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The Roles of Ammoniation, *Direct Fed Microbials* (DFM) and Cobalt (Co) in the Creation of Complete Cattle Feed Based from Oil Palm Trunk

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ABSTRACT

The continuous increase in population and life style, will result in increased demand for animal protein. Animal protein from beef will be among the animal protein sources of paramount importance due to the large size of beef cattle and the volume of meat it can produce at a time. In Indonesia, comparison between national beef production and consumption reveals a deficit and about 51% of beef are imported from other countries. It is estimated that conditions of this nature will continue to increase every year. One of the problems in the development of ruminants is forage availability since forage serves as the main feed for ruminants. Forage availability in Indonesia is affected by differences in geographical conditions in each region and changes in productive forage land for animal feeds into housing, industry and oil plantations. Indonesia has a high oil palm plantation of about 11.9 million Ha in 2016 and is expected to increase to 14.03 million Ha in 2018. Oil palm plantations need to be rejuvenated (replanting) every 25-30 years. Replanting produces wastes especially in the form of oil palm trunks. The pith of the oil palm trunk can be used as animal feed because it has a high fiber content (44.43%) and can serve as a source of energy for ruminants. However, the oil palm trunk has a limiting factor as the high lignin content (15.41%) causes low digestibility. Therefore, it is important to develop technologies that can reduce the lignin content and increase the digestibility. The purpose of this paper is to review the potentials of oil palm plantation wastes (trunks) as high quality beef cattle feed using ammoniation technology, *Direct Fed Microbials* (DFM) and addition of the mineral, cobalt (Co) to increase meat production in beef cattle.

Keywords : Oil palm trunk, Cattle, Ammoniation, DFM, Cobalt

ABSTRAK

Sejalan dengan bertambahnya jumlah penduduk, keperluan hidup akan meningkat pula termasuk keperluan akan protein haiwan daripada daging lembu. Perbandingan antara pengambilan dan pengeluaran daging lembu di Indonesia menunjukkan defisit sehingga harus diimpor sekitar 51% dari negara lain. Diperkirakan keadaan seperti ini akan terus meningkat setiap tahunnya. Salah satu permasalahan dalam pengembangan haiwan ternakan adalah ketersediaan makanan. Terdapatnya perbezaan persekitaran disetiap daerah di Indonesia dan berlakunya perubahan kawasan penghasilan makanan haiwan ternakan kepada kawasan perumahan, industri dan perkebunan telah menurunkan peluang haiwan ternakan mendapatkan makanan semulajadinya. Indonesia memiliki area perkebunan kelapa sawit yang luas iaitu sekitar 11.9 juta Ha pada tahun 2016 dan diperkirakan bertambah kepada 14,03 juta Ha pada tahun 2018. Kelapa sawit perlu ditanam semula (*replanting*) setiap 25-30 tahun. Penebangan pokok untuk penanaman semula ini akan menghasilkan sisa terutama dalam bentuk batangnya. Bagian empulur dari batang kelapa sawit dapat dimanfaatkan menjadi makanan haiwan ternak karena memiliki kandungan serat yang cukup tinggi (44,43%) sebagai sumber tenaga bagi ternak. Akan tetapi batang kelapa sawit juga memiliki faktor pembatas yaitu kandungan lignin yang cukup tinggi (15,41%) yang dapat menyebabkan penghadaman menjadi rendah. Oleh itu pengetahuan berkaitan teknologi untuk menurunkan kandungan lignin serta meningkatkan nilai penghadaman adalah penting untuk dikembangkan. Tujuan dari penulisan ini adalah untuk menerokai potensi sisa perkebunan kelapa sawit sebagai bahan makanan ternak lembu pedaging yang bermutu tinggi dengan menggunakan teknologi amoniasi dan *Direct Fed Microbials* (DFM) serta penambahan mineral Cobalt (Co) untuk meningkatkan penghasilan daging pada haiwan ternakan lembu pedaging.

Kata kunci : Batang kelapa sawit, sapi potong, amoniasi, DFM, Cobalt

INTRODUCTION

Indonesia is the country with the fourth largest human population in the world. The country has a total population of 252.2 million in 2014 and it is projected to increase to 305.7 million by 2035 (Badan Pusat Statistik, 2014). The increase in population will be accompanied by increase life style and demand for goods and services. These goods and services will include the livestock industry needed to provide meat, eggs and milk for the supply of animal protein. An important source of animal protein is meat, derived from beef cattle. According to Dirjen

Peternakan dan Kesehatan Hewan (2016), national beef consumption in 2016 was 0.417 kg/capita/year, equivalent to 1.05 million tons, while national meat production in 2016 was 518,500 tons. This shows a beef supply deficit and importation of cattle from other countries to fulfill the demand. In 2016, Indonesia imported 493,726 tons of cattle (Dirjen Peternakan dan Kesehatan Hewan, 2016). Importation of cattle is expected to increase every year along with the increasing population and the needs for animal protein. Therefore, the production of beef cattle must be increased to be able to supply the domestic needs. One of the problems in the development of ruminants production is the availability of forage (the main feed for ruminants). The availability of forages can be influenced by differences in geographical conditions in each region of Indonesia and the conversion productive forage lands for animal feeds to housing, industries and plantations.

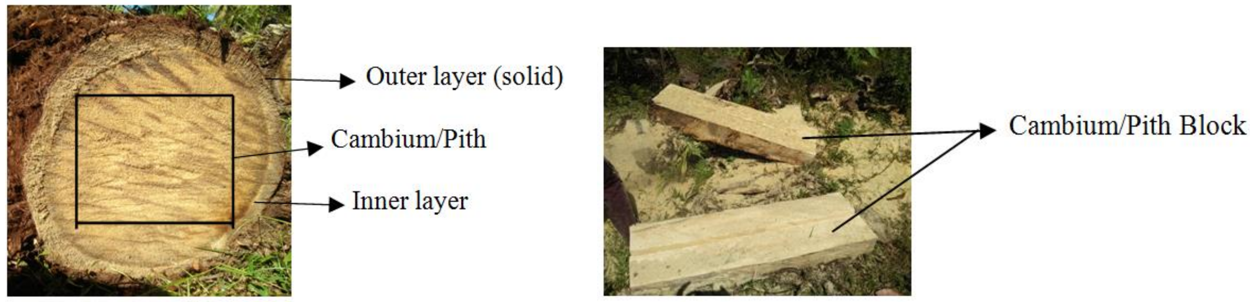
Oil palm plants were brought initially by the Dutch to Indonesia in 1848 and were planted in the Bogor Botanical Gardens. Because oil palm plants thrived well in Indonesia, in 1910, commercial cultivation began and expanded to other parts of Indonesia. In the 1980's the area under oil palm cultivation/plantations reached 200,000 hectares and has continued to increase until now. According to Dirjen Perkebunan (2016), the potential of oil palm plantations in Indonesia is very high with a plantation area of 11.9 million Ha in 2016 and it is expected to increase to 14.03 million Ha in 2018. The largest distribution of plantations in Indonesia is in the provinces of Riau, North Sumatra, Central Kalimantan, South Sumatra, and West Kalimantan (Dirjen Perkebunan, 2016). As the area under oil palm plantations continues to increase in Indonesia over years, replanting or rejuvenation of oil palm plantations will also increase. The consequences of this replanting will lead to the production of large quantities of oil palm wastes, especially oil palm trunk that can be used as animal feed.

Oil palm replantation is an activity to replace the old oil palm plants that are no longer economical compared to new oil palm plants or after 25-30 years old of being planted. The oil palm trunk from replantation that can be used as animal feed is the pith or the inside of the trunk which has been separated from the outer layer. Guritno and Darnoko (2003) stated that the average area of replantation during the period of 2001 - 2005 was 32,155 ha/year. Solid wastes in the form of oil palm trunk was generated at 2.26 million tons per year, while in the period of 2006 - 2010 there was an increase in the area of replanting oil palm plantations, which averaged 89,965 hectares annually. During this period of replanting oil palm plantations solid waste production reached 6.3 million tons per year.

The large quantity of wastes produced from oil palm plantations serve as potential for cattle feed. The pith of the oil palm in particular is of much importance for the production of cattle feed. The percentage of pitch in an oil palm truck is 75%. Thus 4.72 million tons of oil palm pith was available as animal feed. In terms of nutrition, oil palm trunks contain high crude fiber which serves as an energy source for ruminants. However, oil palm trunk as ruminant feed has limitations due to the high lignin content. Therefore, the development of technologies to reduce lignin content and to increase digestibility is important. The purpose of this paper was to review the potential of oil palm plantation wastes as high quality beef cattle feed using ammoniation technology, *Direct Fed Microbial*s (DFM) as well as the addition of the mineral, cobalt (Co) to increase meat production in beef cattle.

Morphology and chemical composition of oil palm trunk pith and its limitations

The morphology of the oil palm trunk consists of the hard outer part, the softer inner trowels and the pith. The trunk of oil palm has more percentage of pith than other parts. Cross section of the oil palm trunk and its pith can be seen in Figure 1.



a. Oil palm trunk morphology   b. Oil palm trunk pith

Figure 1. Cross section of oil palm trunk (a = morphology of pith; b = block of pith after separated from outer layer)

Oil palm pith has a chemical composition containing 49.54% dry matter, 87.56% organic matter, 44.43% crude fiber, 3.64% crude protein, 3.32% crude fat, 55.33% cellulose, 20.35% hemicellulose and 15.41% lignin (Azhary, 2018). The highly fiber fractions contained in oil palm pith is a very good source of energy for ruminants. However, the highly lignin content serves as a limiting factor since lignin can form lignocellulose and lignohemiselulose bonds making it difficult to be degraded by rumen microbes. Lignin is composed of three phenylpropanoid compounds, namely komaril alcohol, alcohol coniferil, and sinapil alcohol, which are arranged randomly to form amorphous or irregular lignin polymers (Higuchi, 1980). Lignin is one of the phenylpropanoid polymers which are difficult to be remodeled (recalcitrant). This is due to its heterogeneous structure and complexity.

Existing technologies for using oil palm trunks

Utilization of oil palm trunks can be done using several technologies with the aim of reducing lignin content and improving quality in terms of nutritional content and digestibility. Some of the technologies commonly used include physical (chopped and evaporated), chemical (alkaline/NaOH and acid/ammonia treatment), biological (fermented and enzymatic) and their combinations. The use of oil palm as ruminant feed is very promising if it is first processed. Ammoniation is an inexpensive, easy and practical technology used in forage processing and can improve the palatability of livestock.

Ammoniated technology

Ammoniation is a treatment of waste feed ingredients with urea $CO(NH_2)_2$. There are three sources of ammonia that can be used in the ammoniated process: NH_3 in the form of liquid gas, NH_4OH in the form of a solution, and urea in solid form. The advantages of using urea for ammoniation is that, it is easy to obtain, the price is relatively cheap, it is easy to handle, it is non-toxic and has a high nitrogen content. This is supported by Siregar (1995), who stated that, urea with the molecular formula $CO(NH_2)_2$ is widely used in ruminant rations because it is easy to obtain, has low price and little poisoning caused by biuret. Physically, urea is solid white and hygroscopic. Ammoniation from urea can cause changes in the composition and structure of cell walls which play a role in breaking the bonds between lignin and cellulose/hemicelluloses (Komar, 1984). The chemical reaction that occurs (by cutting the hydrogen bridge) develops the tissue and increases the flexibility of the cell wall to facilitate penetration by cellulase produced by microorganisms.

Ammoniation has been shown to have good effects on feed. Furthermore, ammoniation increases feed digestibility. After decomposing into NH_3 and CO_2, NH_3 and water molecules will undergo hydrolysis to NH_4^+ and OH. NH_3 has pKa of 9.26, meaning that in a neutral atmosphere (pH=7) there will be more NH^+. Thus, ammoniation will be similar to alkaline treatment. The OH group can break the hydrogen bond between carbon number 2 and one glucose molecule with oxygen. Carbon number 6 is another glucose molecule found in cellulose, lignocellulose and lignohemicellulose bonds. It is known that these last two bonds are alkaline labile which can be broken by alkaline treatment. Thus, the feed will expand making it easily digestible by the rumen microbes. Further expansion of feed will dissolve the lignin deposits found in the walls and space between cells. It means that ammoniation also decreases the level of chemical composition that are difficult even not digested by livestock which results in an increase in the digestibility of feed far more.

The positive effect of ammonia treatment on straw was first reported by Nikolaeva (1941) and more research was conducted by Chomyszyn *et al.* (1961). A shortage of fodder in Scandanavia because of drought in the seasons of 1975 and 1976 accelerated interest in the ammoniation of straw (Sundsteji *et al.*, 1978). According to Parulian (2009), urea is used at 1% in a ration on dry matter basis, not more than 3% in concentrate mixture or not more than 1/3 of the protein requirement. Improved performance of animals receiving ammoniated roughage is dependent upon the concentrate level of the diets fed (Horton, 1979; Garrett, *et al.,* 1979). Furthermore, Warly (1994) stated that ammoniation treatment can increase the digestibility of rice straw both *in vivo*, *in vitro* and *in sacco*, and increase feed consumption and body weight gain of sheep. According to Seed *et al* (1985) stated that ammoniated maize residue can be included in fattening diets for lambs with beneficial effect provided that not more than 60% concentrate is included and The higher dressing percentage indicates a higher fat content in carcasses of lambs fed ammoniated diets (Berg and Butterfield, 1976).

THE ROLE OF DIRECT FED MICROBIALS (DFM)

Manipulation of the rumen ecosystem can be done through feed processing (to increase energy availability and increase protein) and through supplementary feeding. These will stimulate microbial growth and activity in the rumen and increase digestibility and efficient feed usage. The bioprocess activity in the rumen can be manipulated as long as the nutrient requirements of the rumen microbes are supplied, whereas certain nutrient deficiencies needed by rumen microbes will reduce biomass and will result in decreased digestibility of feed, especially fibrous feed (Preston and Leng, 1987). Bioprocess manipulation can be done by feeding supplements that can stimulate microbial growth and activities to increase the digestibility of feed in the rumen. Some of these feed supplements are buffers, defaunation agents, amino acids and precursors, probiotics, minerals and enzymes.

In recent times the use of probiotics, also known as *Direct Fed Microbials* (DFM) for animal production has become intensive among farmers. Haddadin *et al.* (1996) stated that probiotics are organisms and their substances can support the balance of micro-flora in the digestive tract. The use of probiotics is an alternative for controlling rumen fermentation and a more efficient way of feed and nutrient usage. Some strains of microorganisms have been used as probiotics, these strains include yeast and fungi. The use of probiotics can increase the population and activity of rumen microbes to increase feed digestibility. *Saccharomyces cerevisiae* has been widely used and it is known to increase livestock productivity. The use of probiotics in feeds is intended to ensure a balance of useful microorganisms in the degradation process of the components of nutrients in the rumen (William and Newbold, 1990). Enzymatic activity on the degradation of fiber components can be increased if the production of fiber-breaking enzymes can be increased (Gong and Tsao, 1979), this can be achieved by microbial supplementation or probiotics.

The use of these microbes has the advantages of increasing fermentation efficiency in the rumen, increasing forage digestibility and increasing the flow rate of microbial proteins from the rumen (Wallace and Newbold, 1992). Probiotics help to increase body weight gain by increasing consumption of dry matter and protein (Ngadiyono and Baliarti, 2001), providing better rumen conditions (Prihardono, 2001), increasing dry matter digestibility (Apriyadi, 1999) and higher nitrogen retention (Han et al, 2004). The addition of probiotics in rations can stimulate microbial growth in the rumen and increase feed digestibility in ruminants (Zain *et al.*, 2011). The microorganisms commonly used as DFM in ruminants are grouped into three, these are bacteria, yeast and fungi.

DFM of bacteria species

Bacterial species commonly used as DFM in ruminant animals are *Bacillus*, *Lactobacillus* and *Pseudomonas* (Llyod-Evans, 1989; Chen and Chiou, 2004). *Bacillus* is one of the bacterium that can produce various types of enzymes that are able to breakdown chemical compositions such as carbohydrates, fats and proteins into simple compounds that are more easily digestible by livestock (Buckle *et al.*, 1987). *Bacillus* bacteria are widely used as probiotics because of their ability to produce antimicrobial compounds that can inhibit the development of other harmful microorganisms. All types of *Bacillus* groups will produce these antimicrobial compounds under certain conditions if there is an inducer compound that is able to induce the biosynthesis of these antimicrobial compounds in their cells.

Bacillus amyloliquefaciens as a probiotic has been known to produce a variety of important commercial enzymes and metabolism in the last decade (Zhang, 2011). Wizna *et al*., (2007) isolated a cellulolytic bacterium (*Bacillus amyloliquefaciens*) from Lunang Peat forest litter in Pesisir Selatan Regency, West Sumatra. *B. amilolyquifaciens* products as probiotics have been used in the poultry industry to increase their performance. However, the utilization of *B. amylolyquifaciens* has never been tested on ruminants. Therefore, research is needed regarding the use of *B. amyloliquifaciens* in ruminant animals given the ability of these bacteria to produce several enzymes that have the potential to help digest the fiber ruminants take in.

Several studies have reported on the effect of bacteria as DFM for feed supplementation. Qiao et al, (2010) found that *Bacillus subtilis* as a supplement did not produce any effect on rumen fluid characteristics and nutrient digestibility by *in vivo* trial. However, supplementation of *B. licheniformis* increased the average dry matter digestibility, NDF and ADF (Beecher *et al*., 2009). Also, supplementation of *Bacillus licheniformis* in cattle feed increased the average daily body weight gain during 0-8 weeks of feeding trial (Fu *et al*., 2012). Some studies that used bacteria as DFM species in improving the digestibility of feed ingredients are presented in Table 1.

Table 1. The use of DFM of bacteria in increasing digestibility

No	DFM of Bacteria	Species	Result	Reference
1	Suplementation with *L. acidophilus* at 5×10^7 cfu/ml.	Calf	Initial body weight was maintained and control of body weight loss until 2 weeks of age occurred.	Cruywagen *et al*. (1996)
2	Suplementation with *L. acidophilus, L. plantarum*, and *L. acidophilus* strain 27SC at 1.85×10^7 cfu/ml.	Calf	Reduced risk of diarrhea in the first week in calves and *Lactobacilli* increased in calf faeces given liquid feed plus *L. acidophilus* 27SC in it.	Abu-Tarboush *et al*. (1996)
3	Suplementation with *Propionibacterium* strain P169 at 6×10^{11} cfu/day	Cattle rumen fluid (*in vitro*)	Lower concentration of acetate, higher propionate concentration and energy efficiency.	Weiss *et al*. (2008)
4	Suplementation with *Propionibacterium* strain P169 at 6×10^{11} cfu/day	Cattle rumen fluid (*in vitro*)	Increased propionate proportion but did not affect rumen digestion, microbial protein synthesis or feed flow rate.	Lehloenya *et al*. (2008)

DFM of yeast species

The species of yeast commonly used in ruminants is *Saccharomyses cerevisiae* (Shin *et al*., 1989). The use of *S. cerevisiae* as a supplement to live microbes in the rumen will affect the host by improving the balance of rumen microorganisms. *S. cerevisiae* is able to compete with starch bacteria which leads to the prevention of lactate accumulation in the rumen (Lynch and Martin, 2002). In addition, it was reported that *S. cerevisiae* has the ability to stimulate growth factors, such as organic acids or vitamins, thereby stimulating cellulolytic bacterial populations (Chaucheyras *et al*., 1995; Zain *et al*., 2011). The addition of single microorganism such as *S. cerevisiae* to feed is only 1 g/head/day for sheep (Mardalena, 2000).

The principle by which yeast work has been explained briefly by Yoon and Stern (1995). They explained that yeast in the rumen is able to utilize oxygen so as to ensure anaerobic conditions for rumen bacteria and stimulate certain rumen bacterial populations. This situation is followed by increased utilization of ammonia and lactic acid so that the rumen pH is stable. Anaerobic conditions and stable rumen pH allow for more optimal microbial protein synthesis so that the total population of rumen bacteria increases and digestibility of crude fiber increases too. With increasing digestibility of crude fiber, it automatically increases consumption and supply of nutrients to the intestine. In the end it will increase the overall production response. Several studies using yeast as DFM in increasing the digestibility of feed ingredients are presented in Table 2.

Table 2. The use of yeast as DFM in increasing digestibility

No	DFM of Yeast	Species	Result	Reference
1	Supplementation with *S. Cerevisiae* in feed (22.5 g/d and 11.25 g/d).	Sheep	Digestibility of dry matter, NDF and ADF increased at a dose of 22.5 g/d and no effect at a dose of 11.25 g/d.	El-Waziry and Ibrahim (2007)
2	Supplementation with *S. cerevisiae* and addition of bioplus to ration	Sheep	A positive correlation was obtained at a dose of 4 g/day and feed conversion efficiency of 6 kg/kg body weight gain was produced.	Ratnaningsih (2000)
3	Supplementation with *S. Cerevisiae* at 4 x 10^9 CFU per day.	Sannen goat	Increased milk production by 14.4%.	Abd. El-Ghani (2007)
4	Supplementation with *S. Cerevisiae* at 3 g to 12 g in hay rations, corn leaf silage and concentrates.	Friesian Holstein cattle	Did not increase milk production.	Nikkah *et al.* (2004)
5	Supplementation with *S. cerevisae* (PSc) at 5.2 x 10^{11} CFU .	Brahman Cross cattle	Increased in meat production by 0.43 kg/ head/day.	Soeharsono (2010)
	Supplementation with *S. cerevisiae* at 5.2 x 10^{11} CFU	Friesian Holstein cattle	Increased in milk production by 15%.	
6	Use of yeast culture	Buffalo	Produced ADGW of 1.39 kg and 1.34 kg in test and control animals, respectively.	Adam *et al.* (1981)
7	Suplementation with yeast culture (0, 2.5, 7.5 g kg^{-1}/DM) on straw.	Cattle rumen fluid (*in vitro*)	There was an increased in dry matter digestibility in vitro.	Tang *et al.* (2008)
8	Suplementation with *S. Cerevisiae* at 4g/d	Sheep	Reduced the concentration of acetate and increased propionate.	Inal *et al.* (2010)
9	Suplementation with *S. Cerevisiae* at 0%, 0.25%, 0.50% and 0.75%	Cattle rumen fluid (*in vitro*)	Increased bacterial population compared to controls but not significantly different between treatments, 0.29; 4.99; 5.12 and 6.01.	Zain *et al.* (2011)

Note: DM = dry matter, CFU = colony forming unit, NDF = neutral detergent fiber, ADF = acid detergent fiber and ADGW = average daily gain weight

DFM of fungi species

DFM of fungi origin commonly used in ruminant are *Aspergillus niger* and *Aspergillus oryzae* (Llyod-Evans, 1989; Chen and Chiou, 2004). Offer (1990) and Newbold (1990), have used models to describe how probiotics of fungi origin works in ruminants in 6 stages. Increased livestock productivity was as a result increased feed consumption (stage 1). The increase in feed consumption occurred due to increase in fiber digestibility and microbial protein flow rate. In stage 2, increased in the digestion rate of fiber caused an improvement in the growth of microorganisms, due to the supply of minimal life requirements for the proliferation of microorganisms (Hobson and Wallace, 1982). The use of fungi probiotics caused an increased in the population of microorganisms in the rumen, as a result of the improvement in the activities of microorganisms (stage 3). Improved activities of these microorganisms occurred because of the stable rumen pH which is a secondary influence (stage 4). The stability of the rumen pH is thought to be caused by a decrease in lactic acid in the rumen (stage 5). Fungi probiotics form a substance that functions as a cofactor so as to increase lactate absorption by rumen microorganisms (Nisbet and Martin, 1991). This slows the release of oligosaccharide compounds which are precursors of lactic acid (stage 6). Some studies that used fungi as DFM in increasing the digestibility of feed ingredients are presented in Table 3.

Table 3. The use of fungi as DFM in feeds to increase digestibility

No	DFM of Fungi	Species	Result	Reference
1	Supplementation with *Aspergillus oryzae* at 6×10^{11} CFU/ml.	Cattle rumen fluid (*in vitro*)	Increased the total concentration of VFA, propionate, acetate in the rumen and the number of cellulolytic bacteria was higher than the control.	Beharka *et al.* (1991)
2	Supplementation with *Aspergillus oryzae* at 6×10^{11} CFU/ml.	Calf	Increased digestibility in cows and calves fed poor quality feed from 0.57 kg/day to 0.80 kg/day.	Wiedmeier (1989)

Note : CFU = colony forming unit and VFA = volatile fatty acid

The influence of combination of microorganisms as DFM

Researches using a combination of several microorganisms as DFM have been carried out to optimize livestock productivity. Amin (1991) stated that the supplementation of *S. cerevisiae* and *A. oryzae* combination increased the fermentation of feed in the rumen. This was due to the synergistic effects of the two microorganisms. *S. cerevisiae* is able to produce amylase enzymes which functions to digest starch, whereas *A. oryzae* produces cellulase enzymes and hemicellulase which are able to digest crude fiber (cellulose and hemicellulose). This statement was supported by Shin *et al.* (1989) who stated that *S. cerevisiae* is one of the commonly used microbes for livestock as a probiotic, together with bacteria and other fungi such as *A. niger*, *A. oryzae*, *B. pumilus*, *B. centuss*, *L. acidophilu*s, *S. crimers*, *Streptococcus lactis* and *Streptococcus termophilus*.

Widiawati and Winugroho (2007) reported that the supplementation of probiotics consisting of bioplus, *S. cerevisiae* and *Candida utilis* in feed increased milk production by 13%. Supplementation of microorganisms (*Aspegillus oryzae* and *S. cerevisiae*) increased dry matter digestibility, milk production, milk quality, and live weight of ruminants (Kung *et al.* 1997; Alshaikh *et al.* 2002). This occurred because of an increased in the number of cellulolytic bacteria, an increased in degradation of fibers in the rumen, and a change in the rumen's fatty acid content (VFA). Yeast also has the ability to provide growth factors, such as malic acid, which is used by lactate bacteria to improve pH (6.5-7) changes in the rumen. A combination of yeast cultures stimulated the use of hydrogen by rumen acetogenic (Kung *et al.,* 1997; Alshaikh *et al.,* 2002; Miller-Webster *et al.*, 2002; Sniffen *et al.*, 2004).

The combination of microorganisms as DFM in ruminants have not been widely reported. Doto and Liu (2011) reported on the combination of *B. licheniformis* and *Clostridium butyricum*, and a combination of both with yeast culture in vitro as DFM in ruminants. The combination of *B. licheniformis* and *C. butyricum* increased the digestibility of dry matter and organic matter, and were higher when *B. licheiformis* and *C. butyricum* were combined with yeast culture. Rojo *et al.* (2005) stated that, combination of *S. cerevisiae* and *B. licheneformis* increased the digestibility of organic matter in buffalo. Studies using combination of microorganisms as DFM to increase feed digestibility in ruminants is shown in Table 4.

Table 4. The use of DFM combinations in increasing digestibility

No	Combination of DFM	Species	Result	Reference
1	Supplementation with *S. cerevisiae* (57 g/d) and *A. oryzae,* 3 g/d	Dairy cattle	Increased digestibility of organic matter and protein; whereas fungal culture stimulated cellulolytic and proteolytic bacteria.	Yoon and Stern (1995)
2	Supplementation with *S. cerevisiae* (0.5 g/d)*, A. oryzae* (0.5 g/d) and their combination	Sheep	AO and SC + AO supplementations increased DM digestibility by 5% and 9%, respectively; SC, AO and SC + AO increased nitrogen digestibility.	Latif *et al.* (2014)
3	Supplementation with *Lactobacillus acidophilus* at 1×10^{9} CFU/bottle/kg and *S. cerevisiae* at 3×10^{9} CFU/bottle/kg	Calf	ADG and feed efficiency increased and were higher in calves who received feed supplemented with probiotics and enzymes	Malik and Bandla (2010)

Note : CFU = colony forming unit, AO = *Aspergilus oryzae*, SC = *Saccharomyses cerevisiae*, DM = dry matter, ADG = average daily gain

THE ROLE OF COBALT (Co) MINERALS IN RUMINANT

In addition to the use of technologies to reduce lignin content, increasing the quality and for that matter the digestibility of feed derived from oil palm pith can be done by adding the mineral Cobalt (Co). Co is most commonly found in the kidneys, adrenal glands, spleen and pancreas. Only a small amount of Co can be found in blood and milk. Cows' requirement for Co is higher than that of sheep. Ruminants' requirement for Co is relatively high due to their use in vitamin B12 formation and absorption. For calves, B12 is more necessary than adult cows (Parakkasi, 1999). For all ruminants, the NRC recommends a maximum tolerable level of 10 ppm (DM) (NRC of Sheep, 1985; NRC of Dairy Cattle, 2001) However, in the light of the above remarks this limit seems to be too low. The limit of 30 ppm (DM) seems to be sufficiently safe to protect ruminants from Co toxicity. (Puls, 1988)

Rumen microbes use Co for the formation of cyanocobalamin or vitamin B12 molecules. Supplementation with Co can improve appearance due to the recycle process into the rumen through saliva or rumen wall. Co is needed for the formation of vitamin B12 and rumen microorganisms have the ability to synthesize vitamin B12 from Co. Co requirements have been estimated to be 0.10 mg/kg of dry matter for beef (NRC, 2000) and 0.11 mg/kg of dry matter for dairy cattle (NRC, 2001). Co also affects the metabolism of some rumen microorganisms. In some bacteria, the propionate formation pathway from succinate to L-methylmalonyl CoA depends on vitamin B12 and Co (Tiffany and Spears, 2005). Co increases the proportion of propionate in rumen fluid and decreases the ratio of acetate: propionate to finisher sheep (Tiffany, 2003; Tiffany and Spears, 2005). Co supplementation increased colostrum and concentration of vitamin B12 in milk (Akins *et al.,* 2013). Schwarz *et al.* (2000) stated that, the Co requirement for beef cattle is 0.12 mg/kg for maximum body weight gain and 0.16 to 0.18 mg/kg for maximum feed consumption on DM basis. Co supplementation can improve fiber fermentability (Krisidayova, 2001). Studies using Co to influence fermentation in the rumen is presented in Table 5.

Table 5. The use of Cobalt (Co) minerals to influence fermentation in the rumen

No	Cobalt Treatment	Species	Result	Reference
1	Increased Co supplementation in feed (0.19 [a]), 0.37 [b]) or 1.0 [c]) mg Co/Kg of dry matter.	Lactation cattle	Did not affect dry matter digestibility, milk production and composition, plasma or serum concentration of vitamin B12.	Kincaid and Socha (2007)[a]) Kincaid *et al.* (2003)[b]) Akins *et al.* (2013)[c])
2	Supplementation of Co in feed from 0.10 to 1 mg/kg.	Buffalo	Increased consumption, ADG and ADFI; but decreased supplementation of Co to 0.05 mg/kg did not significantly increase ADG.	Tiffany (2003)
3	Supplementation of 0.5 ppm Co-sulfate in basal feed.	Priangan sheep	There was no significant effect on DM digestibility with a value of 932.30 g/head/day but there was a significant effect on body weight gain (BWG) and feed efficiency (FE) with a BWG value of 3940 g and FE of 7.45% compared to the control which was 858.32 g/head/day DM, 3620 g BWG, and 7.41% FE.	Latifudin *et al.* (2002)
4	Effect of concentrate substitution with cassava waste supplemented with cobalt and zinc	Sheep	The treatment of 50% field grass + 50% concentrate + 5 ppm cobalt + 30 ppm zinc gave the highest yield of VFA 213.11 mM, NH_3 15.61 mM, pH 6.96, dry matter digestibility 58.85% and organic matter digestibility 59.75%.	Hernaman *et al.* (2015)
5	Supplementing Co-glucoheptonate at a dose of 0, 5 and 10 ppm in the alfafa, hay, orchard grass, and corn leaves in vitro.	Cattle rumen fluid (*in vitro*)	There was no interaction on dry matter digestibility, organic matter, NDF and VFA.	Hussein *et al.* (1994)
6	Interaction of cobalt and molybdenum supplementation	Beef cattle	Supplementation of 8 mg/day Co increased blood hemoglobin, copper and iron levels in the liver but decrease molybdenum level.	Chapman and Kidder (1963)
7	Effect of cobalt supplementation at a dose of 0.04 mg/kg (control), 0.05, 0.10, and 1 mg/kg (growth phase and finisher phase)	Sheep	Increased the proportion of propionate during the finishing and growing phase and the plasma status of vitamin B12. It was concluded that the sheep until the finishing phase requires about 0.15 mg/kg Co and was not affected by the source of Co.	Tiffany (2003)

Note : ADG = average daily gain, ADFI = average daily feed intake, NDF = neutral detergent fiber and VFA = volatile fatty acid

CONCLUSION

Oil palm pith can be used as beef cattle feed when treated, and this will reduce the lignin content that serves as a limiting factor for its digestibility. The technologies that can be used to reduce lignin and increase digestibility includes ammonia technology using urea/ammonia, *Direct Fed Microbials* (DFM) using different microorganisms such as yeast, bacteria and fungi, and the addition of cobalt (Co). These technologies are capable of increasing fermentation in the rumen leading to increase in the conversion of meat in beef cattle.

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