



## **UTILIZATION OF SAGO DREGS AS RUMINANT FEED BY USING THE FERMENTATION METHOD: LITERATURE REVIEW**

### **Pemanfaatan Ampas Sagu sebagai Pakan Ternak Ruminansia dengan Menggunakan Metode Fermentasi: Tinjauan Pustaka**

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#### **ABSTRACT**

*Every year there is a change in the stock of ruminant feed that occurs in the rainy season and water shortages in the dry season. Utilization of agricultural waste as an alternative feed is one way to overcome these problems. One of the wastes that have the potential to be used as feed ingredients is sago waste. Sago pulp is a waste that is rich in lignocellulose, namely cellulose. Several biotechnology applications in ruminant feed fermentation can improve properties such as taste, aroma, shelf life, texture and nutritional value of food. Fermentation using mold or yeast, as well as bacteria or a mixture of various microorganisms can increase the nutrients in the feed needed by ruminant feed. Processing of lignocellulosic materials is required to obtain optimal degradation results. The degradation process will convert lignocellulosic material into raw materials that are easily digested by the ruminant. Enzymes produced by microorganisms can increase crude protein, crude fat, carbohydrates, crude fiber, vitamins and minerals. Thus, the fermentation method of sago dregs and its use as feed can increase the nutritional value, so that productivity can be increased when given as feed.*

**Keywords:** *Enzymes, fermentation, ruminant feed, sago dregs, waste*

#### **ABSTRAK**

Setiap tahun terjadi perubahan stok pakan ternak ruminansia yaitu pada musim hujan dan kekurangan air pada musim kemarau. Pemanfaatan limbah pertanian sebagai pakan alternatif merupakan salah satu cara untuk mengatasi permasalahan tersebut. Salah satu limbah yang berpotensi untuk dimanfaatkan sebagai bahan pakan adalah limbah sagu. Ampas sagu merupakan limbah yang kaya akan lignoselulosa yaitu selulosa. Beberapa aplikasi bioteknologi dalam fermentasi pakan ternak ruminansia dapat meningkatkan sifat-sifat seperti rasa, aroma, umur simpan, tekstur dan nilai gizi makanan. Fermentasi menggunakan kapang atau khamir, serta bakteri atau campuran berbagai mikroorganisme dapat meningkatkan nutrisi dalam pakan yang dibutuhkan pakan ruminansia. Pengolahan bahan lignoselulosa diperlukan untuk mendapatkan hasil degradasi yang optimal. Proses degradasi akan mengubah bahan lignoselulosa menjadi bahan baku yang mudah dicerna oleh ruminansia. Enzim yang dihasilkan oleh mikroorganisme dapat meningkatkan protein kasar, lemak kasar, karbohidrat, serat kasar, vitamin dan mineral. Dengan demikian, metode fermentasi ampas sagu dan pemanfaatannya sebagai pakan dapat meningkatkan nilai gizi, sehingga produktivitas dapat meningkat bila diberikan sebagai pakan.

**Kata Kunci:** Ampas sagu, enzim, fermentasi, limbah, pakan ruminansia

## INTRODUCTION

Indonesia is a developing country where high population growth and economic progress are driving increased demand for animal-based foods (Widi 2015). As most consumers are Muslim, beef and chicken are the two most popular meat proteins in Indonesia (Agus and Widi 2018). Until now, there has been a gap between the supply of ruminant feed and the demand for beef. Feed plays an important role in the success of a livestock feed business. Breeders go out of village areas and even districts to look for forage sources with distances that can reach up to tens of kilometers (Handayanta et al. 2015).

Although the performance of an animal is greatly influenced by its genetic value, improper feeding, such as feeding poor quality feed, will lead to low performance (Edi and Irwansyah 2020). The results showed that feed had an effect of 60-70% on livestock productivity (Bidura 2017). Forage is the main source of feed for ruminants, so when the production of ruminants increases, it leads to an increase in the supply of ruminant feed in terms of both quality and quantity (Sari et al. 2016). Continuous fluctuations in the availability of forage always occur every year, there is an excess of feed during the rainy season and a shortage of feed in the dry season (Handayanta et al. 2015). If the government does not address this problem, it will have an impact on the price of meat and milk. The diversity of local feeds in Indonesia also creates different feed qualities between regions (Nuraini et al. 2019). Agricultural waste accumulates on farmland and can be used as a feed source.

The utilization of agricultural waste used for alternative ruminant feed can overcome these problems. One of the wastes that have the potential to be used as feed ingredients is sago waste. Indonesia has the largest area of sago forest in the world, with the most growth in the provinces of Papua and West Papua covering an area of 1.28 million ha, with a total population of sago palms estimated at around 27 million trees in the stem stage, 21.1 million trees in the harvest stage and 5.5 million trees in the over-harvesting stage (Dimara et al. 2021, Yater et al. 2019). Sago (*Metroxylon sago*

Rottb.) is a tree species from the Areaceae family (palm family) (Kadir et al. 2022). Sago can be grown in freshwater, peat and swamp areas, around water sources with low salinity, along river banks or in mineral soils containing more than 70% clay and 30% organic matter (Vita 2018). The starch fiber residue from the pieces of the sago palm or better known as sago palm pulp is composed of starch and lignocellulose components, which are 54.6% starch, 31.7% of cellulose and hemicellulose and 3.3% of lignin (Husin et al. 2019).

Fermentation is a desirable process of biochemical modification of major food matrices carried by microorganisms and their enzymes to improve the nutrition of poor constituents (Nkhata et al. 2018). The high polysaccharide content in sago pulp makes this agricultural residue a promising raw material for further fermentation. Sago dregs waste contains lignocellulose which is very rich in cellulose, which is then optimally utilized by bacteria and fungi as a carbon source (Nuraini 2015). Cellulose is a natural polymer that is biocompatible and the most abundant and is very environmentally friendly because cellulose is easily degraded, renewable and non-toxic (Mulyadi 2019).

Cellulose is always found together with other lignocellulose content such as starch, hemicellulose, and lignin (Mulyadi 2019). Hemicellulose is the second most natural carbohydrate biopolymer after cellulose, counted for approximately 20-30 wt% (Lu et al. 2021). Hemicellulose is composed of pentoses ( $\beta$ -D-xylose and  $\alpha$ -L-arabinose), hexoses ( $\beta$ -D-mannose,  $\beta$ -D-glucose,  $\beta$ -D-galactose), glucuronic acid, small amounts of L-rhamnose and L-fucose units, the most abundant hemicellulose polymer is xylan (Ruiz et al. 2013). The hemicellulose content and structure, main chain length and type, and side chain distribution and type vary among lignocellulose types (Lu et al., 2021). Lignin is a complex aromatic cell wall polymer that increases as plants mature (Zhong et al. 2021). Lignin is a complex polymer of phenylpropane units held together by various chemical bonds (Rynk et al. 2022). Lignin can be considered an anti-nutritional factor that limits feed digestibility (Chanjula et al. 2018). The removal of lignin components can be carried out with the help

of microorganisms through a fermentation process and the lignin removal process will be explained in the next sub-chapter (Iram et al. 2021). This review discusses how fermented food biotechnology replaces conventional feed and can reduce the environmental impact of waste.

## THE FERMENTATION OF SAGO DREGS

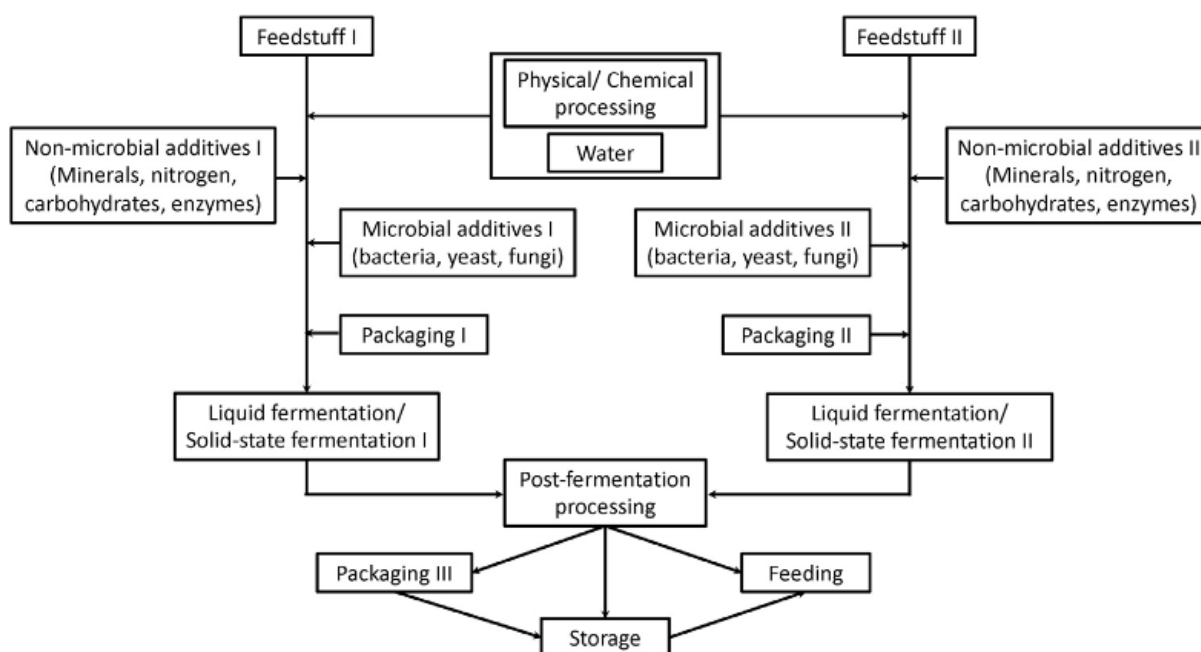
Fermentation is a process or change that causes large organic molecules to be broken down into simpler molecules using microorganisms. For example, yeast enzymes can convert sugars and starches contained in ingredients into alcohol, and proteins into peptides or amino acids. The enzymatic activity of microbes in feed components tends to cause the desired biochemical changes and results in significant changes in ruminant feed. Fermentation is a natural way to improve agricultural waste to produce vitamins, essential amino acids, nutraceuticals, proteins, and the appearance, taste and aroma that cattle liked (Sharma et al. 2020). Acceleration of the fermentation process and the growth of microorganisms require additional nutrients (Suryani et al. 2017). Bacteria require a minimum of nutrients such as water, carbon sources, nitrogen sources and mineral salts to grow (Bonnet et al. 2020). Fermentation can use mold, yeast, bacteria or a mixture of several microorganisms.

Fermentation involves a variety of activities from a mixture of several species of microorganisms or certain microbes (Agustina et al. 2019). To achieve optimal growth and yield, farm animals require a complete and balanced intake of nutrients. In achieving optimal growth and production, ruminants need a complete and balanced nutrient intake. Imbalances between energy requirements and nutrient intake are often associated with fatty liver, ketosis (clinical or asymptomatic), gastric acidosis (subacute or acute), milk fever (asymptomatic) symptomatic or clinical), immune dysfunction (placental defect, met, mastitis) (Wankhade et al. 2017). Nutrients in feed needed by ruminants include crude protein, carbohydrates, amino acids, fatty acids, minerals, vitamins, and water (Erickson and Kalscheur 2019). Fermented feed can be

given to livestock if it has been tested and meets the nutritional standards required by ruminants. The feed standards are set according to the production (milk, meat, eggs, wool), product composition (milk fat content), and physiological status (growth, fetal development) of the animal (Tahuk et al. 2021).

Fermentation of ruminant feed can be done by two methods such as liquid or solid fermentation. Such fermentation may involve one or two steps. In the one-step process of liquid fermentation or solid phase I fermentation, the fermented products can be used directly for ruminant feed. This one-step fermentation product can also be further processed using drying and mixing using other nutrients (fermented or unfermented) and feed additives (e.g. minerals and vitamins). In the two-stage fermentation process, the products of liquid fermentation or solid fermentation I can be used as starting materials for feedstuff II. Feedstuff II can be further fermented to improve its nutritional quality. Physical or chemical treatments of feed ingredients such as desizing, milling, addition of acid or alkaline solutions, extraction of organic solvents and autoclaving and addition of water can be used prior to fermentation. The addition of "non-microbial additives" such as minerals, nitrogen (e.g. ammonia), carbohydrates (sugars) and enzymes (fibrinolytic enzymes, phytases and enzymes that break down nutritional factors) are expected to accelerate the process of fermentation and eliminate anti-nutritional factors (Dai et al. 2019). The schematic diagram of feed fermentation is shown in Figure 1.

Cui et al. (2021) conducted a study using the fungus *A. niger* and *T. koningii* on tea dregs. After fermentation, there was an increase in crude protein content of 33.19% which was fermented by *A. niger* and 14.77% which was fermented by *T. koningii*. The increase in crude protein was due to the increased mycelial growth of the fungus. Protease activity and large amounts of cellulose in the raw material are destroyed by fungi, thus increasing the amount of amino acids. Fermentations carried out by *T. koningii* had higher reducing sugar levels in *A. niger*, indicating that the enzymatic activity of *T. koningii* was higher than that of *A. niger*. The taste of *T. koningii* is superior to



**Figure 1.** Schematic diagram of the techniques and procedures used in the production of fermented feed for livestock feed (Dai et al. 2019).

that of *A. niger* because monosaccharides are a type of water-soluble sugar that can mask bitterness. In addition, increased reducing sugar content has positive effects on digestion, absorption, and energy supply in animals. Suebu et al. (2020) found that the longer the fermentation time used, the more material was degraded into cell mass by microorganisms. Cell mass is a single-cell protein, so it can affect the crude protein content, which increases at the end of the fermentation process.

## THE ROLE OF FEED FERMENTATION

### Color

Falola et al. (2013) in their research stated that the color of a well-fermented ration is close to the color before fermentation. The color change that occurs in fermented feed is caused by an ongoing aerobic respiration process as long as the availability of oxygen is still there, until the carbohydrate content in the material runs out (Christi et al. 2019). According to research by Kung et al. (2018), wet silage with too much acetic acid is also yellow, especially at the bottom of the silo, because the compaction effect increases moisture in that area.

During the fermentation process, there is a temperature instability that can cause a change in the color of a feed from brown to very dark (Christi et al. 2019). Polyorach et

al. (2013) stated that a fermentation process that exceeds heat can cause a change in the color of the feed to become charred. This is in line with research conducted by Christi et al. (2019) using the bacteria *Saccharomyces cerevisiae* and EM-4. The results showed that the resulting fermented concentrate was dark in color while the unfermented concentrate was lighter in color.

Research conducted by Saputro et al. (2015) used the fungus *Trametes sp* and variations in temperature. The results obtained were fermentation for 6 days had a significant effect on the aroma, color and texture of pineapple leaves. This was due to the fact that on the 2<sup>nd</sup> and 4<sup>th</sup> days the growth of the *Trametes sp*. was not yet optimum which causes a color change from lignin degradation by *Trametes sp*. not very visible. The fungus will continue to degrade lignin the longer the fermentation takes, causing the leaf color to appear darker (Saputro et al. 2015).

The only microorganisms that can degrade lignin are wood rot fungi which are classified into the Basidiomycetes class (Saputro et al. 2015). In nature, three groups of fungi can degrade wood components (lignocelluloses), namely brown rot (e.g. *Lentinus lepideus*, *Polyporus sp*, *Phlebia brevispora*, etc), white rot (e.g. *Schizophyllum commune*, *Pynoporus sanguineus*, etc) and soft rot (*Chaetomium globosum*) (Suprapti et al. 2020, Rahim et al.

2019). White rots can degrade both polysaccharides and lignin, but while brown rots can rapidly and extensively depolymerize cellulose, they can only partially modify lignin (Singh and Singh 2016). Brown rot fungi preferentially attack cellulose content, leaving lignin and browning the degraded residue (Saputro et al. 2015). Soft rot fungi enzymatically degrade cellulose in wood tissue, while their hyphae secrete cellulase (Langer et al. 2021).

### Flavor

The good fermented livestock feed should smell good and be mixed with an acid (Budiyanto 2013). Fermentation pH is proportional to total acid. So a lower pH indicates a higher total acid (Miksusanti et al. 2019). According to Suwignyo et al. (2015) complete feed ferments that have been fermented for two weeks have a pungent smell like that of fermented milk due to the long fermentation time and lactic acid. Well-fermented feeds have an acidic taste, and low pH and retains the color of the original material (Marhamah et al. 2019). According to Kung et al. (2018), silage with fruits and sweet flavors is wrongly associated with stable and well-fermented feeds. These odors are usually caused by high concentrations of alcohol (ethanol), which is produced mainly by yeasts, but also by various bacteria. Also, alcohols can react with silage acids to form esters, resulting in fruity aromas.

Many microorganisms can be used during fermentation to produce aromas that livestock feed can enjoy. Feed odor can increase feed palatability and nutrient absorption by cattle, which can increase cattle weight. Some lipolytic microorganisms can degrade phospholipids, lipids and their derivatives, resulting in unpleasant odors (Suningsih et al. 2019). There are also proteolytic microorganisms that can degrade proteins and other nitrogen-containing components, resulting in undesirable odors such as rancidity or decomposition (Suningsih et al. 2019).

Moisture content can also affect fermented foods. Miksusanti et al. (2019) conducted a study on fermented parts with water content varying between 40%, 50%, 60% and 70%. Based on the results

obtained, the physical quality of the portions fermented at 40% moisture indicates a slightly sour aroma, light brown and dry texture. The rations fermented at 50% moisture content appeared similar in both odor and color, but had a moist texture. At 60-70% moisture fermentation, rations with a strong sour smell, light color and very wet texture were obtained. The last two results show that there is a decrease in feed quality and an increase in microbes that can be harmful.

### Texture

The texture is one way of showing the taste of the surface of the material that is made so that it produces a good or bad quality response (Christi et al. 2019). Dry or not a product from fermentation, the texture of the resulting feed depends on the water content of the material (Christi et al. 2019). If there is less water content in the material, then the texture of the fermented product produced is slightly dry and can even become very dry, on the contrary if the water content of the material is high, it produces a slightly wet texture until it becomes wet (Telew et al. 2017).

This is in by following the research conducted by Miksusanti et al. (2019) that fermented rations with 60% and 70% water content decreased crude protein. High water content not only accelerates the growth of beneficial microorganisms, but also leads to the growth of harmful microorganism; a condition that causes microbial growth to increase microbial mass is hampered. Physical appearance shows that some fungi grew in the fermented portion with a sour smell, light color and wet texture (Miksusanti et al. 2019).

Karyono and Novita (2020) conducted research on silage fermentation with variations of the coffee hull, banana suckers, banana weevil, and Banana head. The results of analysis of variance and data tables of fermented coffee husk silage with banana sucker showed no effect on the structure of coffee husk fermented feed. Because the composition of the silage material is coffee husk waste which has almost the same size and shape, the degradation of the material by microorganisms is as good as all the parameters tested. In addition, the resulting

silage texture is medium (not too hard). The structure of the resulting silage from coffee skin can be classified as good because it is close to the original structure (Karyono and Novita 2020).

Livestock generally prefer soft-textured diets to coarse-textured diets (Marhamah et al. 2019). Changes in the chemical and physical properties of livestock feed occur due to microbial activity (Sharma et al. 2020). According to Utama and Christiyanto (2021) fermentation is the transformation of chemical, physical and biological structures from complex to simple in order to achieve efficient digestibility of feed. Traditional biotechnology is a biotechnological process that occurs in feed or food ingredients by adding enzymes or specific microorganisms that cause physical, taste, and external changes as a result of biological processes in the material (Suningsih et al. 2019).

### Proximate content

Ruminants require more forage than other types of feed. Good green fodder depends on its nutritional content and palatability. Complete feed in the form of a mixture of various fermented feed ingredients can be a solution in feeding s under various conditions (Pakpahan and Restiani 2019). Proximate analysis is one of the most widely used variables to determine the nutritional quality of feed ingredients (Wardono et al. 2022). Proximate analysis aims to determine the percentage of protein, fiber, lipid, ash and moisture content of formulated livestock feed (Osman et al. 2019). Based on research conducted by Wardono et al. (2022) using sago dregs by

using variations in fermentation time and a mixture of multi-microbial inoculum in Saus Burger Pakan (SBP) with a dose of 0.5% and the addition of 0.4% urea. The results obtained showed significant differences from sago pulp before and after fermentation which can be seen in Table 1.

The increase in crude protein in Table 1 is thought to be due to the addition of protein donated by microbial cells to produce single cell protein (SCP) (Wardono et al. 2022). This is also supported by the research of Nurhayati et al. (2022) which states that the increase in crude protein content of fermented feed is caused by the large number and types of microbes, so that more and more microbes can decompose complex feed ingredients into simpler ones, which can then be used by microbes to multiply. Where the microbe itself is a single cell protein source, so that the protein content can increase.

Fiber in plants is usually tightly bound to lignin, which causes the fiber to not be broken down in the digestive system, thereby reducing digestibility (Permana et al. 2020). Processing of feed ingredients can be done as an effort to increase the amount of nutrients from feed ingredients that are absorbed in the digestive tract. The processing of sago dregs that can be applied to farmers easily and at low cost is biological processing by fermentation (Hossain et al. 2017). The principle of biological processing by means of fermentation is the use of ideal microbes that have the ability to degrade lignin. Lignin can be considered as an anti-nutritional factor, limiting feed digestibility (Chanjula et al. 2018). Several groups of

**Table 1** Nutrient content of sago dregs before and after fermentation (Wardono et al. 2022)

No.	Description of feedstuff	Dry matter	Ash	Percentage based on dry matter				
				Crude protein	Crude fat	Crude fiber	NDF	ADF
1	SHBF	89.68	11.02	3.50	1.16	38.57	84.53	65.38
2	SHAF 1	68.92	5.80	4.64	1.37	34.81	70.54	53.25
3	SHAF 2	71.89	7.86	3.64	1.18	33.24	83.79	51.02
4	SHAF 3	69.77	7.34	3.28	0.48	33.01	65.63	49.17
5	SHAF 4	70.55	7.37	3.13	0.97	32.01	58.91	50.65
6	SHAF 5	69.15	6.66	3.54	0.16	27.84	59.80	49.33
7	SHAF 6	82.07	7.74	3.71	1.33	31.04	68.94	48.04

Note : SHBF (Sago dregs before fermentation),  
 SHAF 1 (Sago dregs after fermenting for 12 hours)  
 SHAF 2 (Sago dregs after fermenting for 24 hours)  
 SHAF 3 (Sago dregs after fermenting for 48 hours)  
 SHAF 4 (Sago dregs after fermenting for 72 hours)  
 SHAF 5 (Sago dregs after fermenting for 120 hours)  
 SHAF 6 (Sago dregs after fermenting for 168 hours)

fungi have the ability to produce lignocellulose enzymes. Enzymes produced by fungi or bacteria are useful for decomposing compounds containing carbon sources. The enzymes produced work synergistically with oxidative mechanisms to convert lignin into smaller molecules with different chemical properties (Iram et al. 2021). Different types of C–C bonds in phenylpropane units make lignin difficult to degrade (Cagide and Castro-Sowinski 2020).

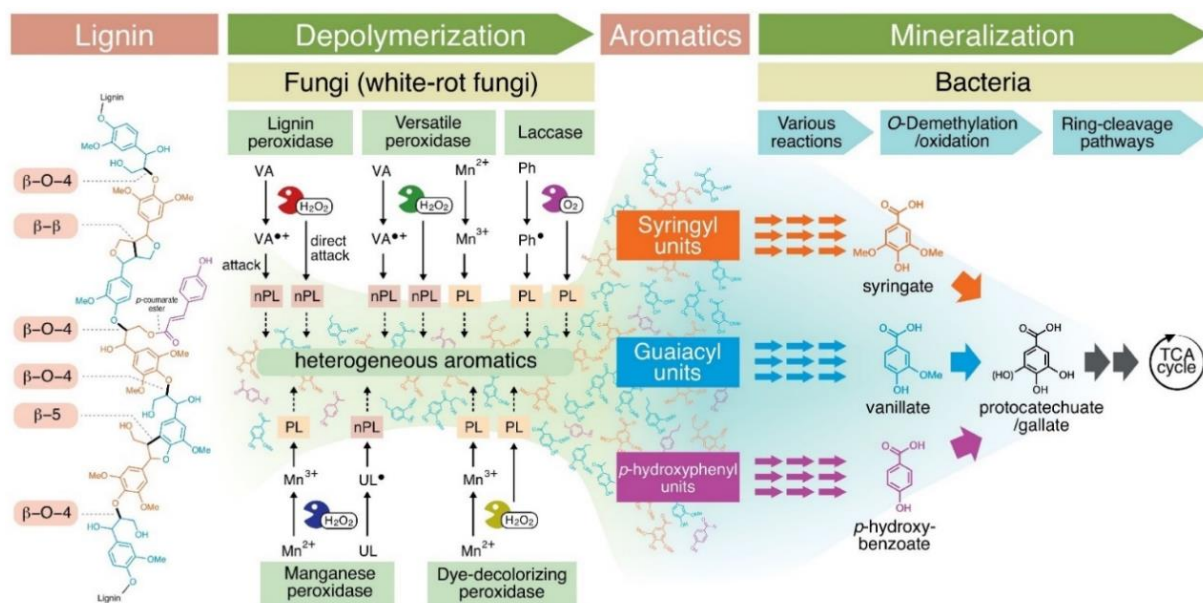
In the last two decades, there is evidence that fungal strains, especially white and brown rot fungi, can degrade plant cell wall components through the production of free hydroxyl ions ( $\text{OH}^-$ ). First, hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) is produced, which aids the oxidation and production of  $\text{OH}^-$ . These free radicals then attack different cell wall components along with lignin and break the bonds between these structures. As a result, plant cell walls become more susceptible to other lignocellulolytic enzymes (as illustrated in Figure 2). The fungus *Phanerochaete chrysosporium*, for example, has enzymes to break down lignin and cellulose. It also has the ability to decompose toxic substances that are persistent, due to the presence of dehalogenase enzymes, lignin peroxidase, and manganese peroxidase (Rahim et al. 2019). Lignin peroxidase (LiP) and manganese peroxidase (MnP) are

extracellular peroxidase enzymes that use  $\text{H}_2\text{O}_2$  to degrade lignin. LiP is an extracellular peroxide enzyme that oxidizes aromatic compounds (phenolic and non-phenolic) by transferring 1 electron to produce phenoxy (Dimawarnita and Panji 2019).

LiP catalyzes the  $\text{H}_2\text{O}_2$ -dependent oxidative depolymerization of various non-phenolic lignin compounds (diarylpropane), non-phenolic lignin model compounds  $\beta$ -O-4 and various phenolic compounds (eg guaiacol, vanillyl alcohol, catechol, syringic acid, acteosyringone) with potential redox. LiPs oxidize substrates in multi-step electron transfer and form intermediate radicals, such as phenoxy radicals and veratryl alcohol radical cations. These intermediate radicals undergo non-enzymatic reactions such as radical coupling and polymerization, side chain cleavage, demethylation and intramolecular addition and rearrangement. Unlike other peroxidases, such as MnP, LiP is able to oxidize non-phenolic aromatic substrates and does not require the participation of a mediator due to its very high redox potential (Iram et al. 2021, Pollegioni et al. 2015).

## FERMENTATION MECHANISM

The limited protein content and high fiber content of sago pulp as feed can be processed using fermentation technology.



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**Figure 2** Different enzymatic pathways of microbial lignin degradation (Iram et al. 2021)

Therefore, the use of fermented food processing technology is expected to maintain or even improve the quality of nutrition and improve the taste, making it a new feed for farmers (Irwansyah and Junaedi 2019). In Table 2, it can be seen some microorganisms that are often used in fermenting livestock feed in order to produce good nutritional quality.

The mechanism of fermentation of sago dregs as livestock feed occurs in 4 phases. The first phase is known as the aerobic phase. At this stage, the sago dregs should be packed and sealed to remove as much oxygen as possible. During this early stage, the remaining oxygen is depleted as the forage cells absorb the oxygen and convert it to carbon dioxide through cellular respiration. Aerobic bacteria also use the remaining oxygen from this stage in combination with plant sugars to produce carbon dioxide, water, and heat. It continues until it is exhausted (Adesogan and Newman 2010).

The second phase is known as the lag phase. The condition of the bacteria in this phase is that the bacteria adapt to environmental conditions; they are mature and have not had a chance to divide (Benjamín et al. 2022). When the remaining oxygen is used up, the plant cells break down and are used by bacteria as a food source. At this stage, the bacteria produce

enzymes that can break down complex carbohydrates, starches and fibers into simple sugars that the bacteria can easily use. Enzymes also break down plant proteins at this time, making the protein more soluble. The bacteria use the cell juices produced in the late phase to grow in the fermentation phase. At this time the pH of sago pulp fermentation decreased (and acidity increased) to around 5.7-5.5 (Pretz 2020).

Livestock cannot synthesize enzymes for the breakdown of cellulose (Siagian 1998). Cellulose is a polysaccharide that is the main part of plant support materials, such as cell walls. Because livestock cannot synthesize cellulose, the breakdown of cellulose and cellobiose can only occur with the help of enzymes produced by microbes (Siagian 1998). Research conducted by Yuliana and Chuzaemi (2019) using *A. oryzae* succeeded in increasing crude protein up to 27.04%.

Fungus *A. oryzae* is known as one of the molds that produces the most enzymes such as cellulase,  $\alpha$ -amylase, and  $\alpha$ -galactosidase (Yuliana and Chuzaemi 2019).  $\alpha$ -amylases catalyze the cleavage of  $\alpha$ -1,4-glycosidic bonds from starch with the formation of lower molecular weight compounds, such as glucose, maltose, and oligosaccharides (maltotriose, dextrin) (Blaga et al. 2022). Cellulases play a major

**Table 2** Some microorganisms that can be used in livestock feed

Microorganism	Enzymes produced	Function	Reference
<i>Streptococcus sp.</i>	Extracellular amylase enzyme	Starch hydrolysis	Tjahjaningsih et al. 2016
<i>Pediococcus sp.</i>	Lipase enzyme	Fats hydrolysis	Tjahjaningsih et al. 2016
<i>Saccharomyces cerevisiae</i>	Invertase and zimase enzymes	Disaccharides hydrolysis	Rizwan et al 2018
<i>Bacillus lentus</i>	Protease enzyme	Protein hydrolysis	Artha et al. 2019
<i>Lactobacillus acidophilus</i>	Protease and $\beta$ -galactosidase enzymes	Protein and lactose hydrolysis	Suradi et al. 2019
<i>Bacillus subtilis</i>	Subtilisin enzyme and mettaloproteases enzyme	Hydrolyzes peptide bonds and ester bonds	Razzaq et al. 2019
<i>Aspergillus niger</i>	Chitinase enzyme	Chitin hydrolysis	Purkan et al. 2016
<i>Trichoderma reseei</i>	Phytase enzyme	Phytic acid hydrolysis	Paloheimo et al. 2016
<i>Serratia marcescens</i>	Cellulase enzyme	Cellulose hydrolysis	Kurniawati et al. 2021
<i>Lasiodiplodia theobromae</i>	Lypoxygenases	Oxygenated on polyunsaturated fatty acids containing cis-1,4-pentadiene in the lipid	Patel et al. 2015



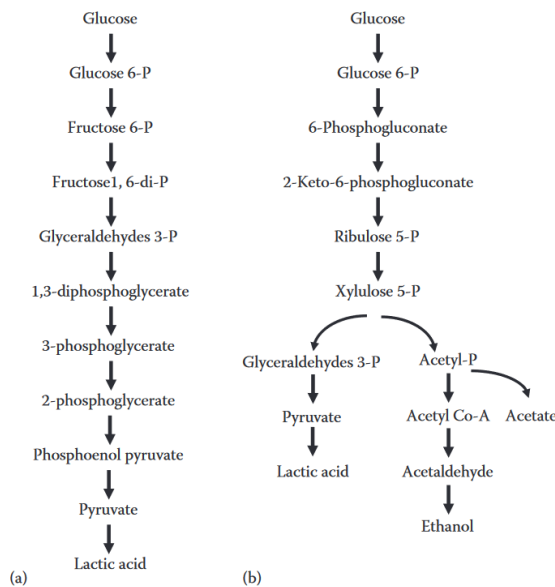
role in the hydrolysis of cellulose substrates and their conversion to monomeric products. They hydrolyze the  $\beta$ -1,4-glycosidic bonds of cellulose and are synthesized by microbial strains during growth on cellulosic material (Singh et al. 2019). Cellulase is a complex enzyme consisting of endocellulase or endo- $\beta$ -1,4-glucanase, exocellulase or exobiohydrolase, and  $\beta$ -1,4 glucosidase or cellobiase (Nugrahini et al. 2016).

Exo- $\beta$ -1,4 glucanase or known as cellobiohydrolase works by removing units from cellobiose at the end of the cellulose chain (Megawati and Damayanti 2020). In crystalline cellulose, the activity is very high, but in amorphous cellulose it is very low (Nugrahini et al. 2016). Endo- $\beta$ -1,4-glucanase hydrolyzes cellulose randomly which will then produce cellodextrin, cellobiose, and glucose (Megawati and Damayanti 2020). This enzyme is active in breaking the bonds of amorphous cellulose such as carboxyl methyl cellulose (CMC) (Anindyawati 2009).  $\beta$ -1,4-glucosidase or cellobiase hydrolyzes short cello-oligomers and cellobiose which then produces glucose (Anindyawati 2009).

The third stage is known as the log phase, where the conditions of lactic acid bacteria begin to grow at a constant time interval and will continue indefinitely, produce lactic acid and acetic acid, and ferment the remaining sago to increase

acidity (Benjamín et al. 2022, Pretz 2020). Lactic acid is stronger than acetic acid, so it lowers the pH of fermentation in sago dregs more than acetic acid. There are two types of lactic acid bacteria: homofermentative and heterofermentative. It can be seen in Figure 2 the pathways of two types of lactic acid bacteria. Homofermentatives mainly produce lactic acid, while heterofermentatives mainly produce lactic acid, acetic acid, ethanol and carbon dioxide. Homofermentative bacteria preferred in fermentation because it runs faster, providing more nutrients to the cows and allowing better preservation of the livestock feed. At least 70% of the total acid in properly preserved sago pulp must be lactic acid. Higher temperature and higher humidity lead to higher initial levels of lactic acid bacteria in sago dregs fermented feed. The anaerobic fermentation stage typically lasts about two weeks and the feed is cooled to near room temperature (Pretz 2020).

The fourth phase is known as the stable phase. The pH of sago dregs fermentation should be between 3.8 and 4.2 which is a sign that the fermentation has reached a stable stage where fermentation is maintained and bacterial growth has slowed or stopped. If the available sugar is depleted before the pH of the sago dregs reaches a low pH value, fermentation stops before the sago dregs reaches a steady state, resulting in a poor fermentation profile, reduced



**Figure 3** (a) Homofermentative and (b) heterofermentative pathways of lactic acid bacteria. Homofermentative bacteria consume or ferment glucose that yields lactic acid as the primary end metabolite. Heterofermentative bacteria also consume glucose; common end products are lactic acid, ethanol, and carbon dioxide (Kumar et al. 2015).

palatability and reduced nutrient availability for livestock feed. Forage quality improvement can last for four to six months of storage due to the continuous activity of bacteria and nuclear enzymes that help solubilize protein (Adesogan and Newman 2010). This is in line with research conducted by Effendi (2018), using *L. acidophilus* for 7 days is considered to be able to reduce crude fiber levels in food and can increase crude protein levels according to Indonesian National Standards (SNI).

In the fermentation process, these bacteria produce protease enzymes and  $\beta$ -galactosidase enzymes (Suradi et al. 2019). Proteases represent a broad group of enzymes that degrade or hydrolyze proteins or peptides. Proteases act on and cleave peptide bonds that connect adjacent amino acid residues within protein molecules, forming shorter peptides and amino acids (Razzaq et al. 2019). In terms of peptide bond termination sites, proteases can be divided into endopeptidases or proteinases and exopeptidases (Cahyaningati 2019).

Exopeptidases act only near the ends of polypeptide chains. Based on the N-terminal or C-terminal site of action, they are divided into aminopeptidases and carboxypeptidases. Aminopeptidases act at the free N-terminus of polypeptide chains, liberating single amino acid residues, dipeptides, or tripeptides. Carboxypeptidases also act at the C-terminus of polypeptide chains releasing single amino acids or dipeptides. Carboxypeptidases can be divided into three major groups based on the nature of the amino acid residues in the enzyme's active site: serine peptidases, metallopeptidases, and cysteine peptidases. Endopeptidases are characterized by preferentially acting at peptide bonds in the internal region of the polypeptide chain. Endopeptidases are classified into four subgroups based on their catalytic mechanisms: serine proteases, cysteine proteases, aspartic proteases, and metalloproteases (de Souza et al. 2015).

The  $\beta$ -galaktosidase enzyme produced by bacteria can break down lactose into glucose (Khusniati et al. 2015). Glucose fermentation has a principle consisting of two stages, in the first stage there is a breakdown of the carbon chain from glucose and also the release of at least two pairs of

hydrogen atoms, which then produces other carbon compounds that are more easily oxidized than glucose. The oxidized compound is then reduced again by a hydrogen atom released in the first stage, and then forms another compound as a result of the fermentation process (Leoanggraini and Muhadi 2011).

The addition of probiotic supplements or by adding live microorganisms is a method that can be used in fermentation technology. The workings of the probiotic itself is to help inhibit the growth of disturbing microorganisms in the digestive system and can reduce the degree of acidity (Suryani et al. 2015). Effendi (2018) stated that the use of probiotic microorganisms in livestock feed can support optimal conditions in the rumen and can stabilize conditions in the rumen. The addition of probiotic microorganisms can also be used as an agent in helping cow digestion to break down carbohydrates in existing livestock feed so that it is easier to digest (Effendi 2018).

## CONCLUSION

The method of processing sago dregs by fermentation as livestock feed can improve nutrition, improve texture, aroma, and taste even better so that it can increase its productivity if given to livestock as feed. The use of this fermentation technique is expected to increase the nutritional content of the feed, increase the palatability of livestock to feed and assist livestock in the absorption of feed, and can also assist in the provision of feed in the dry season for the community. The application of bacteria and fungi for the food or feed biotechnology industry sees potential and requires further research, especially the addition of pretreatment before fermentation in order to improve the performance of enzymes produced by bacteria and fungi. The method of fermentation of sago dregs should be investigated and also evaluated further in order to obtain a product with high nutritional value.

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