Provision of *Indigofera zollingeriana* and turmeric (*Curcuma domestica*) in quail rations on in vitro value of dry matter and protein digestibility

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**Abstract.** Animal husbandry development has an important role in the effort to meet the needs of animal protein in the community. Digestion of feed substances is one of the benchmarks in determining the quality of feed ingredients, in addition to their chemical composition. To study digestibility and fermentation in the digestive tract, a very successful and widely used method is the In-vitro technique. Turmeric (*Curcuma domestica*) is a plant that pledged as phytobiotic feed additive group at poultry. *Indigofera zollingeriana* is a legume plant that has the potential as a feed ingredient for protein sources with high protein content along with relatively low fibre content and a high digestibility rate. This plant is very good as a source of forage feed in poultry.

The design used in this study was a completely randomized design (CRD) with 4 treatments and 5 replications. The composition of the research treatment was as follows: without TPDI and turmeric flour (P0), substituting 6% soybean meal protein with 10% TPDI and 2.5% turmeric flour (P1), substituting 9% soybean meal protein with 15% TPDI and 2.5% turmeric flour (P2), 12% substitution of soybean meal protein with 20% TPDI and 2.5% turmeric flour (P3). Changes observed were dry matter digestibility and protein digestibility. The results showed that the substitution of soybean meal protein with *Indigofera zollingeriana* shoot flour at different levels and the addition of turmeric significantly affected dry matter digestibility and protein digestibility (p<0.05). The average digestibility of dry matter P1 gave a significantly lower difference with P2 and P3 (p<0.05) and in P3 protein digestibility the difference was significantly lower with P0 and P1 (p<0.05). This study concludes that the administration of *Indigofera zollingeriana* at the level of 10% and 2.5% turmeric was able to improve dry matter digestibility by 84.96% and protein digestibility by 35.99%.

**1. Introduction**

Digestion of food substances is one of the benchmarks in determining the quality of feed ingredients, in addition to their chemical composition. To study digestibility and fermentation in the digestive tract, a method that is very successful and has been widely used is in-vitro techniques. Measurement of the digestibility of feed ingredients or rations can be done directly on poultry because poultry has very fast growth in a short time so that the optimal absorption of food substances can be seen. Digestion measurement is an attempt to determine the number of substances that can be absorbed by the digestive tract. By measuring the amount of food consumed and the amount of food released through feces.
Indigofera zollingeriana is a type of legume feed and has very high nutrients, can grow in almost all soils in Indonesia, the average tree is not too high but has dense leaves and can produce a lot. Turmeric (Curcuma domestica) as a feed additive can be used as a combination feed in poultry, the active substances possessed by turmeric are curcumin and essential oils that function as cholagogue (can increase the secretion of bile). In addition to essential oil, another content contained in turmeric is curcuminoids which can increase appetite, which in turn will increase body weight. Curcumin is the most dynamic compound that outfits turmeric with its characteristic yellow color and is recognized as being mindful for most of its helpful impacts, counting antibacterial, antifungal, antiprotozoal, antiviral, antioxidant, anti-inflammatory and hypocholesterolemia exercises [1].

Indigofera zollingeriana is a legume plant that has potential as a source of food for protein and high digestibility, this plant is very good as a source of forage in poultry. The addition of turmeric flour in poultry rations can improve the working system of digestive organs that can help the absorption of food in the body. It also serves to increase the body's resistance to livestock. Dry matter digestibility and protein digestibility need to be known to achieve efficient use of feed in a given ration. This cannot be separated from the energy and protein content in the ration which greatly affects the consumption of feed. Factors that meet poultry's need for protein are temperature, environment, age, species/ nation/strain, amino acid content, and digestibility.

Based on the description above, a study was conducted on the effect of giving Indigofera buds (Indigofera zollingeriana) to substitute soybean meal and the addition of turmeric (Curcuma domestica) feed additives in feed on the digestibility of dry matter and protein digestibility.

2. Material and methods

2.1. Material

The materials used in the research of protein digestibility and dry matter are phosphate buffer (0.1 M; pH 6.0), phosphate buffer (0.2 M; pH 6.8) HCl 0.2 M, 1 M HCl, pepsin, 0.5 ml chloramphenicol, 100 ml ethanol, NaOH (0.6 M), 1 M NaOH, EDTA 0.2 M, 30% acetic acid, litmus paper, 96% ethanol, 99.5% acetone, sodium carbonate, aquades, pepsin, basal ration consisting of Indigofera zollingeriana, turmeric flour, corn, bran, soybean meal, fish meal, meat and bone meal (MBM), lysine, methionine. The tools used in protein digestibility research are analytical scales, 125 mm Erlenmeyer, centrifuge tubes, porcelain cups, shake water baths, dropper pipettes, filter paper, crucibles, ovens, desiccators, incubators.

2.2. Methods

2.2.1. Ration composition. The ingredients of the ration consist of ground maize, bran, soybean meal, coconut cake, fish meal, meat and bone meal (MBM), oil, dicalcium phosphate (DCP), lysine, methionine, turmeric flour, and shoot flour of Indigofera zollingeriana. Protein digestibility research is an experimental study using an in vitro method developed [2] that has been given a modification to the administration of pancreatin enzyme into pepsin, this method through three stages, namely: First, the ration sample is weighed (0.500 g ± 1 mg) into the 125 mm Erlenmeyer flask. Subsequently added 25 mL phosphate buffer (0.1 M; pH 6.0) and 10 mL 0.2 M HCl are added to each flask then the flask is shaken until all ingredients are mixed, after that the pH of the solution is adjusted to 2 ± 0.01 by adding 1 M HCl or 1 M NaOH. One millilitre of pepsin solution (25 mg pepsin /mL; P7000, Sigma Aldrich, St. Louis, MO) is added to each flask, and add 0.5 mL of 0.5 g chloramphenicol solution to prevent bacterial growth and samples placed in shake water bath the bath is heated at 39°C for 2 hours. Second, after 2 hours, 10 mL phosphate buffer (0.2 M; pH 6.8) and 5 mL NaOH (0.6 M) were added to each flask and the pH was adjusted to 6.8 ± 0.01 by adding 1 M HCl or 1 M NaOH. One millilitre of 1M HCl solution is added to each flask and the sample is placed in a bath at 39°C for 4 hours. Third, after 4 hours, 10 mL of 0.2 M EDTA solution was added to each flask and the pH was adjusted to 4.8 ± 0.01 with 30% acetic acid. Viskozyme (Viscozyme L V2010, Sigma-Aldrich, St. Louis, MO) was added to each flask with an
amount of 0.5 mL. The pumpkin is placed in a shake water bath with a temperature of 39°C for 18 hours. To determine digestibility, the undigested residue in the flask was filtered on previously weighed and containing 545 seleis (0.400 g ± 5 mg; Sigma Aldrich, St. Louis, MO). The undigested material collected in the crucibles is washed twice with 10 ml of ethanol (96%) and twice with 10 mL acetone (99.5%). The crucibles are dried in an oven for 2 hours at 130°C, cooled in a desiccator, and weighed to measure the total DM (dry matter) residue. DM digestibility in vitro was calculated by a method developed [3].

2.2.2. Experimental design. The design used in this study was a completely randomized design (CRD) with 4 treatments and 5 replications. Protein digestibility test and dry matter uses a combination of Indigofera zollingeriana flour and uses a percentage of turmeric flour addition of 2.5% based on [4]. The composition of the research treatment as follows: P0: Control; P2: Indigofera zollingeriana leaf flour 10% (equivalent to 6% substitution of soybean meal protein) + turmeric rhizome flour 2.5%; P3: Indigofera zollingeriana leaf flour 15% (equivalent to 9% substitution of soybean meal protein) + turmeric rhizome flour 2.5%; P4: Indigofera zollingeriana leaf flour 20% (equivalent to 12% substitution of soybean meal protein) + turmeric rhizome flour 2.5%.

2.2.3. Statistical analysis. The data obtained were processed using variance with a Completely Randomized Design (CRD) with 4 treatments and 5 replications, analysis of the data used Analysis of Variance (ANOVA) following the linear additive of the Complete Randomized Design with the according to model [5].

3. Result and discussion

Based on table 1, the results of the study obtained the average digestibility of dry matter and protein digestibility with the addition of shoot flour Indigofera zollingeriana and turmeric.

| Treatment | DM Digestibility (%) | Protein Digestibility (%) |
|-----------|----------------------|---------------------------|
| P0        | 85.44 ± 0.81         | 32.44 ± 9.41              |
| P1        | 84.96 ± 0.75         | 35.99 ± 1.28              |
| P2        | 86.01 ± 0.11         | 27.05 ± 5.09              |
| P3        | 86.25 ± 0.28         | 23.96 ± 1.69              |

Superscripts with different letters in the same column show a real effect (p<0.05). P0 = control; P1 = Indigofera zollingeriana leaf flour 10% + turmeric rhizome flour 2.5%; P2 = Indigofera zollingeriana leaf top flour 15% + turmeric rhizome flour 2.5%; P3 = Indigofera zollingeriana leaf flour 20% + turmeric rhizome flour 2.5%.

The results of statistical analysis showed that the treatment of soybean meal protein substitution with Indigofera zollingeriana leaf buds at different levels and the addition of turmeric showed a significant effect (p>0.05) on dry matter and protein digestibility.

3.1. Dry matter digestibility

The highest average dry matter digestibility (DM) was obtained in the P3 treatment which was 86.25% and the lowest was in the P1 treatment which was 84.96% (table 1). This shows that the increase in the administration of Indigofera zollingeriana leaf flour to a level of 20% has increased the digestibility of dry matter statistically in the ration so that the flour Indigofera zollingeriana can be used as a substitute.
for soybean meal against the digestibility of dry matter. The high digestibility value of dry matter shows high-quality ration [6].

Duncan's test results showed a difference between the treatments P0 with P1, P2, and P3 (p>0.05) but the P1 gave a significantly lower difference with P2 and P3 (p<0.05). The results of the digestibility of dry matter showed that the digestibility of dry matter tended to increase starting from treatments P1, P2, and P3. One that causes an increase in the digestibility of dry matter is the value of crude fibre in the ration. In this study, the diet experienced an increase in crude fibre P0 (3.32%), P1 (4.40%), P2 (4.75%) and P3 (5.10%) which were able to increase the digestibility of dry matter. This is consistent with the opinion [7] which states that the level of crude fibre ration will affect the digestibility of crude fibre and is directly proportional to the digestibility of dry matter.

The above conditions indicate that curcuminoids can improve the work of organs for digestion of dry matter so that high crude fibre values in rations can be digested properly in the digestive tract of quail. Undigested coarse fibres in poultry can carry digested food substances from ingredients other foods and will be found again in the faeces [8]. According to [9] the digestibility of dry matter will be greatly influenced by the consumption of dry ingredients ration, crude fibre, crude fat, crude protein.

This study showed the high digestibility of dry matter was found in P3 (86.25%), the quality and quantity of dry matter could affect the amount of ration consumption in livestock by in vivo showed results in the administration of Indigofera zollingeriana leaf shoots. with a level of 20% and the addition of turmeric flour 2.5% which is 102 g/e/week. This has decreased the amount of ration consumption from the control ration, but this result is higher than the study of [10] which shows that quails given commercial rations have an average consumption of rations 101.65 g/e/week. The influence of dry matter digestion and ration consumption is explained by [9] digestibility is often closely related to consumption, namely the administration of old forages that are very voluminous and slow to digest compared to plants that are not fibrous.

Morrison [11] explained that the digestibility of dry matter is part of protein digestibility so that when dry matter increases the protein digestibility will also increase. However, [12] explained that the digestibility coefficient or the digestibility level of a ration is influenced by the balance of food content between protein and crude fibre, not merely the eyes saw from digestion, such as the digestibility level of dry matter.

3.2. Protein digestibility

The statistical analysis showed that the administration of Indigofera zollingeriana and turmeric had a significant effect (p<0.05) on protein digestibility in vitro. The average results obtained ranged from 23.96% - 35.99%. This is because Indigofera zollingeriana flour has a high protein composition. One of the factors that influence protein digestibility is the protein content in rations consumed by livestock. Rations with low protein content, generally have low digestibility and vice versa. High or low protein digestibility is influenced by the protein content of the ration material and the amount of protein that enters the digestive tract [9]. Duncan test results obtained that the treatment of P3 experienced significantly lower differences with P0 and P1 (p<0.05) while P0 did not experience significant differences with P1 and P2 (p>0.05). Protein digestibility by giving Indigofera zollingeriana 15% and 20% levels has decreased, this is suspected at the level of 15% and 20% have high crude fibre content. The results of the laboratory analysis of Animal Feed Chemistry show crude fibre content at the level of 15% which is 5.56% and at the level of 20% which is 6.68%.

Protein digestibility on the addition of Indigofera zollingeriana leaf flour with a level of 10% showed the highest digestibility whereas the dry matter digestibility experienced the lowest digestibility with those without the administration of Indigofera zollingeriana leaf flour and turmeric 2.5%. The level of digestibility of food is central to the digestive system of livestock. The consumption of rations on the administration of Indigofera zollingeriana flour with a level of 10% and turmeric 2.5% was able to increase the consumption of rations in quail. Digestion or digestibility is part of the nutrient ration that is not excreted in the phases and is assumed to be absorbed by livestock [13].
Protein digestibility is only around 23.96% - 35.99%, indicating that protein digestibility is low, according to [14] that ration quality based on the level of digestibility there are 3 categories, namely: digestibility value in the range of 50-60% is low quality, between 60-70% of medium quality and above 70% of high quality. The addition of 2.5% turmeric flour in quail ration serves to increase the work of digestive organs that can help the absorption of food in the body but also serves to increase the endurance of livestock. The use of turmeric water extract by 2.5% can improve histomorphology performance and does not have side effects on performance [4]. One source of protein in this study is soybean meal which is thought to be one of the factors affecting the digestibility of proteins that have anti-nutrient trypsin. High or low digestibility value depends on the ingredients of ration and the amount of protein that enters the digestive tract [12].

This situation will certainly also affect the ability of broilers in digesting and absorbing protein [15]. The function of turmeric to improve the work of the digestive organs of poultry is to stimulate the wall of the gallbladder to release bile and to stimulate the release of pancreatic sap containing amylase, lipase, and protease enzymes that are useful for improving digestion of rations such as carbohydrates, fats, and proteins.

4. Conclusion
It can be concluded that the *Indigofera zollingeriana* at the level of 10% and turmeric 2.5% can improve the dry matter digestibility 84.96% and protein digestibility 35.99%.

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