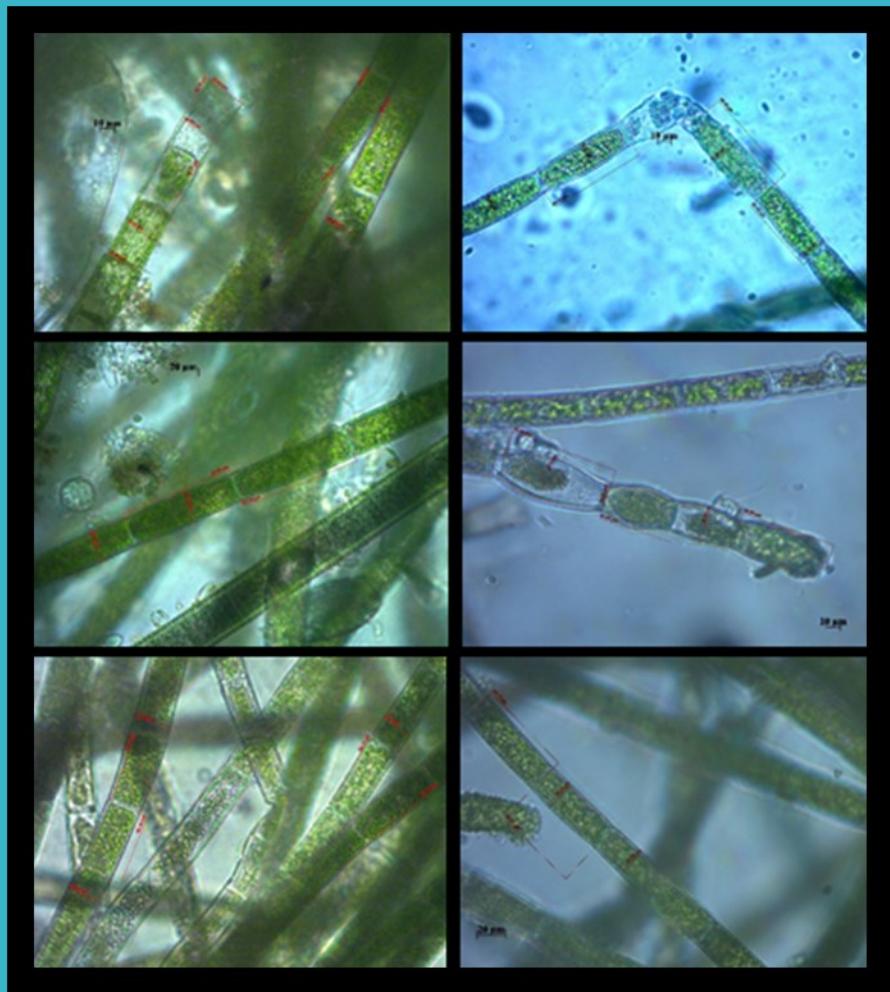


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Keterangan foto cover depan: Pertumbuhan *Oedogonium* sp. pada perlakuan cahaya yang berbeda. *Oedogonium* sp. Pada kultur Outdoor tampak lebih padat daripada kultur indoor, sesuai dengan halaman 309
(Notes of cover picture): (*Growth of Oedogonium* sp. at different light treatments. *Oedogonium* sp in outdoor culture appeared denser than in indoor culture, as in page 309)



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THE IMPORTANCE OF RUMEN ANAEROBIC FUNGI ON FIBER DEGRADATION IN RUMINANTS: REVIEW

[Pentingnya Fungi Anaerob Rumen dalam Mendegradasi Serat pada Ruminansia: Review]

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ABSTRACT

Forage is a feed source for ruminant livestock, but one of the limiting factors of forage is the high fiber content in old forage plants. Rumen microbes can only degrade the fiber content in forage. One of the rumen microbes that has fiber degrading activity is rumen anaerobic fungi because it can produce very active enzymes to degrade lignocellulose. The rumen anaerobic fungi are divided into several genus which are grouped base on the number of flagella in zoospores, thallus morphology and rhizoid type. The presence of fungi in the rumen is very important because fungi can form rhizoid which will penetrate the feed particles and degrade plant cell walls physically and chemically. In addition, fungi can produce fiber degrading enzymes such as cellulase, hemicellulase, pectinase, and lignocellulase which can increase feed digestibility. However, in Indonesia there are not many studies of the potential for rumen anaerobic fungi, so this review paper aims to discuss the potential of anaerobic fungi rumen in improving fiber digestibility in livestock.

Key words: anaerobic fungi, fiber degradation, lignocellulase enzyme, rumen

ABSTRAK

Hijauan merupakan sumber makanan untuk ternak ruminansia, namun salah satu faktor pembatas dari hijauan adalah tingginya kandungan serat terutama pada hijauan yang sudah tua. Kandungan serat pada pakan hanya bisa didegradasi oleh mikroorganisme rumen. Salah satu mikroorganisme rumen yang memiliki aktivitas pendegradasi serat adalah fungi anaerob rumen karena dapat memproduksi enzim yang sangat aktif untuk mendegradasi lignosellulosa. Fungi anaerob rumen ini dibagi menjadi beberapa genus yang dikelompokkan berdasarkan jumlah *flagela* pada zoospora, morfologi *thallus* dan tipe *rizoid*. Keberadaan fungi di dalam rumen sangat penting karena dari siklus hidupnya fungi dapat membentuk *rizoid* yang akan menembus partikel pakan dan mendegradasi dinding sel tanaman baik secara fisik maupun kimia. Selain itu fungi juga dapat memproduksi enzim pendegradasi serat seperti selulase, hemiselulase, pektin, dan lignoselulase yang dapat meningkatkan kecernaan pakan. Namun di Indonesia kajian mengenai potensi fungi anaerob rumen belum banyak dilakukan sehingga penyusunan makalah ini bertujuan untuk membahas potensi fungi anaerob dalam meningkatkan kecernaan serat pada ternak.

Kata kunci: degradasi serat, enzim lignoselulase, fungi anaerob, rumen

INTRODUCTION

Forage is the main feed for ruminants. One of the problems in developing ruminant business is the low of feed quality, especially in the dry season. According to Lado and Aoetpah, (2009), in general, forage quality on the dry season has already exceeded the flowering phase so it has a very high fiber content. The high fiber content in feed can reduce digestibility and it will affect livestock productivity. Krisnan *et al.* (2009) stated that the productivity of ruminants is determined by rumen performance to digest fiber in feed. Rumen microbes are one of the factor that might affect the performance of livestock in digesting fiber especially due to the presence of cellulolytic microorganisms such as bacteria and fungi.

Pamungkas and Anggraeny, (2006) said that cellulolytic microorganisms play an important role in the digestion of high fiber feed, so the weaning ruminants require probiotics to aid the development

of the rumen ecosystem. Probiotics are additive feed derived from living microbes that can improve the digestive function of host animals by manipulating digestive tract microflora to increase livestock productivity (Fuller, 1989). One of the microbes that can be used as a probiotic candidate is rumen anaerobic fungi. Paul *et al.* (2004) stated that rumen anaerobic fungi are indispensable in rumen because fungi can produce highly active enzymes to degrade lignocellulose. In addition, fungi also have the ability to break down and penetrate feed fiber particles with mycelium, so the available surface for the action of other microbes was increase (Paul *et al.*, 2004). Wood *et al.* (1985) reported that *Neocallismatix frontalis* isolated from sheep rumen has high cellulase enzyme activity in dissolving and damaging hydrogen bonds in feed fiber. Lee *et al.* (2000) also reported that rumen anaerobic fungi isolated from sheep were able to increase lignocellulose consumption. The result of in vitro experiment

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conducted by Paul *et al.* (2004) also showed that anaerobic fungi isolated from buffalo rumen fluid could increase the degradation of feed lignocellulose. However, in Indonesia, there are not many studies of the potential for rumen anaerobic fungi, so this review paper aims to discuss the potential of anaerobic fungi rumen in improving fiber digestibility in livestock.

FUNGAL TAXONOMY

Rumen anaerobic fungi in taxonomy bloom placed into phylum Neocallimastigomycota (Figure 1). It has one order (Neocallimastigales) and one family (Neocallimastigaceae) (Dollhofer *et al.*, 2015). Dollhofer *et al.* (2015) said that rumen anaerobic fungi are divided into eight genus consisting of four monocentric fungi (*Neocallismatix*, *Piromyces*, *Oontomyces*, and *Buwchfawromyces*), two polycentric fungi (*Anaeromyces* and *Orpinomyces*), one monocentric bulbous fungi (*Caecomyces*) and one polycentric bulbous fungi (*Cyllamyces*). However, other genus of new fungi were found along with development research. Hanafy *et al.* (2017) found *Pecoramycetes ruminantium* isolated from cattle and sheep feces. These fungi have one thallus (monocentric) and one flagella (monoflagel) with varying shapes (spherical, ovoid and ellipsoid) (Hanafy *et al.*, 2017). In 2018, Hanafy *et al.*, also found a new genus of rumen anaerobic fungi *Feramyces austini* isolated from wild Barbary sheep dan deer (*Dama dama*). *Feramyces austini* has a monocentric thallus with sperical zoospores ($9.6 \pm 1.9 \mu\text{m}$ in diameter) and 7-16 flagella (polyflagella) (Hanafy *et al.*, 2018). *Liebetanzomyces polymorphus* was found in 2018 by Joshi *et al.*, isolated from the rumen of a goat. Joshi *et al.*, (2018) stated that *Liebetanzomyces* is an anaerobic fungi that produces uniflagellate zoospore with monocentry thallus. These fungi has morphological characteristics similar to *Piromyces*, *Buwchfawromyces*, *Oontomyces* and *Pecoramycetes* but genetically near to *Anaeromyces* (Joshi *et al.*, 2018). Recent study conducted by Hanafy *et al.*, (2020) found seven new genus isolated from five wild (axis deer, white-tailed deer, Boer goat, mouflon, and Nilgiri tahr), one zoo-housed (zebra), and three domesticated (horse, sheep, and goat)

herbivores in US that is *Agriosomyces*, *Aklioshbomyces*, *Capellomyces*, *Ghazallomyces*, *Joblinomyces*, *Khoyollomyces*, and *Tahromyces*.

Nagpal *et al.* (2011) stated that the determination of fungi genus is based on the number of flagella in zoospores, thallus morphology (monocentric and polycentric) and rhizoid type (filamentous or vegetative). Paul *et al.* (2018) stated that the morphological variations in each genus and species anaerobic fungi make the morphological and phenotypic classification of these fungi difficult. Therefore, the determination of anaerobic fungi genus must also be done by molecular analysis. Joshi *et al.* (2018) stated that morphological and molecular identification must be used to determine the specific characteristics of each anaerobic fungal. The internal transcribed spacer (ITS) are the DNA target regions for analyze the diversity of anaerobic fungi (Kittelman *et al.*, 2012; Sirohi *et al.*, 2013; Wang *et al.*, 2017; Edwards *et al.*, 2017). Callaghan *et al.* (2015) and Dagar *et al.* (2015) stated that genetic characterizations of anaerobic fungi could be carried out on ITS1 and ITS2 regions. Paul *et al.*, (2018) and Edwards *et al.* (2019) stated that phylogenetic analysis using ITS1 is more accepted than ITS2 because it is phylogenetically more informative to identifying genus and species level of anaerobic fungi.

The function of anaerobic fungi in the rumen

Aydin *et al.* (2017) reported that fungi have a very important role in degrading plant cell walls because rumen anaerobic fungi such as *Piromyces* has the ability to degrade strong plant cells, *Neocallimastix* has ability to attack or remodel parts of the lignin plant cells by physically or enzymatically, and *Orpinomyces* has ability to dissolve lignocellulose and produce the effective enzyme for hydrolyzing cellulose and hemicellulose. This also agreed by Rabee *et al.* (2018) that fungi have a very significant role to breakdown lignocellulose, so it can be provide a substrate for other microbes. McSweeney *et al.* (1994) reported that fungi have the ability to breakdown lignin bonds so the components of plant cells wall can be dissolved and digested. Bonerman *et al.* (1992) said that anaerobic fungi have the ability to damage the

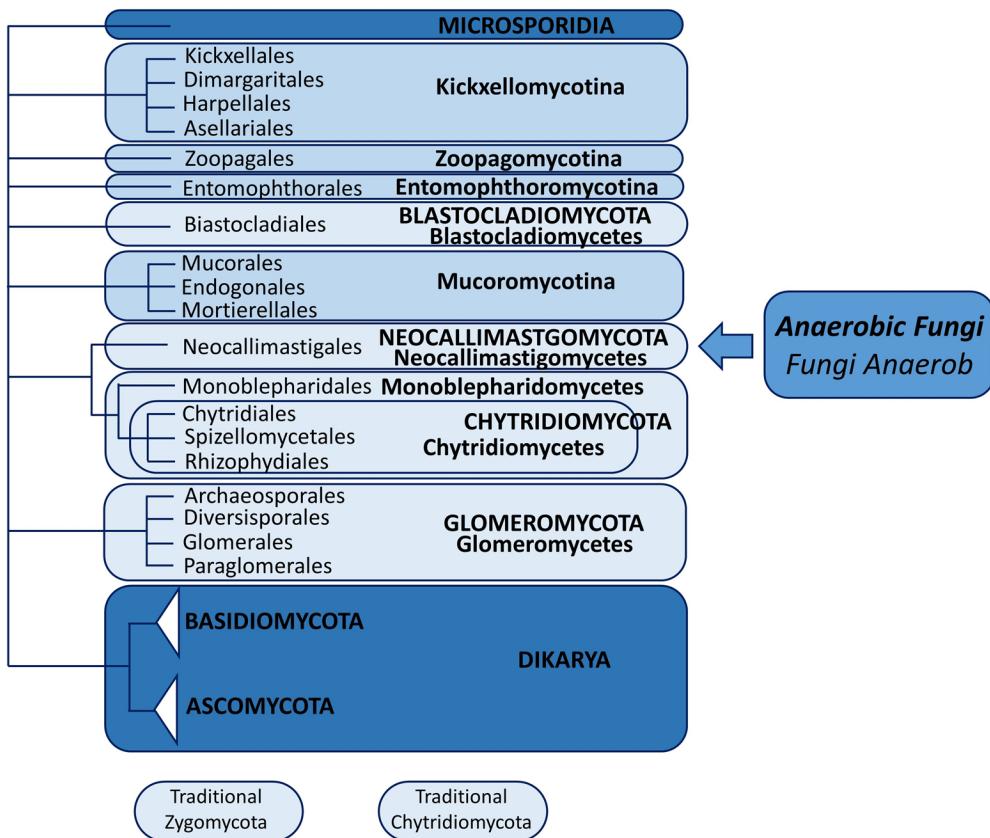


Figure 1. Phylogeny and classification of fungi (Adopted from Hibbet et al., 2007). (*Filogeni dan klasifikasi fungi*) (Diadopsi dari Hibbet et al., 2007).

connecting bond of lignin and hemicelulose, because fungi produce feruloyl and p-coumaroyl esterase. Kittelman et al. (2012) stated that the presence of fungi in the rumen could improve the performance of animal host because the result of cells wall fermentation such as hemicellulose and cellulose will produce VFA (volatile fatty acid) which can be used by the host as an energy source.

Life cycle of rumen anaerobic fungi

The life cycle of rumen anaerobic fungi starts from the production of zoospores as asexual reproduction released from sporangium. Orpin, (1975) stated that the life cycle of rumen anaerobic fungi began when motile zoospores were released from sporangium. The new zoospores will move and search feed sources, especially new feeds to colonize and produce monocentric or polycentric thallus (Haitjema et al., 2014). Ho and Abdullah, (1999)

stated that the time of attaching flagella on feed tissue occurs very quickly ie 15-30 minutes after feed enters the rumen. In all monocentric such as *Piromyces*, *Neocallimastix* and *Buwchfawromyces* the nucleus will be concentrated in sporangium (Haitjema et al., 2014) so the released process of zoospores can cause vegetative thallus ruptured and fungi cannot develop further, it just can enlarge only (Dollhofer et al., 2015).

On the other hand, Haitjema et al. (2014) stated that fungal polycentric such as *Anaeromyces* and *Orpinomyces*, its nucleus is distributed in all zoospores and rhizomycelium. Polycentric fungi can still develop by rhizoid fragmentation when the sporangium breaks (Haitjema et al., 2014). Zoospores that are released from sporangium, will form rhizoid and enter the plant tissue through the enzymatic reaction and hydrostatic pressure

(Dollhofer *et al.*, 2015). Gordon and Phillips, (1998) said that the attached process of fungi and feed begin with the attraction of zoospores to new feed particles. Flagel zoospores will move chemotactically to search carbohydrate receptors in feed such as glucose, mannose, sorbitol and sucrose (Nagpal *et al.*, 2011). Ho and Abdullah, (1999) stated that zoospores in *N. frontalis*, *Piromyces communis*, *Orpinomyces joyonii* and *Anaeromyces* sp. are doing chemotaxis to respond the presence of phenolic acids such as ρ -coumaric acid, ferulic acid and syringic acid found in plant lignin tissue. After the fungi enter the plant tissue, it will develop by enlarging the sporangium and mature sporangium will be released zoospores to search new substrates as fungi growth media like presented in Figure 2. Bauchop, (1989) reported that fungi colonies could form 15 minutes after fungi attach to substrate and after 2 – 3 hours the fungi would form larger rhizoid. Dollhofer *et al.*, (2015) stated that mature sporangium will produce zoospores eight hours after attaching the plant tissue and its entire life cycle will be completed in 24 – 32 hours.

Enzymes produced by rumen anaerobic fungi

Cheng *et al.*, (2018) stated that fungi have the potential to digest fiber because fungi can produce cellulase, hemicellulase, pectinase, and lignocellulase. The experiment of Akin and Borneman, (1990) showed that all anaerobic fungi in the rumen have enzymes exoglucanase, endoglucanase, β -glucosidase, xylanase, dan xylosidase with different enzyme activities in each type of fungi. The result of Morrison *et al.*, (2016) showed that rumen anaerobic fungi type *Orpinomyces* sp. strains C1A were isolated from Angus cattle feces have several high activity enzymes such as β -glucosidase, β -galactosidase and β -xylosidase and low activity xylanase enzyme. Ljungdahl, (2008) carry out more study to identify the detail of *Orpinomyces* enzymes, it showed that this type of fungi had 17 cellulase enzyme consisting of 10 GH family cellulase enzymes 6 (CelA, CelC, CelD, CelF, CelH and CelI), 4 enzymes cellulase family 5 (CelB, CelE, CelG, and CelJ), one enzyme β -glucosidase (Bg1A) and five hemicellulase enzymes are consisting of Xylanase XynA GH11,

lichenase or β -glucanase LicA GH16, mannase ManA GH5, acetyl xylan esterase AxeA, and feruloyl esterase FaeA.

Cellulose degradation mechanism by rumen anaerobic fungi

Cellulose is a polysaccharide consisting of several glucose units with β -1,4-glycosidic bonds (Behera *et al.*, 2017). Cellulose is the most commonly component found in plant cell walls as a single form or binds with hemicellulose and lignin (Behera *et al.*, 2017). It causes the presence of cellulase enzyme in the rumen is very important to degraded cellulose in cell walls. Paul *et al.*, (2004) reported that rumen anaerobic fungi have the ability to produce cellulase enzymes to digest feed fiber. Aydin *et al.* (2017) also stated that *Neocallimastix*, *Piromyces* and *Orpinomyces* have genes that can produce endoglucanase and exoglucanase enzymes. Behera *et al.* (2017) stated that cellulase (endoglucanase, exoglucanase, and cellobiase) are very important enzymes for hydrolyzing β -1,4-glucosidase bonds.

The first enzyme in degrading cellulose is endoglucanase or endo-1,4- β -D-glucanase (EG) (Behera *et al.*, 2017). This enzyme will attack the amorphous part of cellulose randomly and the internal glycan bonds to produce reducing or non-reducing cellooligosaccharides (Behera *et al.*, 2017). After that, exoglucanase or cellobiohydrolase (CBH) will cut reducing sugar side on cellooligosaccharide so the amount of glucose from cellooligosaccharide will be decreased as Figure 3 (Behera *et al.*, 2017). Cellobiose formed in the breakdown process of cellooligosaccharide will be altered by β -glucosidase or β -D-glucosideglucanohydrolase to glucose (Behera *et al.*, 2017).

The effect of rumen anaerobic fungi on in vitro fiber

The presence of anaerobic fungi in the rumen is very important in fiber digestibility. Wood *et al.*, (1986) stated that fungi have strong cellulase enzymes so the presence of fungi in the rumen can improve the digestibility of fiber. This is agreed with

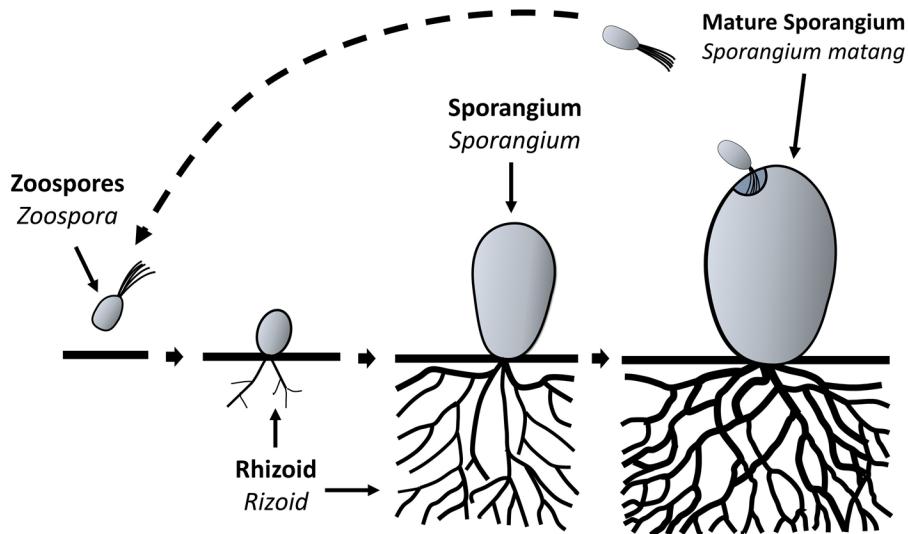


Figure 2. Life cycle of rumen anaerobic fungi (Adopted from Bauchop, 1989). (*Siklus hidup fungi anaerob rumen*) (Diadopsi dari Bauchop, 1989).

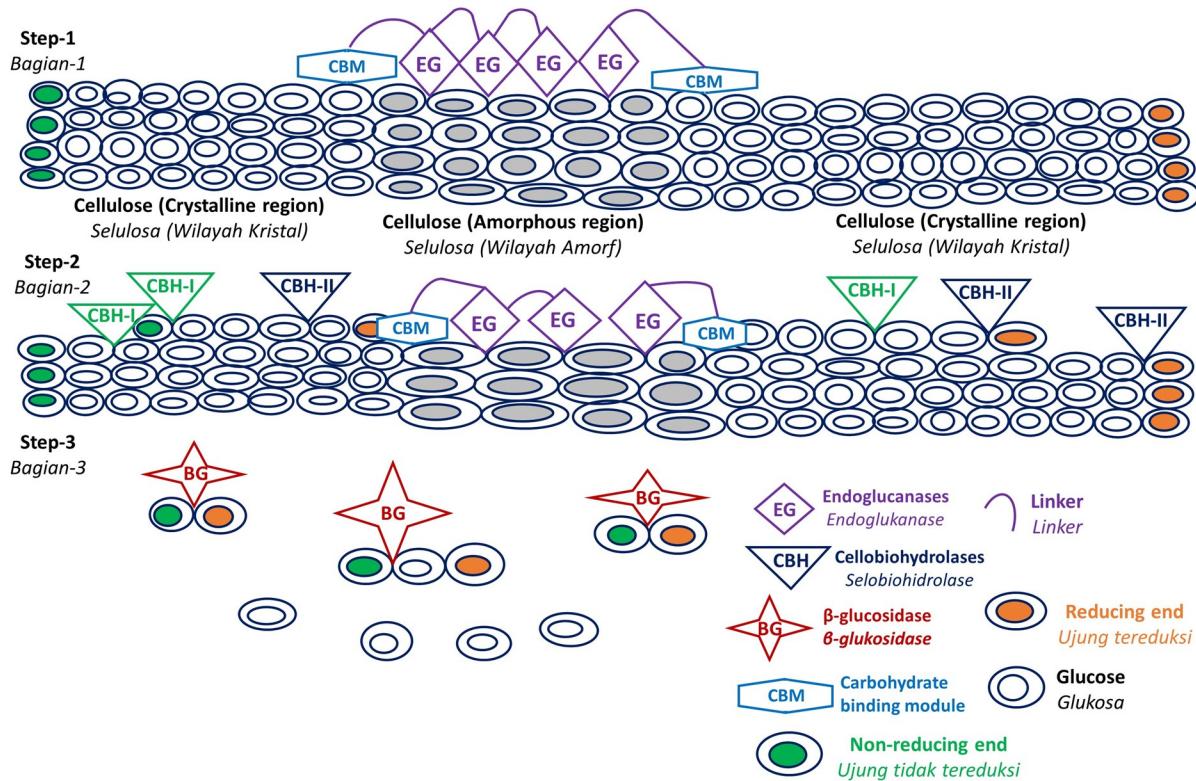


Figure 3. The mechanism of cellulose breakdown by rumen anaerobic fungi (Adopted from Behera et al., 2017). (*Mekanisme perombakan selulosa oleh fungi anaerob rumen*) (Diadopsi dari Behera et al., 2017).

Akin and Bonerman, (1990) study which showed that alfalfa and Bermuda grass that was given fungi isolates had percentage dry weight loss (40.8% and 53.8%) higher than alfalfa and bermuda grass that was given with bacteria isolates (35.7% and 46.4%). The in vitro study of Nagpal *et al.* (2011) showed that straw supplemented with *Caeomyces* sp., *Orpinomyces* sp., and *Necallimastix* sp. incubated for 72 hours had a higher dry matter digestibility (48.9%, 45.8% and 40.6%) compared with straw without anaerobic fungi supplementation (38.6%). Paul *et al.*, (2010) showed that wheat straw added with *Pyromyces* sp. isolated from wild cattle and blue bull had a higher NDF digestibility i.e., 51.5% and 34.3 respectively compared with a wheat straw without fungi addition that only had 26.9% NDF digestibility. Thareja *et al.*, (2006) also showed that after 72 hours incubation straw with the addition of *Anaeromyces* sp. and *Neocaliismatix* had in vitro dry matter digestibility (IVDMD) higher ($30.7 \pm 0.2\%$ and $34.4 \pm 0.1\%$) than straw without addition of fungi ($25.0 \pm 0.2\%$).

Effect of feed on rumen anaerobic fungi populations

Feed Composition. Orpin, (1994) and Bauchop, (1979) reported that differences in the feed would affect anaerobic fungi populations in the digestive tract because fiber and lignocellulose content in feed is a critical factor for the growth of anaerobic fungi. Kostyukovsky *et al.*, (1991) found that cattle were fed with hay had a high number of anaerobic fungi populations and Gordon, (1985) stated that cattle were fed with high concentrate had a low number of fungi populations, because concentrate contains easily fermented carbohydrates so that can reduce the rumen pH. Gordon and Phillips, (1998) stated that a decrease in pH rumen could affect the fungal population. The study of Bauchop, (1979) showed that sporangia or zoospores of *Phycomycetous* fungi were not found in sheep fed with low fiber.

Sulfur. Gordon, (1985) said that in the pasture, sulfur in hay or forage is very influential on rumen

anaerobic fungi populations. Akin *et al.*, (1983) also stated that the number of anaerobic fungi populations in the rumen would increase after the pasture was given S fertilizer. Gordon and Phillips, (1998) also found that anaerobic fungi would not be found in the rumen if ruminants fed forage with low S mineral content.

The potential of anaerobic fungi as a probiotic

Paul *et al.*, (2011) stated that anaerobic fungi have a very high potential to be used as a feed additive because it can improve the growth performance of livestock fed with straw based diet. Anaerobic fungi are significantly contribute to digest fiber content in feed (Gordon *et al.*, 2000). Joblin and Naylor (2010) stated that increased the anaerobic fungi in rumen potentially increase the ruminant productions. This also agreed with the statement of Puniya *et al.* (2015) that the anaerobic fungi have a positive relationship with the increased digestibility of fibrous feed. Gruninger *et al.* (2014) said that the addition of anaerobic fungi could increase in vivo digestibility because it affects the rumen fermentation, populations of rumen microbial and activity of cellulolytic enzyme. In 2004, Dey *et al.*, study showed that the addition of 160 ml fungal culture (*Orpinomyces* sp.) weekly increase feed conversion ratio, average daily gain, digestibility energy and crude fiber digestibility in calves. Sehgal *et al.* (2008) also showed that the supplementation of 250 ml broth culture *Neocallimastix* sp. GR1 every four days in female Murrah buffalo calves increase daily weight gains, feed efficiency and nutrients digestibility (dry matter, crude protein, NDF and energy). Other research that showed the potential of anaerobic fungi as probiotics for livestock was carried out by Paul *et al.* (2011). Its showed that the supplementation of *Neocallimastix* in buffalo calves were given every week for four weeks had weight gain, organic matter digestibility, total digestible nutrient, VFA production higher than buffalo calves without anaerobic fungi supplementation (Paul *et al.*, 2011). Saxena *et al.* (2010) reported that the lactating buffalo were administered with 250 ml anaerobic fungi genus *Orpinomyces* sp. C-14 or *Piromyces* sp. WNG-12

had milk production and milk fat content higher than control (without anaerobic fungi addition).

CONCLUSION

Based on their ability to digest fiber, rumen anaerobic fungi can potentially be used as probiotics for livestock. It is expected to improve the digestibility and productivity of livestock especially in animals fed with high fiber. Several studies showed that addition of anaerobic fungi could improve in vitro dry matter digestibility (IVDMD) and fiber digestibility. However, the presence of fungi in the rumen was influenced by the composition of feed so it needs to be considered to maximize the potential of rumen anaerobic fungi.

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Pedoman Penulisan Naskah Berita Biologi

Berita Biologi adalah jurnal yang menerbitkan artikel kemajuan penelitian di bidang biologi dan ilmu-ilmu terkait di Indonesia. Berita Biologi memuat karya tulis ilmiah asli berupa makalah hasil penelitian, komunikasi pendek dan tinjauan kembali yang belum pernah diterbitkan atau tidak sedang dikirim ke media lain. Masalah yang diliput harus menampilkan aspek atau informasi baru.

Tipe naskah

1. Makalah lengkap hasil penelitian (*original paper*)

Naskah merupakan hasil penelitian sendiri yang mengangkat topik yang *up to date*. Tidak lebih dari 15 halaman termasuk tabel dan gambar. Pencantuman lampiran seperlunya, namun redaksi berhak mengurangi atau meniadakan lampiran.

2. Komunikasi pendek (*short communication*)

Komunikasi pendek merupakan makalah hasil penelitian yang ingin dipublikasikan secara cepat karena hasil temuan yang menarik, spesifik dan atau baru, agar dapat segera diketahui oleh umum. Hasil dan pembahasan dapat digabung.

3. Tinjauan kembali (*review*)

Tinjauan kembali merupakan rangkuman tinjauan ilmiah yang sistematis-kritis secara ringkas namun mendalam terhadap topik penelitian tertentu. Hal yang ditinjau meliputi segala sesuatu yang relevan terhadap topik tinjauan yang memberikan gambaran '*state of the art*', meliputi temuan awal, kemajuan hingga issue terkini, termasuk perdebatan dan kesenjangan yang ada dalam topik yang dibahas. Tinjauan ulang ini harus merangkum minimal 30 artikel.

Struktur naskah

1. Bahasa

Bahasa yang digunakan adalah Bahasa Indonesia atau Inggris yang baik dan benar.

2. Judul

Judul diberikan dalam bahasa Indonesia dan Inggris. Judul ditulis dalam huruf tegak kecuali untuk nama ilmiah yang menggunakan bahasa latin, Judul harus singkat, jelas dan mencerminkan isi naskah dengan diikuti oleh nama serta alamat surat menyurat penulis dan alamat email. Nama penulis untuk korespondensi diberi tanda amplop cetak atas (*superscript*). Jika penulis lebih dari satu orang bagi pejabat fungsional penelitian, pengembangan agar menentukan status sebagai kontributor utama melalui penandaan simbol dan keterangan sebagai kontributor utama dicatatkan kaki di halaman pertama artikel.

3. Abstrak

Abstrak dibuat dalam dua bahasa, bahasa Indonesia dan Inggris. Abstrak memuat secara singkat tentang latar belakang, tujuan, metode, hasil yang signifikan, kesimpulan dan implikasi hasil penelitian. Abstrak berisi maksimum 200 kata, spasi tunggal. Di bawah abstrak dicantumkan kata kunci yang terdiri atas maksimum enam kata, dimana kata pertama adalah yang terpenting. Abstrak dalam Bahasa Inggris merupakan terjemahan dari Bahasa Indonesia. Editor berhak untuk mengedit abstrak demi alasan kejelasan isi abstrak.

4. Pendahuluan

Pendahuluan berisi latar belakang, permasalahan dan tujuan penelitian. Perlu disebutkan juga studi terdahulu yang pernah dilakukan terkait dengan penelitian yang dilakukan.

5. Bahan dan cara kerja

Bahan dan cara kerja berisi informasi mengenai metode yang digunakan dalam penelitian. Pada bagian ini boleh dibuat sub-judul yang sesuai dengan tahapan penelitian. Metoda harus dipaparkan dengan jelas sesuai dengan standar topik penelitian dan dapat diulang oleh peneliti lain. Apabila metoda yang digunakan adalah metoda yang sudah baku cukup ditulis sitasinya dan apabila ada modifikasi maka harus dituliskan dengan jelas bagian mana dan hal apa yang dimodifikasi.

6. Hasil

Hasil memuat data ataupun informasi utama yang diperoleh berdasarkan metoda yang digunakan. Apabila ingin mengacu pada suatu tabel/ grafik/diagram atau gambar, maka hasil yang terdapat pada bagian tersebut dapat diuraikan dengan jelas dengan tidak menggunakan kalimat 'Lihat Tabel 1'. Apabila menggunakan nilai rata-rata maka harus menyertakan pula standar deviasinya.

7. Pembahasan

Pembahasan bukan merupakan pengulangan dari hasil. Pembahasan mengungkap alasan didapatkannya hasil dan arti atau makna dari hasil yang didapat tersebut. Bila memungkinkan, hasil penelitian ini dapat dibandingkan dengan studi terdahulu.

8. Kesimpulan

Kesimpulan berisi infomasi yang menyimpulkan hasil penelitian, sesuai dengan tujuan penelitian, implikasi dari hasil penelitian dan penelitian berikutnya yang bisa dilakukan.

9. Ucapan terima kasih

Bagian ini berisi ucapan terima kasih kepada suatu instansi jika penelitian ini didanai atau didukungan oleh instansi tersebut, ataupun kepada pihak yang membantu langsung penelitian atau penulisan artikel ini.

10. Daftar pustaka

Tidak diperkenankan untuk mensitis artikel yang tidak melalui proses *peer review*. Apabila harus menyitir dari "laporan" atau "komunikasi personal" dituliskan '*unpublished*' dan tidak perlu ditampilkan di daftar pustaka. Daftar pustaka harus berisi informasi yang *up to date* yang sebagian besar berasal dari *original papers* dan penulisan terbitan berkala ilmiah (nama jurnal) tidak disingkat.

Format naskah

1. Naskah diketik dengan menggunakan program Microsoft Word, huruf New Times Roman ukuran 12, spasi ganda kecuali Abstrak spasi tunggal. Batas kiri-kanan atas-bawah masing-masing 2,5 cm. Maksimum isi naskah 15 halaman termasuk ilustrasi dan tabel.

2. Penulisan bilangan pecahan dengan koma mengikuti bahasa yang ditulis menggunakan dua angka desimal di belakang koma. Apabila menggunakan Bahasa Indonesia, angka desimal ditulis dengan menggunakan koma (,) dan ditulis dengan menggunakan titik (.) bila menggunakan bahasa Inggris. Contoh: Panjang buku adalah 2,5 cm. Length of the book is 2.5 cm. Penulisan angka 1-9 ditulis dalam kata kecuali bila bilangan satuan ukur, sedangkan angka 10 dan seterusnya ditulis dengan angka. Contoh lima orang siswa, panjang buku 5 cm.

3. Penulisan satuan mengikuti aturan *international system of units*.

4. Nama takson dan kategori taksonomi ditulis dengan merujuk kepada aturan standar yang diajui. Untuk tumbuhan menggunakan *International Code of Botanical Nomenclature* (ICBN), untuk hewan menggunakan *International Code of Zoological Nomenclature* (ICZN), untuk jamur *International Code of Nomenclature for Algae, Fungi and Plant* (ICAFP), *International Code of Nomenclature of Bacteria* (ICNB), dan untuk organisme yang lain merujuk pada kesepakatan Internasional. Penulisan nama takson lengkap dengan nama author hanya dilakukan pada bagian deskripsi takson, misalnya pada naskah taksonomi. Penulisan nama takson untuk bidang lainnya tidak perlu menggunakan nama author.

5. Tata nama di bidang genetika dan kimia merujuk kepada aturan baku terbaru yang berlaku.

6. Untuk range angka menggunakan en dash (-), contohnya pp.1565–1569, jumlah anakan berkisar 7–8 ekor. Untuk penggabungan kata menggunakan hyphen (-), contohnya: masing-masing.

7. Ilustrasi dapat berupa foto (hitam putih atau berwarna) atau gambar tangan (*line drawing*).

8. Tabel

Tabel diberi judul yang singkat dan jelas, spasi tunggal dalam bahasa Indonesia dan Inggris, sehingga Tabel dapat berdiri sendiri. Tabel diberi nomor urut sesuai dengan keterangan dalam teks. Keterangan Tabel diletakkan di bawah Tabel. Tabel tidak dibuat tertutup dengan garis vertikal, hanya menggunakan garis horizontal yang memisahkan judul dan batas bawah.

8. Gambar
Gambar bisa berupa foto, grafik, diagram dan peta. Judul gambar ditulis secara singkat dan jelas, spasi tunggal. Keterangan yang menyertai gambar harus dapat berdiri sendiri, ditulis dalam bahasa Indonesia dan Inggris. Gambar dikirim dalam bentuk .jpeg dengan resolusi minimal 300 dpi, untuk *line drawing* minimal 600dpi.
9. Daftar Pustaka
Situs dalam naskah adalah nama penulis dan tahun. Bila penulis lebih dari satu menggunakan kata ‘dan’ atau *et al.* Contoh: (Kramer, 1983), (Hamzah dan Yusuf, 1995), (Premachandra *et al.*, 1992). Bila naskah ditulis dalam bahasa Inggris yang menggunakan sitasi 2 orang penulis maka digunakan kata ‘and’. Contoh: (Hamzah and Yusuf, 1995). Jika sitasi beruntun maka dimulai dari tahun yang paling tua, jika tahun sama maka dari nama penulis sesuai urutan abjad. Contoh: (Anderson, 2000; Agusta *et al.*, 2005; Danar, 2005). Penulisan daftar pustaka, sebagai berikut:
 - a. **Jurnal**
Nama jurnal ditulis lengkap.
Agusta, A., Maehara, S., Ōhashi, K., Simanjuntak, P. and Shibuya, H., 2005. Stereoselective oxidation at C-4 of flavans by the endophytic fungus *Diaporthe* sp. isolated from a tea plant. *Chemical and Pharmaceutical Bulletin*, 53(12), pp.1565–1569.
 - b. **Buku**
Anderson, R.C. 2000. *Nematode Parasites of Vertebrates, Their Development and Transmission*. 2nd ed. CABI Publishing. New York. pp. 650.
 - c. **Prosiding atau hasil Simposium/Seminar/Lokakarya.**
Kurata, H., El-Samad, H., Yi, T.M., Khammash, M. and Doyle, J., 2001. Feedback Regulation of the Heat Shock Response in *Escherichia coli*. *Proceedings of the 40th IEEE Conference on Decision and Control*. Orlando, USA pp. 837–842.
 - d. **Makalah sebagai bagian dari buku**
Sausan, D., 2014. Keanekaragaman Jamur di Hutan Kabungolor, Tau Lumbis Kabupaten Nunukan, Kalimantan Utara. Dalam: Irham, M. & Dewi, K. eds. *Keanekaragaman Hayati di Beranda Negeri*. pp. 47–58. PT. Eaststar Adhi Citra. Jakarta.
 - e. **Thesis, skripsi dan disertasi**
Sundari, S., 2012. Soil Respiration and Dissolved Organic Carbon Efflux in Tropical Peatlands. *Dissertation*. Graduate School of Agriculture. Hokkaido University. Sapporo. Japan.
 - f. **Artikel online.**
Artikel yang diunduh secara online ditulis dengan mengikuti format yang berlaku untuk jurnal, buku ataupun thesis dengan dilengkapi alamat situs dan waktu mengunduh. Tidak diperkenankan untuk menseptisasi artikel yang tidak melalui proses peer review misalnya laporan perjalanan maupun artikel dari laman web yang tidak bisa dipertangung jawabkan kebenarannya seperti wikipedia.
Himman, L.M., 2002. A Moral Change: Business Ethics After Enron. San Diego University Publication. <http://ethics.sandiego.edu/LMH/oped/Enron/index.asp>. (accessed 27 Januari 2008) bila naskah ditulis dalam bahasa inggris atau (diakses 27 Januari 2008) bila naskah ditulis dalam bahasa indonesia

Formulir persetujuan hak alih terbit dan keaslian naskah

Setiap penulis yang mengajukan naskahnya ke redaksi Berita Biologi akan diminta untuk menandatangani lembar persetujuan yang berisi hak alih terbit naskah termasuk hak untuk memperbaiknya melalui artikel dalam berbagai bentuk kepada penerbit Berita Biologi. Sedangkan penulis tetap berhak untuk menyebarluaskan edisi cetak dan elektronik untuk kepentingan penelitian dan pendidikan. Formulir itu juga berisi pernyataan keaslian naskah yang menyebutkan bahwa naskah adalah hasil penelitian asli, belum pernah dan tidak sedang diterbitkan di tempat lain serta bebas dari konflik kepentingan.

Penelitian yang melibatkan hewan dan manusia

Setiap naskah yang penelitiannya melibatkan hewan (terutama mamalia) dan manusia sebagai obyek percobaan/penelitian, wajib menyertakan ‘ethical clearance approval’ yang dikeluarkan oleh badan atau pihak berwenang.

Lembar ilustrasi sampul

Gambar ilustrasi yang terdapat di sampul jurnal Berita Biologi berasal dari salah satu naskah yang dipublikasi pada edisi tersebut. Oleh karena itu, setiap naskah yang ada ilustrasinya diharapkan dapat mengirimkan ilustrasi atau foto dengan kualitas gambar yang baik dengan disertai keterangan singkat ilustrasi atau foto dan nama pembuat ilustrasi atau pembuat foto.

Proofs

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