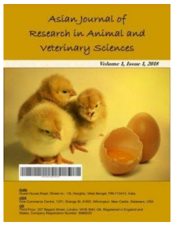

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Comparison of Two Mathematical Models to Describe the Rumen Fermentation Parameters of Some Sources of Plant and Animal Protein Using *in Vitro* Gas Method

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Authors' contributions

This work was carried out in collaboration among all authors. Author EGC designed the study, wrote the protocol, and wrote the first draft of the manuscript. Authors JS and FMAG managed the analyses of the study. Author HA managed the literature searches. Author RS performed the statistical analysis. All authors read and approved the final manuscript.

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ABSTRACT

Aims: To compare, on an experimental basis, the respective relevance of two mathematical models estimating the rumen fermentation parameters of some plant and animal protein sources: the "exponential" model by Ørskov & McDonald (EXP) and the" sigmoid" model by France et al. (FRC).
Study Design: The study was conducted at the University of Ardebil (Iran) between 2014 and 2016. In order to conduct the experimental part of the study, sources of plant protein (soybean meal, rapeseed meal and cottonseed meal) and sources of animal protein (poultry offal meal, fish meal and blood meal) were obtained from the agricultural sector and the local slaughterhouse.

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Methodology: Gas production was measured for 6 feeding contents in 3 repeats at 3 separate periods. The volume of gas produced after 2, 4, 6, 8, 10, 12, 16, 24, 36, 48 and 72 hours incubation was measured and checked against two models estimating gas production parameters and ruminal fermentation kinetics.
Results: The amounts of gas production potential and the rate constant gas production according to both models, EXP and FRC, was not significantly different. However, the two models differ significantly regarding the length of the lag phase (T lag) which is significantly longer in the model EXP, than in the model FRC; due to model EXP substantially overestimating the actual time-lags.
Conclusion: The sigmoid model FRC, proposed by France et al., appears providing more relevant estimates than does the exponential model EXP by Ørskov & McDonald, at least regarding the duration of the lag phase before starting of the fermentation process. Accordingly, it seems that the sigmoid FRC model should be preferred over the exponential EPXP model.

Keywords: In vitro fermentation; mathematical models; protein sources.

1. INTRODUCTION

Gas production *in vitro*, is related to fermentation parameters, and rumen digestion kinetics are valuable descriptions in the evaluation feeds [1]. In this *in vitro* gas production fermentation, a certain amount of feed in the rumen fluid was incubated and the volume of gas produced at regular intervals and row that showed the speed of feed digestion is measured. The results of the tests is described mainly by fitting them into two models of EXP and FRC is done [2]. Therefore, comparing the performance and capability of two models can highly be influential model for choice. Some of the differences between the two models may be related to the test conditions and the type of feed. Some of models, like the model France sigmoid structure have established that due to the use of this structure; the presence of microbial activity in the rumen has been reported [3]. But some other of models like the model of Ørskov and McDonald have non-Sigmoid structure. So today, for greater reliability of gas production test results by the researchers, a variety of models non- Sigmoid and Sigmoid structure is used and in this regard, various formulas have been proposed [4,5]. In most studies related to rumen fermentation parameters by *in vitro* gas production of the exponential equation Ørskov and McDonald [6] as (EXP) y=A (1- e-ct) is used. Ørskov and McDonald model is one of the most well-known models used-in predicting rumen fermentation parameters. This model assumes that the rate of gas production in the rumen depends only on the availability of feed has been reported [7]. Another model that is used to predict gas production, is the model of France (FRC). As mentioned, France model had sigmoid structure and great flexibility in fitting the data of gas production. France model assumes that the rate of gas production is directly linked to the rate of feed degradation and this condition is dependent on fermentation time and time identification or adherence of bacteria to feed components (lag phase) [2]. In addition, there are models that have been proposed by other researchers but have received little attention. However, you need to expantiate on the statement for better understanding of the concept [8]. According to the comprehensive comparison between the two models of France and Ørskov and McDonald for described ruminal fermentation parameters plant and animal some protein sources using gas test method and since the evaluation tests of feed has been done more than alfalfa hay as a standard feed and with important in ruminant nutrition. Therefore, in this study the accuracy of the proposed methods in terms of goodness of fit and to describe the ruminal fermentation parameters in some plant and animal protein sources evaluated using the gas production method.

2. MATERIALS AND METHODS

In order to conduct the experiment, sources of plant protein (soybean meal, Rapeseed meal and cottonseed meal) and sources of animal protein (poultry offal meal, fish meal and blood meal) were obtained from the agricultural sector and the local slaughterhouse. The chemical composition of the feed by conventional methods [9] was carried out. The *in vitro* method [1] was used to measure the amount of produced gas in laboratory conditions and the amount of gas production measured and recorded at 2, 4, 6, 8, 10, 12, 16, 24, 36, 48 and 72 hours of incubation, respectively. In this study, the different mathematical models have been developed to analyze gas production data by two models of digestion by France et al [2] and Ørskov and McDonald [6] with regard to the lag phase was used to evaluate the digestive process. For this purpose of 54 series data obtained from the tests (three separate periods with 3 repeat and 3 levels of feed and 2 feed per period) for fitted data's and T-test was used to compare their means for each parameter of the two models.

Models include:

Ørskov and McDonald model [6] with regard to the lag phase
G=A (1-e^{-ct+L})

Model France et al. [2]

G = A (1-e-c (t-L)-d (√ t -√ L))

Where G is equal to the accumulation of gas produced per unit time, A is equal to the total amount of gas produced (ml), c is equal to a fixed rate of gas production (ml per hour), d is equal to a fixed rate of gas production (ml at ½ h), L equal to the lag phase, t time and t ½ equal to half of the total gas production time is cumulative.

3. RESULTS AND DISCUSSION

3.1 Chemical Composition

The chemical compositions of test feed are presented in Table 1. Blood meal contents have a higher percentage of protein than any of the other plant and animal protein. The maximum amount of crude fat 31.3% for poultry offal meal (POM) and the highest ash content of 20% was observed for fish meal (FM). Highest of NDF and ADF (70.6% and 58.4%) for cotton seed meal (CM) and the lowest NDF and ADF were obtained 45.7 and 33.3% for soybean meal (SM) respectively. The results related to predicted parameters by the model France (FRC) and the Ørskov and McDonald (EXP) are presented in Table 2. As observed the gas production potential (A) for all feed samples testing in the model FRC and EXP respectively, 133.407 and 131.790 ml per gram dry matter was predicted and a significant difference was observed between the two models in terms of gas production potential. The gas production rate constant (c) for all feed tested in the FRC and EXP respectively 0.089 and 0.082 ml per hour, which was not significantly different between the two models.

However, when the individual protein sources were fitted in terms of the two models of France and Ørskov and McDonald, it was observed that rapeseed meal had a significant difference in gas production rate. Only the two models had a significant difference in terms of the lag time (T lag) except for cotton seed meal (P <0.05). According to the results of the tables, T lag was higher in the Ørskov and McDonald's model than the France model. T lag or the time colony production is an important parameter that is associated with feed fibre degradability [10]. There was less time to start the colony by the France model for all plant and animal protein sources. The lag phase for France was 0.44 hours as against 1.96 hours for the Ørskov and McDonald Model observed as shown in Table 2. The longer lag phase for all protein sources in the Ørskov and McDonald model indicated that in this model, microorganisms were observed to have started to recognize and colonize on the digestible substrate in a delayed and time-consuming behaviour compared to the France model.

It is desirable to reduce the production time of the colony for a fermentable substrate and easily fermented, and especially for samples containing fiber and cell wall and certain physicochemical characteristics in the cell wall. Among the studied protein sources, cotton seed meal had the lowest T lag (Table 7) in both models. However, other sources of plant and animal protein in this study, despite their high fibre and cell wall structure (NDF) had less T lag than that of cottonseed meal but the two models in the T lag have shown significantly different values for the protein sources. In this comparison, the France model has the lowest lag phase for either plant or animal protein sources (P <0.05).

This shows that the Ørskov and McDonald model could have an overestimate for lag phase. Therefore, it can be concluded that the French model estimates less lag phase for sources of protein with less fibre. Reis, Sidnei Tavares Dos, et al., [11] stated that the correlation between the cumulative production phase and the total carbohydrate degradation is strong and high, but some differences in this relation could be due to the model used for the analysis.

T Lag represents the amount of time that microbes spent for attachment to raw material or substrate fermentable and adhesion to the insoluble substrate is as a predigesting condition and beginning the process of digestion. The shorter lag phase may be a faster fermentation rate. So among those protein sources, those with a lower lag phase have shown higher fermentation or degradation rates, as well as more gas production. The structure of the

Table 1. Chemical composition of some plant and animal protein sources

Protein sources	DM	CP	EE	Ash	NDF	ADF
Plant						
Soybean meal	92.4	50	1.6	6.1	45.7	33.3
Rapeseed meal	91.4	37	1.2	8	51.5	46.1
Cottonseed meal	93	24	1.4	4.7	70.6	58.4
Animal						
Poultry offal meal	94.4	55	31.3	7.3	48.9	34.8
Fish meal	93.6	50	18.1	20	61.2	40.6
Blood meal	70.6	59	1.6	5	55.3	33.4

**DM = dry matter (percent); CP = crude protein (%DM); EE= crude fat (%DM); Ash = ash (%DM); NDF = Neutral detergent fiber (%); ADF= Acid detergent fiber (%)*

Table 2. Comparison of two models (France and Ørskov and McDonald) based on the estimated parameters between the plant and animal protein sources

		Model		
	Parameters	**France**	**Ørskov and McDonald**	**P value for T-test**
Plant and animal protein sources	A	133.41	131.79	0.93
	c	0.09	0.08	0.59
	T lag	0.44	1.96	<0.001

**A = potential gas production (ml); c = constant rate gas production (ml per hour); T lag = lag phase (hours)*

Table 3. Comparison of two models (France and Ørskov and McDonald) based on the estimated parameters between the plant and animal protein sources

		Model		
	Parameters	**France**	**Ørskov and McDonald**	**P value for T-test**
Plant protein	A	204.74	202.09	0.90
	c	0.06	0.05	0.27
	T lag	0.37	1.48	0.002

**A = potential gas production (ml); c = constant rate gas production (ml per hour); T lag = lag phase (hours)*

Table 4. Comparison of two models (France and Ørskov and McDonald) based on the estimated parameters between the plant and animal protein sources

		Model		
	Parameters	**France**	**Ørskov and McDonald**	**P value for T-test**
Animal protein	A	62.08	61.49	0.96
	c	0.12	0.11	0.74
	T lag	0.50	2.45	<0.001

**A = potential gas production (ml); c = constant rate gas production (ml per hour); T lag = lag phase (hours)*

Table 5. Comparison of France and Ørskov and McDonald models based on the estimated potential gas production parameters of the individual protein sources

	Model		
Source protein	**France**	**Ørskov and McDonald**	**P value For T-test**
	A	**A**	
Soybean meal	287.04	287.48	0.96
Rapeseed meal	215.99	219.68	0.79
Cottonseed meal	111.16	99.12	0.28
poultry offal meal	118.33	117.75	0.95
Fish meal	38.12	37.67	0.94
Blood meal	29.78	29.03	0.81

**A = potential gas production (ml)*

Table 6. Comparison of France and Ørskov and McDonald models based on the estimated constant rate gas production parameters of the individual protein sources

	Model		
	France	Ørskov and McDonald	
Source protein	**c**	**c**	**P value For T-test**
Soybean meal	0.08	0.07	0.23
Rapeseed meal	0.06	0.04	0.01
Cottonseed meal	0.04	0.04	0.89
poultry offal meal	0.12	0.10	0.29
Fish meal	0.10	0.09	0.60
Blood meal	0.13	0.14	0.89

**c = constant rate gas production (ml per hour)*

Table 7. Comparison of France and Ørskov and McDonald models based on the estimated lag phase parameters of the individual protein sources

	Model		
	France	Ørskov and McDonald	
Source protein	**T lag**	**T lag**	**P value For T-test**
Soybean meal	0.34	1.35	0.02
Rapeseed meal	0.62	2.47	0.002
Cottonseed meal	0.16	0.63	0.31
poultry offal meal	0.52	2.21	0.002
Fish meal	0.51	2.39	0.008
Blood meal	0.46	2.74	0.001

**T lag = lag phase (hours)*

solution fraction of each feed serves as an energy substrate for rapid fermentation by attached microbes, and the suitable colonization of microorganisms onto substrate materials, followed by increased fermentation and ultimately reduced lag phase.

However, the importance of the solution fraction to start the degradation and gas production is significant when larger amounts of cell wall components can be provided to microorganisms by better colony and more microbes [12].

4. CONCLUSION

According to the goodness-of-fit tests, the two compared models differ substantially from each other, in particular regarding the estimation of the time-lag preceding the fermentation process. Namely, the sigmoid model FRC, proposed by France et al., appears providing more relevant estimates, in this respect, than does the exponential model EXP by Ørskov & McDonald. For this reason, the sigmoid model FRC should arguably be preferred over the exponential model.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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