DEVELOPMENT OF A NEURAL NETWORK MODEL TO PREDICT THE EXCRETION OF PURINE DERIVATIVES IN THE URINE OF COWS

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Abstract

DEVELOPMENT OF A NEURAL NETWORK MODEL TO PREDICT THE EXCRETION OF PURINE DERIVATIVES IN THE URINE OF COWS.

A Neural Network Model to predict the urinary excretion of purine derivative nitrogen (UPDN) in cows is presented. The input variables of the model are dry matter intake (DMINT), NDF intake (NDFINT), total soluble nitrogen (SP), total soluble non-protein dry matter (SNPDM), total degradable nitrogen (DCP), total degradable non-protein dry matter (DNPDM), hourly available CP in the rumen (HACP), hourly available non-protein dry matter (HANPDM), three different gross indexes of synchronization, namely SYNCA (SP/SNPDM), SYNCB (DCP/DNPDM) and SYNCK (HACP/HANPDM) and two variables describing some metabolic aspects of purine derivative excretion such as live weight of the cow (LW) and milk yield (MILKY). The Model developed uses the Multi Layer Perceptron (MLP) utility, with 13 nodes in the input layer, 8 nodes in the hidden layer and 1 node in the output layer. The Model performances have been tested over 24 observations not previously used to train the model. When compared to a linear regression approach, the Neural Network model showed better performance but under predicted the daily excretion of UPDN for values around 20 g/day. When evaluated in terms of behaviour and depicted scenario the model responded to changes of live weight (LW) and milk yield (MILKY) and to modifications of the pattern of nutrients supplied to rumen microbes.

1. INTRODUCTION

Several models aimed at describing the extent of microbial protein synthesis in the rumen and its response to dietary and animal parameters have been developed. These approaches can be dynamic and mechanistic [1, 2], empirical [3, 4] or static and mechanistic [5].

The urinary excretion of purine derivatives (PD) represents a useful tool to predict the yield of microbial protein in the rumen [6] and has been validated as an indicator in dry and lactating cows [7]. Different approaches have been defined to link PD excretion and microbial synthesis in the rumen in cattle. Some authors [8] have proposed that purine derivatives excretion can be partitioned into an endogenous contribution, which is a function of the metabolic live weight of the animal, and an exogenous contribution which is directly related to microbial purines absorbed in the intestine. In this case the body mass is the sole factor that is recognised to affect endogenous PD contribution. Other authors [9] have proposed that endogenous PD excretion can vary according to the physiological status of the cattle, distinguishing an alternative endogenous PD excretion for dry and lactating cows.

In all these models the endogenous contribution is subtracted from total urine PD excretion in order to estimate the exogenous fraction related to microbial synthesis; however, as the regulation of purine degradation in tissues and organs presents a high degree of complexity, several factors could affect the endogenous excretion of these metabolites, thus the latter contribution could be different from that expected according to the models described.

An alternative approach to study the complex metabolic events concerning PD excretion could be the use of a learning model, such as the Neural Networks.

Neural Networks can be used for real-world applications such as engineering, sensor processing and data analysis [10], and Berg et al. [11] recently used Neural Networks to develop a model for the prediction of pork carcass composition from electromagnetic scans.

A Neural Network typically consists of nodes (*processing elements*) related each other by connections. Each node has a local memory and a *transfer function* which usually has a subfunction called the *learning law*; this latter responds to input signals arriving to the node and adapts the input-out behaviour of the transfer function [10].

The urinary excretion of PD is expressed on weight or molar basis when describing the effect of different factors such as nutrition, species and physiological state of the animal. The same excretion can be referred to as urinary purine derivative nitrogen (UPDN) to obtain a simple synthetic measure to be converted into rumen microbial nitrogen yield. Studies at our Department have shown that UPDN is related to rumen microbial nitrogen yield when estimated by linear regression [9].

An attempt to overcome the liner approach is presented here through the development of a neural network model to predict UPDN excretion in cattle. Such a prediction will be based on aspects affecting UPDN excretion such as metabolic parameters and the pattern of nutrients available in the rumen, which respectively influence the endogenous and exogenous proportion of total UPDN.

2. MATERIAL AND METHODS

2.1. Neural Network: an overview

The present model has been designed adopting a Multi-Layer Perceptron Neural Network, a procedure included in the NEURAL CONNECTION 2.0 software application [12, 13]. Multi-Layer Perceptron (MLP) is a supervised neural network that implements the mapping between the input and output data presented during the *model training*. The *processing elements* (nodes) of the Neural Network are grouped in three different layers: input, hidden and output layers. Nodes within the same layer are not connected to each other "horizontally", but have "vertical" connections to those of the preceding and following layers (Figure 1).

The model assigns a *weight* to each connection between nodes. Weights are modified according to the *learning law* that operates during the Neural Network training process. The inputs to any of the nodes are the result of the input data and the associated weights, the output from a node is the result of input handling by the transfer function. Neural networks have to be trained, and MLP has a training process that follows a supervised training where the network is supplied with an input vector $\mathbf{x}$ and produces an output vector $\mathbf{y}$: each input $\mathbf{x}_i$ is entered in the network with the correct output $\mathbf{y}_i$ as a series of correct input/output pairs [10].

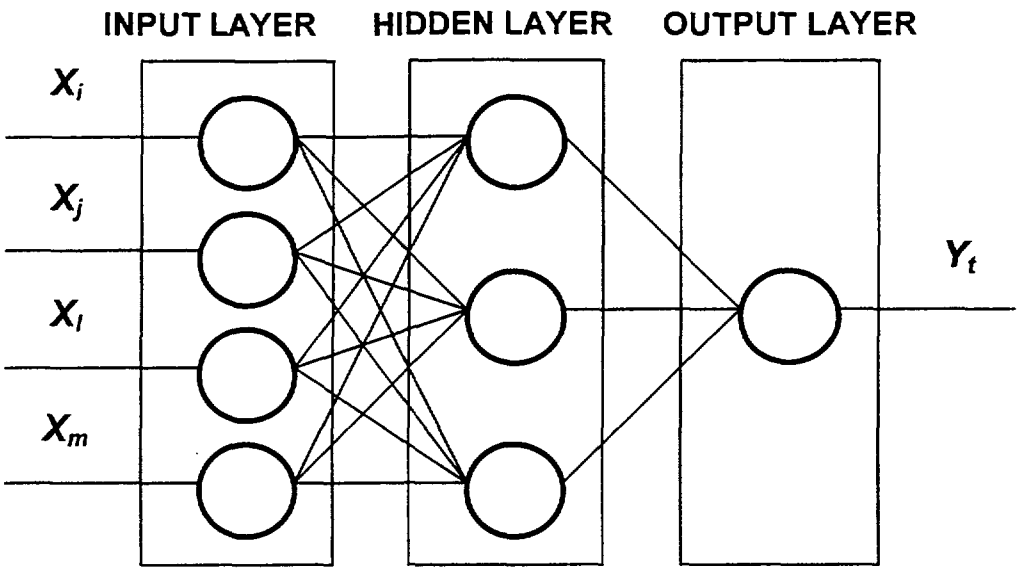

FIG.1. A Schematic diagram of a Neural Network.

In NEURAL CONNECTION 2.0 [12, 13] the training process of the MLP proceeds as follows. First the model randomly sets small initial values for weights and biases, then a training pattern is applied to the input layer and forwarded to the hidden and the output layers. The difference between the *actual* and the *correct* output values is calculated and, in order to reduce the distance between the two outputs, changes in the connection values are automatically established by the software and back propagated from the output layer to the input layer. In the training process the input pattern $\mathbf{X_p}$ that is related to the *correct* output pattern $\mathbf{T_p}$ is presented to the MLP. The initial values of the weights are set randomly to calculate the output from each node in a layer. The output from a node k in the second layer is:

$$\mathbf{z_k = f \sum w_{jk} x_j}$$

where f is the transfer function of the node, $\mathbf{w_{jk}}$ is the weight between nodes j and k, and $\mathbf{x_j}$ is the input value.

The weights between nodes are adjusted according to the back propagation process as follows:

$$\mathbf{w_{jk}(s+1) = w_{jk}(s) + h d_{pk} z_{pk}}$$

where $\mathbf{w_{jk}(s)}$ is the weight between nodes j and k at step s, **h** is the activation function, $\mathbf{d_{pk}}$ is the error of the pattern p at node k and $\mathbf{z_{pk}}$ is the output value at the node k.

The training process can be evaluated in terms of accuracy by comparison of the *correct* output (i.e. the output values used for training) and the *actual* output (that is an estimation of the correct output) of the model. The model is targeted to achieve the lowest error between actual outputs and correct outputs, that represents the best solution of the model.

Two data sets are required to evaluate the model. The data set used to let the network learn the solution of the problem is called the *training data set*. An additional independent input/output data set should be used to test the accuracy of the model. This second data set is called the *validation data set*. The validation process requires the correct interpretation of results. By processing continuously to the lowest training error, the network can also learn errors associated with the training data set. In other words, the model can lose the ability to generalize, in a situation that is defined by the term *over training*. In this case, the network has

learned too precisely the "noise" that accompanies training data and the validation error increases while the training error tends to flatten. The validation data set is then required to monitor the training cycle.

2.2. Input variables, training and validation data set

Data were collected from seven different N digestibility trials with dry and lactating cows, performed at the University of Udine (82 observations) and at the Federal Research Institute for Agriculture in the Alpine Regions in Irdning (48 observations), giving a total of 130 observations. The available measurements were DM, CP and NDF content, intake and digestibility. Moreover, ruminal degradability of CP and non-protein dry matter (NPDM) for the diets offered was also estimated by in situ nylon bag technique [14].

To develop the model, different input variables were considered. Some were used because of their importance in describing ruminal degradability and the availability of nutrients for microbial synthesis in the rumen, (dry matter intake, NDF intake, total soluble nitrogen (SP), sum of the "a" fractions for CP - total soluble non-protein dry matter (SNPDM), sum of the "a" fractions of NPDM - total degradable nitrogen (DCP), sum of the "b" fractions for CP - total degradable non-protein dry matter (DNPDM), sum of the "b" fractions for NPDM - hourly available CP in the rumen (HACP), sum of the b*c terms for CP - hourly available NPDM (HANPDM), sum of the b*c terms for NPDM. Some of these variables were also combined to calculate three different gross indexes of synchronization between carbohydrates and protein availability in the rumen, namely SYNCA (SP/SNPDM), SYNCB (DCP/DNPDM) and SYNCK (HACP/HANPDM). To link the excretion of UPDN to the intermediate metabolism of the animal, further variables were taken into consideration, such as live weight of the cows (LW) and milk yield (MILKY).

The main dietary and animal parameters, the PD excretion along with microbial nitrogen synthesis estimated according to the Italian PDI system [14] of the complete data set are summarised in Table I.

TABLE I. MAIN DIETARY AND ANIMAL PARAMETERS, PD EXCRETION AND MICROBIAL NITROGEN ESTIMATION

	Min	Max	Mean	SD	CV (%)
DM intake (kg/d)	6.92	23.95	12.91	3.69	28.6
NDF intake (kg/d)	3.66	9.66	6.01	1.36	22.6
Soluble crude protein (SOLP) (g/d)	107	1398	541	278	51.4
Soluble non-protein DM (SNPDM) (g/d)	610	4922	2324	1028	44.2
Potentially degradable crude protein (DCP) (g/d)	319	2873	1095	526	48.0
Potentially degradable non-protein DM (DNPDM) (g/d)	2854	11721	6302	1831	29.1
SYNCA	0.113	0.532	0.248	0.107	42.9
SYNCB	0.079	0.280	0.170	0.048	28.1
SYNCK	0.021	0.456	0.166	0.080	48.4
Live weight (LW) (kg)	494	800	622	60.57	9.7
Milk yield (MILKY) (kg/d)	0	35.8	12.3	9.69	8.8
Microbial N [1] (g/d)	27	514	194	99.1	51.0
Urinary PD nitrogen (UPDN) (g/d)	3.1	24.7	10.5	4.57	43.5

[1] estimated according to the Italian PDI system [14]

3. RESULTS AND DISCUSSION

3.1. Development of the neural model

The complete set of 130 observations was randomized into three groups: *training*, *validation* and *testing* data sets. The training data set was used to let the model learn the case study, the validation data set was used to monitor the model error during the learning process and avoid *over training*, the testing data set was finally used to evaluate the performance of the trained model. This latter partition of the complete data set in three different groups was aimed at conducting an *internal validation* of the neural network model; thus the use of the *test data set* was not meant to validate the model. In fact alternative observations from different experiments would have been needed for a classical validation of the model.

Several combinations of the number of nodes in the hidden layer were tested to choose the best Neural Network Model for the data set provided. The number of nodes adopted for the hidden layer depends on the final performance of the model. Thus, increasing the number of nodes can help the model to learn further underlying features of the data set, resulting in a better performance of the neural network. On the other hand increasing further the number of nodes could cause a decrease of the performance of the model as the network could start to learn the *background noise* associated with the data used for the model training.

For the transfer function of the nodes in the hidden layer we adopted a sigmoid function which is a smooth non-linear function with a continuos positive first derivative value. The parameter we adopted to select the different possible models was the standard error (SE) of the output performed by the model on the *testing data set.*

Figure 2 shows how the standard error for testing, training and validation data sets changed according to the number of nodes provided for the hidden layer. As a consequence of the approach we chose the Neural Network with 8 nodes in the hidden layer, as this had the lowest SE for the testing data set. Therefore, the model we adopted was a 13 - 8 -1, a conventional indication for a Multi Layer Perceptron neural network consisting of an input layer with 13 nodes, an hidden layer with 8 nodes and an output layer with a single node.

The ability of the Neural Network model to predict daily UPDN excretion is presented in Figure 3, showing observed (OBS, g/d) and predicted (PRED, g/d) values over the test data set. The function describing the observed vs predicted (OBS = 0.470 + 0.928 × PRED; SE ± 2.329) had average determination coefficient of $R^2 = 0.641$, but the regression coefficient was not significantly different from 1.0 indicating a correct prediction. Figure 4 shows the residuals of the model and the OBS values are also plotted, revealing a tendency of the model to under predict daily UPDN excretion for values around 20 g/day.

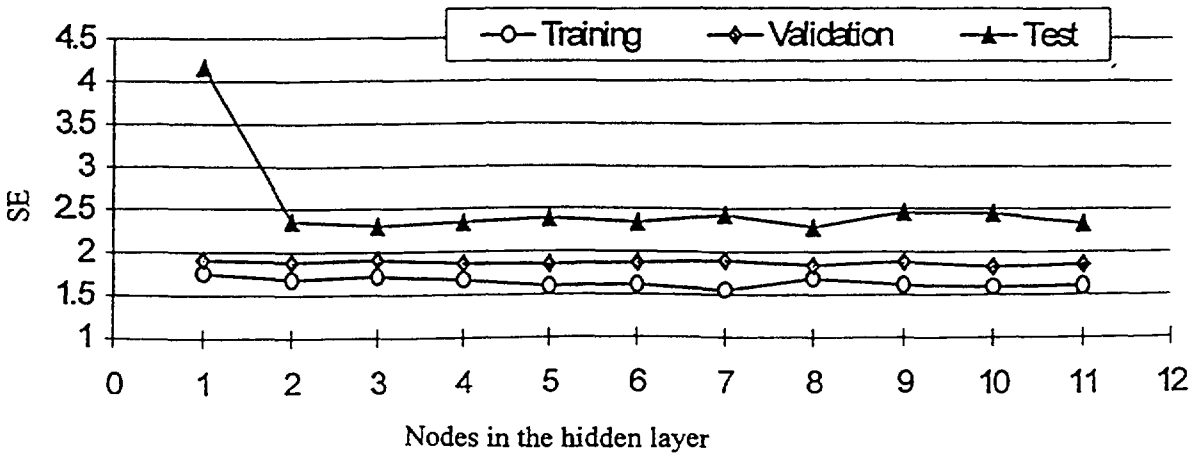

FIG.2. Standard error trend of the Neural Network Model.

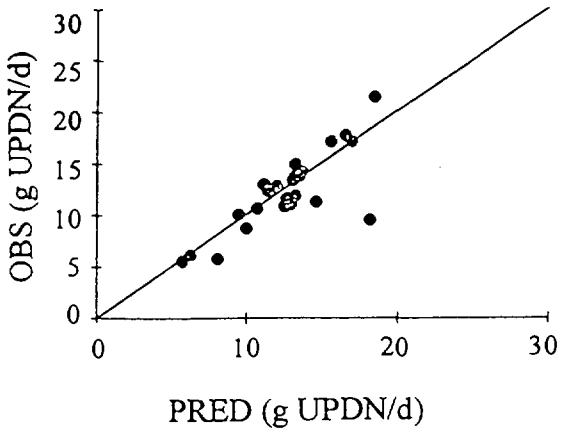

FIG.3. Neural Net Work model prediction.

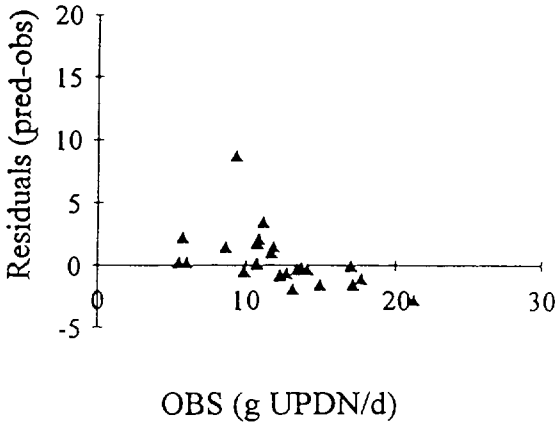

FIG.4. Residuals with Neural Network model.

The choice of adopting a Neural Network model to predict the daily UPDN excretion in cows was also evaluated by comparing the alternative results obtained by two approaches.

First we used the same observations used for training the Neural Network to obtain a linear multiple regression based on 13-input variables and we tested the prediction ability of the latter on the same testing data set used to measure the Neural Network 13-8-1 model performances. Results of this approach are shown in Figures 5 and 6. In the former, observed values (OBS) of daily UPDN excretion and values predicted (PRED) by the multiple regression equation are plotted (OBS = 0.551 + 0.737 × PRED; R^2 = 0.657; SE ± 2.276). In this case the multiple linear regression over predicts the daily PDN excretion, as the regression coefficient b_1 is significantly different from 1.0 ($P < 0.05$). The over prediction can also be appreciated in the plot of residuals vs observed values presented in Figure 6.

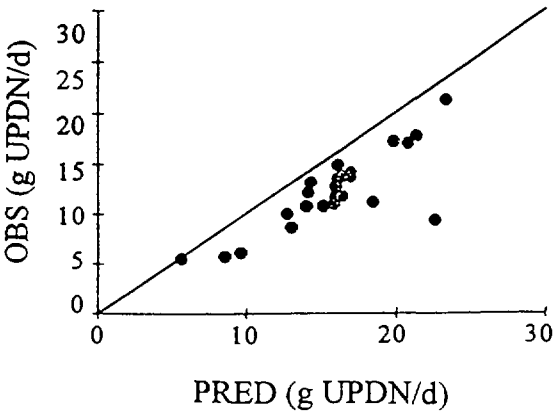

FIG.5. Linear regression prediction.

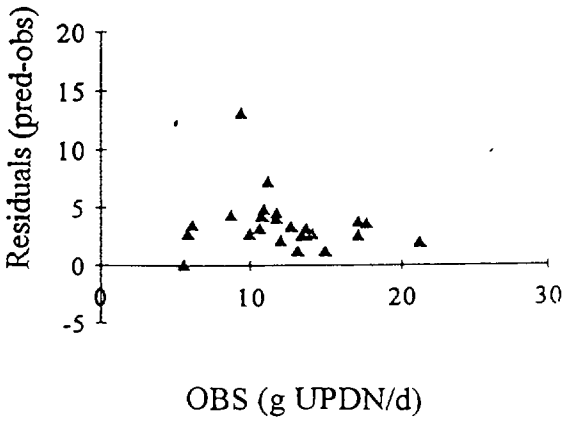

FIG.6. Residuals with linear regression prediction.

We then applied the following empirical equation developed previously at the University of Udine [9] to evaluate the Neural Network.

$$UPDN\ (mg/W^{0.75}) = 12.25 + 29.1 \times L + 44.15 \times TMN$$

This linear equation is aimed at predicting the excretion of purine derivatives nitrogen in the urine of cows from the physiological status of the cows (L, dry or lactating) and total microbial nitrogen synthesis in the rumen (TMN) estimated by the Italian PDI system [14]. The empirical equation was applied to the same testing data set used to evaluate the Neural Model.

The plot of the UPDN excretion predicted by the empirical equation vs the observed values (OBS = 4.084 + 0.561 × PRED; R^2 = 0.388; SE ± 3.040) is shown in Figure 7. The residuals of this prediction are plotted in Figure 8, showing that the equation over predicts UPDN and that the plot of residuals is similar to that of the multiple linear regression seen in Figure 6.

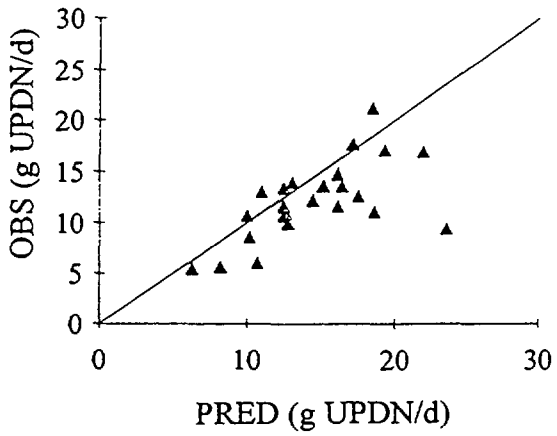

FIG.7. Empirical equation prediction.

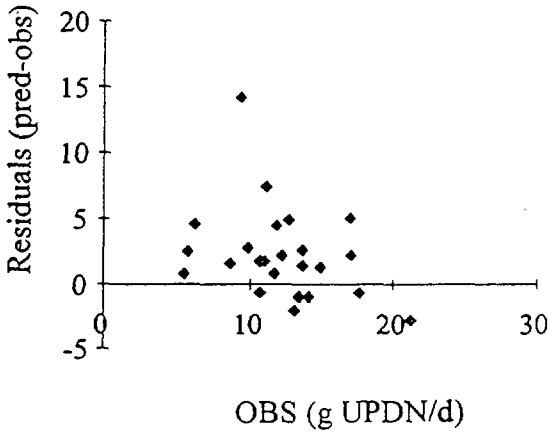

FIG.8. Residuals with empirical equation.

3.2. Model behaviour and scenario

3.2.1. Metabolic aspects

In order to describe the behaviour of the present Neural Network model and the scenario it is able to represent, we plotted two different graphs showing different aspects of the purine derivatives excretion. In Figure 9 we have plotted the model output when changes in the animal's live weight (LW) and daily milk yield (MILKY) are imposed and the other input variables are held constant. It can be noticed that the model predicts a higher UPDN excretion as a consequence of an increased live weight and milk yield.

This scenario is typical of the model developed but relies also on physiological basis. The urinary excretion of PD is dependent on body mass [8] and yielding cows undergo high body tissue turn over particularly during early lactation. Such an increase in tissue turnover may reasonably drive a high endogenous UPDN excretion. We tried to isolate endogenous UPDN excretion related to milk production from that related to the live weight of the cows. For this reason we set LW to zero and changed MILKY to generate a linear relationship between UPDN and MILKY. The equation UPDN = 11.926 + 0.052 × MILKY (SE ± 0.025;

R^2 = 0.998), showed that UPDN increased by 52 mg/kg of milk produced. Conversely, to investigate the effect of LW on UPDN excretion, we set MILKY to zero, varying LW to obtain a linear function. The equation UPDN = 10.974 + 0.002 × LW (SE ± 0.023; R^2 = 0.995), showed that UPDN increased by 2 mg/kg LW. Clearly the latter estimations of 52 and 2 mg of UPDN have to be considered as relative outputs of the model for MILKY and LW variations individually, but despite this limitation it is worth noting that the model was sensitive to these metabolic factors.

When expressed in terms of millimoles, the endogenous PD excretion represented by the present model is equal to 0.268 mmol/kg/$W^{0.75}$ and 0.928 mmol/kg milk produced. These figures can be compared to the endogenous PD excretion estimations presented previously in the literature of 0.385 mmol/kg$W^{0.75}$ [8], and 0.219 mmol/kg$W^{0.75}$ and 0.738 mmol/kg$W^{0.75}$ for dry and lactating cows respectively [9]. In comparison with available literature the neural network model appeared to give reasonable and robust estimation of endogenous contribution of PD excretion.

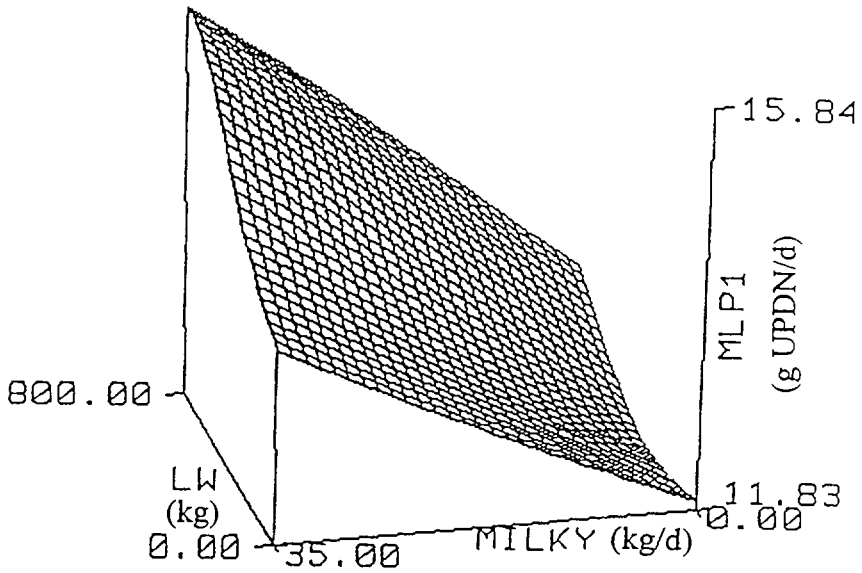

FIG.9. Neural Network model behaviour: Model output (MLP1; g UPDN/d) with respect to variations in live weight (LW; kg) and milk yield (MILKY; kg/d).

3.2.2. Supply of nutrients for rumen microbial population

The model reaction to variation of some ruminal parameters is presented in Figures 10 and 11. Among these, more relevance was given to indexes of synchronization of nutrient availability in the rumen, i.e. nitrogen and carbohydrates, considering their recongnized importance for microbial growth [15, 16].

The model output was investigated by varying dry matter intake (DMINT) and rumen synchronization of the soluble fractions, SYNCA (Figure 10) and when variations in dry matter intake and rumen synchronization of hourly available protein and non-protein dry matter (SYNCK) were imposed (Figure 11). The model was consistently sensitive to variations in dry matter intake. Figure 10 shows that the output UPDN is larger when an increased intake is accompanied by a synchronous availability of the soluble fractions in the rumen, at least at low levels of DMINT. Figure 11, however, shows that the output increases when hourly availability of the non-protein dry matter tends to exceed that of proteins. This latter behaviour was likely due to the fact that hourly available protein in the rumen of the training data set did not limit microbial synthesis.

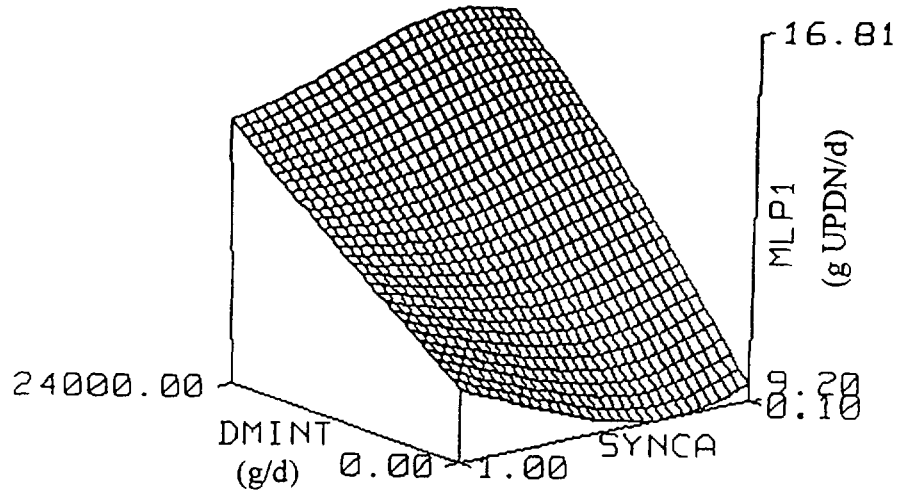

FIG.10. Neural Network model behaviour: Model output (MLP1; g UPDN/d) with respect to variations in dry matter intake (DMINT; g/d) and synchronisation of the soluble fractions (SYNCA; g/g).

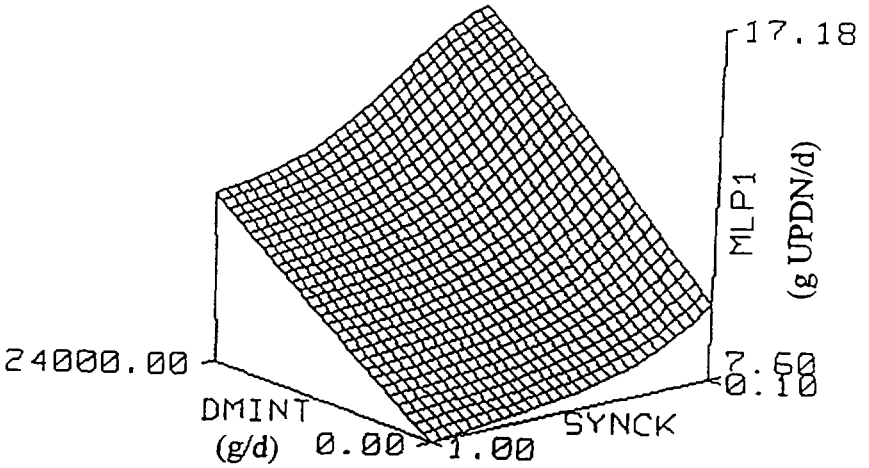

FIG.11. Neural Network model behaviour: Model output (MLP1; g UPDN/d) with respect to variations in dry matter intake (DMINT; g/d) and synchronisation of hourly available protein and non-protein dry matter (SYNCK; g/h / g/h).

4. CONCLUSIONS

The model developed represents an attempt to apply a Neural Network approach to the excretion of purine derivatives in the urine of cows. Although the model showed better performance when compared to classical linear regression approaches, further data from different experiments would be needed to validate the model. The scenario depicted by the model is promising because the model is able to react to variations in the metabolic status of the cow and patterns of nutrients supplied to rumen microbial population. According to the model presented here other factors apart from live weight should be considered to define the fraction of urinary PD that is of endogenous origin. Milk yield and stage of lactation deserve further investigation to clarify their role. Therefore, this model can be useful in the study of physiological principles affecting UPDN in cows.

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