Van soest and mineral analysis of wild buffaloes feces 
(Bubalus bubalis L.) in Savana Baluran National Park, East Java Indonesia

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Abstract. Climate change has affected livestock production systems through impair feed intake, metabolic activities and defense mechanisms. Furthermore, it is also caused the greatest reduction of herbage yield and increase lignifications in plant tissues and hence decrease the digestibility of forage by the animal. The research has been done in Savana Bekol, Baluran National Park, East Java. The research was conducted in June 2019. Objectives: To know Van Soest analysis of wild buffaloes feces sample, so we know the type of plant that consumed. Methods: Observations were carried out by withdrawal feces sample with purposive sampling. Observation time is carried out for 8 hours (8.00-16.00). Van Soest analysis was conducted at Laboratory of Nutrition Technology of Animal Husbandry Faculty, Bogor Agricultural University. Results: The result for Van Soest analysis were Neutral Detergent Fiber (NDF) (71.60%), hemicellulose (23.19%), Acid Detergent Fiber (ADF) (48.41%), cellulose (2.22%) and lignin (32.91%). Mineral analysis showed that feces of buffaloes consists of Nitrogen Free Extract (NFE) (32.05%), Calcium (Ca) (2.00%), Phosphorus (P) (0.52%), and Sodium Chloride (NaCl) (0.10%). Conclusion: These results indicate the NDF, hemicelluloses, cellulose of wild buffaloes were lower than pet buffaloes, but ADF and lignin of wild buffaloes were higher than pet buffaloes.

1. Introduction
The effect of climate change on buffalo is well known, but much less is known about the effects of climate change on faeces of wild buffalo. The objective of this research was to evaluation the faeces in wild buffalo in Baluran National Park. In order to know the effect of climate change on wild buffalo in National Baluran Park, faeces were brought to analysis the digestibility and balance of nutrients and metabolism.

Global warming is one of the causes of global climate change that can directly or indirectly affect livestock productivity. The direct effect of global warming can occur on the health, growth and reproduction of livestock, while the indirect effect is through the availability of grains for additional feed and on the production and quality of forage. According to [1] crop yields are projected to fall in the tropics and subtropics by 10 to 20% by 2050 due to combination of warming and drying, but in some places yield losses could be more severe. Thus in these areas crop productivity will be a critical
constraint for livestock systems.

Another impact of climate change is expected to heighten the vulnerability of livestock systems and reinforce existing factors that are affecting livestock production systems [2], plants, so as to reduce the biodiversity that exists in Indonesia and also the world. Climate change is one of the characteristics and imbalance of temperature, wind and long-term rainfall from a particular region that affects livestock production in this era, crop-livestock systems, mainly acting on forage availability and quality, animal health and productivity [3]. Climate change is acting as a threat multiplier for the issues normally faced by crop production. The potential impacts on livestock include changes in production and quality of feed crop and forage [4-5].

The indirect effect of changes in environmental temperature on livestock performance mainly results in changes in the quality and quantity of feed buffaloes. Forage is the main source of feed for ruminants, so to increase ruminant buffaloes production must be followed by an increase in supply of sufficient forage both in quantity and quality. Forage quality is estimated by chemical analyses, such as structural carbohydrates (neutral detergent fibre (NDF), which includes cellulose, hemicellulose and lignin as the major components [6] and acid detergent fibre (ADF). Forage digestibility in ruminants is constrained by the extent of cell wall (NDF) digestion [7]. The digestibility of forage and the capacity of ruminants to consume it, are largely influenced by its content of neutral detergent fibre (NDF). The digestible proportion of NDF has the greatest impact of all energy supplying nutrients on energy supply to the ruminant, in forage-based feeding systems [8]. Summer and drought cause the greatest reduction in biomass yield of various types of grass in low-lying environments [9]. Feed buffaloes is a feed ingredient that contains the fiber needed by buffaloes to maintain the normal functioning of the digestive tract.

Trace minerals are required for the normal functioning of basically all biochemical processes in the body. They are part of numerous enzymes and coordinate a great number of biological processes, and consequently they are essential to maintain animal health and productivity [10]. The availability of quality forage is influenced by complex interactions among climate, soil, animals, plants, water, and humans.

2. Experimental methods
The evaluation of feces content of neutral detergent fiber (NDF), acid detergent fiber (ADF) and cellulose were analyzed by the [6] methods. Hemicellulose content was calculated by subtracting ADF from NDF. Nitrogen content in feces was determined by micro-kjeldhal method and CP calculated as Nx 6.25. Mineral extract of feces were brought to the laboratory for further analysis. Van Soest analysis and mineral tests were carried out at the Laboratory of Nutrition Technology of Animal Husbandry Faculty, Bogor Agricultural University in Indonesia. The collected data were tabulated and analyzed descriptively.

3. Methodology
Equipment used for Van Soest analysis includes: a. Analysis of neutral detergent fiber (NDF) and acid detergent fiber (ADF), cellulose, lignin, silica, including: digital scales, cup glasses 600 mL, hot plates, vacuum pumps, oven 105°C, desiccators, G3 glass pots, funnels Buchner, and brace. The materials used for the analysis of NDF and ADF are neutral detergent solution (Neutral Detergent Solution/NDS), hot distilled water and acetone and acid detergent solution (Acid Detergent Solution/ ADS), sulfuric acid solution (H2SO4) 72%, and solution of 72% sulfuric acid HBr 48%.

3.1. Van Soest Analysis [6]
Van Soest analysis is used to determine the value of Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), levels of cellulose, hemicellulose, and lignin feces of wild buffaloes.
3.1.1. Neutral Detergent Fiber (NDF) levels. 1 g feces sample (a) is put into a 600 ml beaker and 100 ml is added to the Neutral Detergent Solution (NDS) and then heated. The sample is extracted for 60 minutes from starting to boil. The extracted sample was filtered using a previously weighed G3 sand glass plate (b). The residue is rinsed using hot water and acetone. The glass plate and the residue are dried in a 105°C oven for ±4 hours until the weight is stable. The sample is removed and cooled in an excicator then the cup is weighed (c). Calculation:

\[
\% \text{NDF} = \frac{c - b}{a} \times 100\%
\]

3.1.2. Levels of Acid Detergent Fiber (ADF). Each sample of 1 gram (A) and put in a cup of 600 ml cup, then added 100 ml of ADS solution. The sample is extracted for 60 minutes from starting to boil. The liquid is filtered using a pre-weighed (B) weighing glass cup. The residue is rinsed using hot water and acetone. The residue is dried in a 105°C oven for ±4 hours until the weight is stable. The cup is removed and cooled in the excicator. After chilling, the cup is weighed (C). Calculation:

\[
\% \text{ADF} = \frac{C - B}{A} \times 100\%
\]

3.1.3. Cellulose content. Cellulose analysis is a continuation of ADF analysis. The ADF analysis sample that had been weighed (C) plus a 72% sulfuric acid solution (H2SO4) until submerged for 3 hours. After 3 hours, the residue was rinsed using hot water and acetone. The residue is dried in a 105°C oven for ±4 hours until the weight is stable, remove and cool in an excicator. After chilling, the cup is removed from the excicator and weighed (D). The amount of cellulose content is calculated using the following equation:

\[
\% \text{Cellulose} = \frac{C - D}{A} \times 100\%
\]

\[
\% \text{Hemicellulose} = \% \text{NDF} - \% \text{ADF}
\]

3.1.4. Lignin levels. Lignin analysis is a continuation of ADF and cellulose analysis. The dried sample (D) is then burned in a furnace with a temperature of ±600°C. Chill the saucer in the excicator and weigh (E). The amount of lignin is calculated using the following equation: Calculation:

\[
\% \text{Lignin} = \frac{C - E}{A} \times 100\%
\]

3.1.5. Silica level. Silica analysis (SiO2) is a continuation of lignin analysis. Lignin residue was added 4 ml of 48% HBr solution and allowed to stand for one hour. After one hour washing the residue using acetone and heated in a furnace with a temperature of ±600°C. Chill the saucer in the explorer and weigh it (F). The amount of silica content is calculated using the following equation: Calculation:

\[
\% \text{Silica} = \frac{F - B}{A} \times 100\%
\]

4. Results and discussion

4.1. Results

Fiber fraction was measured using the method of Van Soest [6]. The measured fiber fraction consists of Neutral detergent fiber (NDF), Acid detergent fiber (ADF), lignin, cellulose and hemicellulose. The results showed that livestock and plants will be highly affected by climate change and biodiversity loss [13], this can be seen in Table 1 below.

4.1.1. Results on Van Soest analysis are presented in table 1. The data in Table 1 indicates that the crude fiber (cellulose, hemicelluloses, and lignin) content of wild buffaloes feces show significant differences, if be compared with pet buffaloes. The low digestible fibre (cellulose, lignin) and high
digestible fibre (hemicellulose) of feces were determined as described by Van Soest [6]. The data in Table 1 shows that the NDF and ADF content of wild buffaloes have similarities with the pet buffaloes. The levels of NDF, ADF and crude fiber and lignin content of feces can be seen in Table 1.

Table 1. Van Soest analysis of feces [14].

| Feces          | NDF (%) | ADF (%) | Hemicellulose (%) | Cellulose (%) | Lignin (%) |
|----------------|---------|---------|-------------------|---------------|------------|
| Wild buffaloes | 71.60   | 48.41   | 23.19             | 2.22          | 32.91      |
| Pet buffaloes* | 82.16   | 33.33   | 48.83             | 22.28         | 12.39      |

NDF (Neutral Detergent Fiber), ADF (Acid Detergent Fiber)

Based on Table 1 above shows that the content of NDF, hemicelluloses, cellulose, were lower than buffaloes pet feces, but the ADF and lignin were higher than buffaloes pet feces. The high lignin content in wild buffalo’s forage feed causes the lignin content in wild buffaloes feces to be increase, so that the digestibility of ADF could not be digested.

NDF and ADF have lignose cellulose bonds that are difficult to digest by buffaloes. The content of NDF and ADF is low, so the level of digestibility is increasing and this showed that the quality of feed is getting better. Decreased levels of NDF and ADF will affect the levels of crude fiber. NDF and ADF levels decrease, crude fiber levels will also decrease and crude protein levels increase so that it will affect the quality of feed [15]. In addition, according to [16] stated that the level of plant components that could not be digested (lignin and silica) is part of the ADF. It can cause low digestibility.

Cellulose and hemicellulose are the main components of cell walls in a large quantity in the plants. Cellulose and lignin is one of the criteria that demonstrate the power of fiber. Cellulose consist of highly ordered areas in fibres that retard hydrolysis. Enzymatic hydrolysis of cellulose is a process that requires physical contact between glycoside hydrolase and its substrate, which is inhibited by lignin in various ways [17].

The lignin is always bound to holocellulose (a combination of cellulose and hemicellulose). Structurally, lignin is fenol propane and cannot be digested by rumen microorganisms. Not only that, lignin also decreases the digestibility of other components. Therefore, forage expected to be provided to livestock at a low lignin level in order not to inhibit the action of rumen microorganisms [18].

4.1.2. Results on nitrogen (N), calcium (Ca), phosphorus (P) of feces are presented in table 2. The results in Table 2 revealed that NFE no significant difference in wild buffaloes if compared with pet buffaloes. But Ca, P and NaCl content of wild buffaloes showed the contrary with the pet buffaloes. Ca, P and NaCl level of wild buffaloes were very low when compared to pet buffaloes.

Table 2. The identified mineral test from feces of wild buffaloes and pet buffaloes.

| Feces          | NFE (%) | Ca (%) | P (%) | NaCl (%) |
|----------------|---------|--------|-------|----------|
| Wild buffaloes | 32.05   | 2.00   | 0.52  | 0.10     |
| Pet buffaloes  | 32.20   | 28.20  | 6.70  | 1.10     |

NFE (Nitrogen Free Extract), Ca (Calcium), P (Phosphor), NaCl (Natrium Chloride)

The data in Table 2 is also indicate that the mineral of wild buffaloes feces such as Ca, P and NaCl content show significant differences with pet buffaloes.

Calcium has special relevance with regard to the mineral constitution of several types of forages and is used to increase the productivity of ruminant animals. These minerals are generally present in significant concentrations in forage feed.

4.2. Discussion
The climate change will increase vulnerability to livestock feed in terms of quality and quantity [20]. Impacts on forage quantity and quality depend on the region and length of growing season [5]. The
results showed wild buffalo feces lignin content is higher than pet buffalo. According to [20], increased lignification in plant tissue can reduce the digestibility of forage by animals. Similarly, with [21], on evaluation of climate change impact on forage quality indicate that high temperatures tend to increase lignification in plant tissue, but reduce digestibility of forage and this simultaneously induced a shift from C3 grass species to C4 grasses which has direct implications for forage supply.

Neutral-detergent fibre (NDF) consists of cellulose, hemicellulose and lignin, which represent the major cell wall fractions. The cell walls of plants composed of cellulose, hemicellulose, and lignin which are linked each other and as a component of crude fiber. The increased production of crude fiber was also caused by the process of lignifications are increasing. Increased lignin and cellulose causes the rod will be even greater, and growing cambium stem becomes hard and large [18]. Crude fiber fraction influenced digestibility, because only a small fraction of fibers can be digested by microbes.

Lignin is one component of plant cell walls that cannot be digested by ruminant. The content of lignin in ruminant feed will affect the feed digestability. Lignin is one of the limiting factors in forage crops that will affect the digestibility of feed. Low lignin content can still be digested by cattle, while the high lignin content may decrease the digestibility because it is hard to be broken. Additionally, the cellulose, hemicellulose and lignin can also bind to the proteins in the cell walls of plants. Lignin is insoluble in rumen fluid that inhibit the action of rumen microorganisms and enzymes to digest the feed plant [18]. Furthermore, forages of low nutritional value exist which contain high amounts of lignin cellulose, low concentrations of fermentable sugars, and proteins of low quality; these characteristics affect the microbial activity and thus the profile of the fermentation products [22].

According to [23], hemicelluloses, cellulose, and lignin contents of feeds and feces. The cellulose digestibility, as were the correlations between digestibilities of hemicelluloses with total as well as with insoluble noncellulosic polysaccharides. The results showed NDF content of wild buffaloes was lower than pet buffaloes. NDF is determiner major component of the cell wall such as cellulose, hemicellulose and lignin [7]. Lignin is a phenolic compound that can bind cellulose so that cattle cannot digest cellulose. Lignin and silica could not be digested by digestive enzymes in ruminants. Therefore, cellulose and hemicellulose are substances that useful in the NDF.

The lignin content is negatively correlated with digestible energy in ruminant diets. Reduced feed intake: due to the association with polysaccharide constituents, lignin forms a physical barrier and thus hinders the access of rumen microbes to fermentable cell wall components. Consequently, the passage rate of feeds through the rumen is slowed down, thus reducing the feed intake capacity. So there is the optimal limit of lignin that can be tolerated by ruminant livestock, especially buffaloes, if it exceeds the limit it will affect the digestibility of other feed substances [24]. Furthermore, the high lignin content has a low level of digestibility and limits the bioconversion of forages into products from cattle [25].

Quality of feed crops and forage may be affected by increased temperatures and dry conditions due to variations in concentrations of water-soluble carbohydrates and nitrogen. Temperature increases may increase lignin and cell wall components in plants [5] [26], which reduce digestibility and degradation rates [5], leading to a decrease in nutrient availability for livestock [27]. Furthermore, season significantly different affect to organic matter digestibility of herbage [28].

The climate change has also affected the quality of buffalo forage feed in Baluran National Park, so plants suffer from a lack of important nutrients such as Ca, P and NaCl, as a result the buffalo digestibility also decreases. The quality of minerals in forage is low, causing the amount of minerals present in feces to be low. According to [10] the concentrations of trace minerals in plants largely depend on plant genotype, soil environment, climate, and stage of maturity. Some trace mineral concentrations in animal products are related to animal dietary concentration. The homeostatic control of essential trace element flux through the body is mediated by the down-regulation of absorption and/or stimulation of excretion (urine, faecal excretion from endogenous sources) [29].

5. Conclusion
The climate change will affect wild buffaloes feces and consequently feed security, the nutritional content of wild buffaloes feces, which are one of the suppliers of carbohydrate (cellulose) and lignin.
Global warming affects the nutritional quality of wild buffalo feed, thus affecting the wild buffalo feces. The content of ADF and wild buffalo feces lignin in Baluran National Park is higher than pet buffalo. Nevertheless, the NDF, hemicelluloses, cellulose, and wild buffalo feces in Baluran National Park are lower than those of pet buffaloes.

NFE level of wild buffalo is almost the same as pet buffalo, but the levels of Ca, P and NaCl of wild buffalo are very low when compared to pet buffaloes.

Acknowledgment
The authors are thankful to the staff of Laboratory of Nutrition Technology of Animal Husbandry Faculty, Bogor Agricultural University in Indonesia, Universitas Negeri Jakarta and Baluran National Park Indonesia for supported.

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