Table 4.3. PM silage has lower concentration (0.48 mg/ml) than PEM (0.64 mg/ml) but the value is not statistically significant between both silage (P > 0.05).

Figure 4.3: Standard Curve of Glucose standard solution

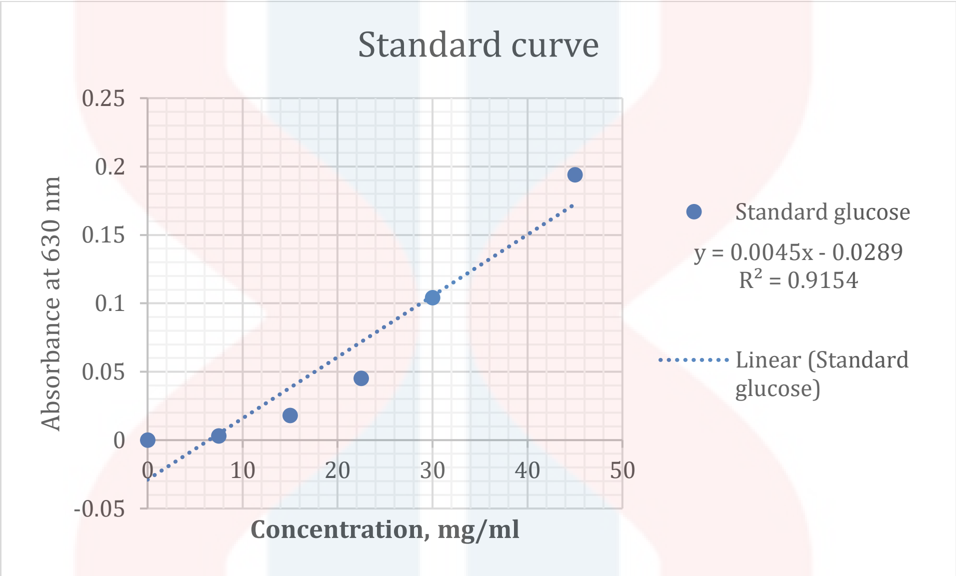

Table 4.3: Concentration of both pineapple leaves silage based on standard curve

Silages	Concentration, mg/ml
PM	0.48 ± 0.09[a]
PEM	0.64 ± 0.19[a]

Means followed by ± standard error mean
Same letters indicate no significant differences between treatments (P > 0.05)

During the first 5 days of ensiling total sugars is high while reducing sugar also high and then gradually decrease until 10 days of ensiling when no significance change for the total sugar and reducing sugar (Bender, Bosshardt, & Garrett, 1941). No significance change to the total sugar and reducing sugar is because acetic content was increased along with pH for the fermentation to occurred. In this study, reducing sugar as concentration for PM and PEM are 0.48 mg/ml and 0.64 mg/ml as in Table 4.3. There was low concentration of reducing sugar when the reading was taken at both silages were at the age of more than a month. Lignocellulose consists of cellulose which is a glucose polymer (Sanderson, 2011). High crude fibre before ensiling in pineapple leaf mean there also high glucose. After becoming silages, the lignocellulose was broken down through fermentation process and result in reducing sugar. From the mean of both silages we can safely assume that fermentation occurred for both silage as the value of reducing sugar is low when the lignocellulose has been broken down.

Sugar content in silage is necessary in preserving the silage for up to 6 months. It is advisable for the silage to have 3% or higher sugar content. However, both PM and PEM silage were lower sugar content from the recommendation. The main cause was delayed in silage preparation (wilting, cutting & storing) the leaves after harvest. Silage preparation could not be carried out at the same day of harvest because of constrains in the laboratory equipment only can be used during office hours. Thus the leaves were left more than one day until wilting were carried out. This resulted with sugar content were declined by the uses from the living leaves and undesirable bacteria.

The addition of EM on PEM showed that sugar content in the silage is higher than PM. It indicates that lactic acid bacteria in EM prevent further sugar breakdown during fermentation while keeping the pH value from dropping further. This study finding agreed with Yitbarek and Tamir (2014) where high sugar concentration along with high protein content when EM was added. PEM silage should have higher pH value and less acidic than PM silage which is good for the cattle consumption.

CHAPTER 5

CONCLUSION

On the whole of study, the objective was achieved whereas pineapple leaves that undergo fermentation process as manipulation towards cattle consumption generally suitable for cattle consumption. Crude fibre content was reduced through fermentation of pineapple leaf. Thus, the high fibrous content in the leaves for cattle digestibility was overcomed in this study. This render the pineapple leaves suitable for cattle to digest as an alternative feed. Next objective also was achieved where the composition of dry matter, moisture content, ash content, crude protein, crude fat and crude fibre of PM and PEM were determined and compared. Although all the elements analysed were not statistically significant between addition of 5% EM to the pineapple leaves silage with 5% of molasses but there are still improvements for some nutrients of the silages. PEM silages has high nutrient composition of DM, crude protein and crude fibre than PM silage that high nutrient composition for ash content, crude fat and moisture content. Both PM and PEM silages are still considered low nutrition value than other popular silages such as corn silage, Napier grass silage and wheat silage. However, it is still can be considered as an alternative feed for the cattle.

RECOMMENDATION

In term of handling pre-harvest and pre-ensile must be carry out carefully to avoid contamination on the pineapple leaves that can affect the quality of silages and nutrient composition. In a worst scenario, the silage must be discarded as it can be harmful for the cattle consumption. Harvesting season also affect the composition and quality of the silage. Summer season may produce different nutrient composition and quality to the silage produced. In term of analysis, it is better when proximate analysis was carried out before and after fermentation occurred to clearly identify the different nutrient compositions. It is also advisable if the physical quality of the silage were carried out to identify different treatment characteristic and physical qualities. Butyric acid concentration analysis also needs to be carried out to identify whether the silage undergo clostridial fermentation. Lastly, study of different percentage of EM additive on pineapple leaves silage also can be done in the future to know the optimize percentage of EM that can produce the highest quality of silage.

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APPENDIX A

Table A.1.i : Independent Samples Statistics for Ash Content from SPSS

	Ash	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	.0825	.05052	.02917
	2.00	3	.0812	.01622	.00936

Table A.1.ii : Independent Samples Test for Ash Content from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	1.845	.246	.040	4
	Equal variances not assumed			.040	2.408

Table A.2.i : Independent Samples Statistics for DM from SPSS

	DM	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	36.1326	1.91864	1.10772
	2.00	3	39.9317	3.13454	1.80973

Table A.2.ii : Independent Samples Test for DM from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	1.423	.299	-1.790	4
	Equal variances not assumed			-1.790	3.314

Table A.3.i : Independent Samples Statistics for moisture content from SPSS

	Moisture	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	71.1467	3.65749	2.11165
	2.00	3	66.4033	2.94629	1.70104

Table A.3.ii : Independent Samples Test for moisture content from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	.371	.575	1.749	4
	Equal variances not assumed			1.749	3.827

Table A.4.i : Independent Samples Statistics for CP from SPSS

	Protein	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	2.9865	.18855	.10886
	2.00	3	4.1194	1.18525	.68430

Table A.4.ii : Independent Samples Test for CP from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	9.000	.040	-1.635	4
	Equal variances not assumed			-1.635	2.101

Table A.5.i : Independent Samples Statistics for CF from SPSS

	Fat	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	3.4816	3.25277	1.87799
	2.00	3	2.6355	2.32141	1.34027

Table A.5.ii : Independent Samples Test for CF from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	.126	.741	.367	4
	Equal variances not assumed			.367	3.618

Table A.6.i : Independent Samples Statistics for Crude Fibre from SPSS

	Fibre	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	3	9.2995	.43384	.25048
	2.00	3	11.8965	1.78612	1.03122

Table A.6.ii : Independent Samples Test for Crude Fibre from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	5.854	.073	-2.447	4
	Equal variances not assumed			-2.447	2.235

Table A.7.i : Independent Samples Statistics for Reducing sugar from SPSS

	Conc	N	Mean	Std. Deviation	Std. Error Mean
PL	1.00	6	.4795	.22067	.09009
	2.00	6	.6409	.47408	.19354

Table A.7.ii : Independent Samples Test for Reducing sugar from SPSS

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PL	Equal variances assumed	1.385	.266	-.756	10
	Equal variances not assumed			-.756	7.069

APPENDIX B

Figure B.1 : Chopper Machine

Figure B.2 : Crude fibre analysis was undergone

Figure B.3 : Sample preparation for fat analysis

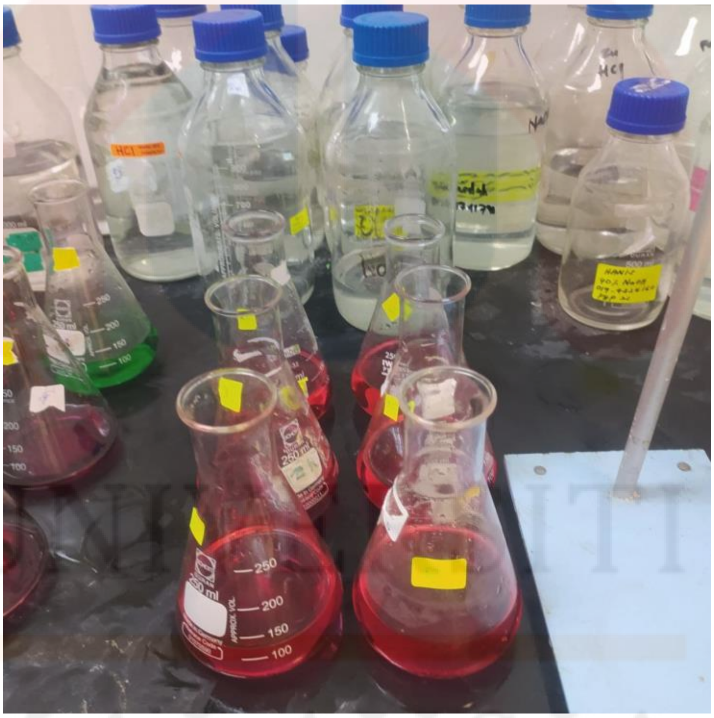

Figure B.4 : Reciver chemical for titration in Kjedahl Method

APPENDIX C

Manipulation Of The Pineapple Leaves For The Development Of Silage Suitable For The Cattle Feed

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