Classification of forage sorghum by application of principal component analysis on near infrared spectroscopic data of sorghum silage

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Introduction

Near infrared (NIR) spectroscopy is widely used to evaluate the nutritional quality of feed crops.

In order to calibrate data, samples are divided into two groups at random. Equations are developed by taking the data from one group and validating the information with predicted data from the other group.

Forage sorghum was classified into several types by their practical use and the importance of these types on making calibrations was investigated.

Materials and methods

Samples

Many varieties and lines of forage sorghum (105 samples) were provided for this experiment. They were harvested at a suitable stage of growth and prepared into silage. Two kinds of silage were made, whole crop silage and stover silage from which the panicles were removed. After they were opened, the feed composition was analysed by chemical methods. In this report, the data on the organic cell wall (OCW), low digestible fraction (LDF) of OCW and crude protein (CP) were investigated.

Forage sorghum were classified into seven groups in order of their practical use and by special genes which reduce lignification of tissue (Table 1). After classifiication into three types of sor-

Table 1. Practical classification of forage sorghum and the sumber of samples.

	(6) Bmr, Bm group	(7) bmr, bm group
(1) Grain type	15	6
(2) Dual purpose type	26	9
(3) Sorgo type	28	
(4) Sudan type (sorghum sudangrass hybrid)	13	
(5) Sudangrass	8	

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ghum—grain type, sorgo type, dual purpose type—and sorghum sudangrass hybrid (called Sudan type) and Sudangrass, two other groups were also analysed in this report. From the grain type and dual purpose type, the lines which have brown midrib (bmr) or bloomless (bm) genes were classified as group (7). The remaining two types of samples were named as group (6). It is known that the bmr and bm genes lower the content of lignin in the leaves and stems of sorghum (Masaoka 1981[1] and Akin 1986[2]). A highly digestible line or variety which are developed from these genes has begun to breed in Japan.

Chemical components expressing fibre

OCW is a feed component which is frequently used in Japan. Samples of feed are incubated with amylase and proteinase. After filtration and ashing, the weight of the residual without ash is called OCW. This is of almost the same value as NDF (in this case, NDF must not contain ash). The same residuals are then incubated with cellulase. After filtration and ashing, the weight of insoluble fraction is measured. This fraction means a low digestible part of OCW. We use the term LDF to express this fraction. LDF is not equal to ADF.

There are several equations which can be used to estimate digestibility of roughage from the components. Sometimes ADF or LDF are used as an independent variable in these equations. In Japan it is said that the accuracy of the equation using LDF is superior to that of the equation using ADF.

Data acquisition, calibration and principal component analysis

The instrument used for spectral data acquisition was an NIRSystems 6500 spectrometer, which scanned the wavelength range 1100–2500 nm and the spectral data were recorded at 2 nm intervals giving 700 datapoints per sample. Calibrations were performed using the multiple regression provided in the NSAS software.

Applying principal component analysis (PCA) to the spectral data, the PC score of the samples was calculated by ISI software, "Symmetry". It selects principal components that have a high contribution rate for all data information on individual chemical components and makes a 3-dimensional space consisting of three major principal components. The position of each of the samples by PC score in the space were plotted.

Results and discussion

Some components in the samples are shown in Figure l.

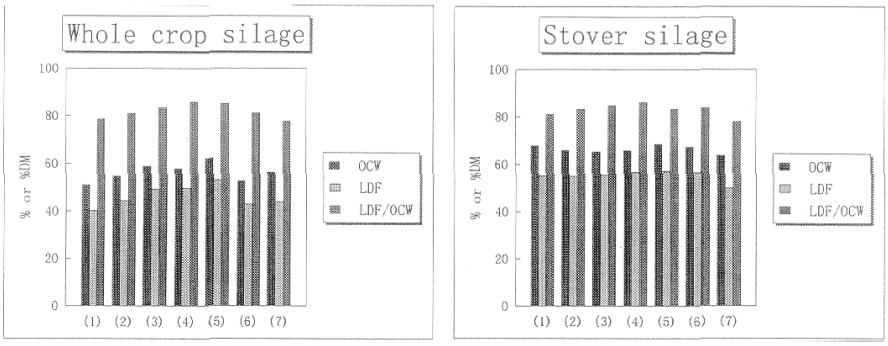

Figure 1. Fibre content of each group. (1)–(7): No. of groups as shown in Table 1; OCW: organic cell wall (% DM); LDF: low digestible fraction of OCW (% DM); LDF/OCW: (%).

Table 2. Calibration developed from all samples.

	Whole crop silage			Stover silage		
	Range of value	Precision		Range of value	Precision	
	(% DM)	*R*	*SE*	(% DM)	*R*	*SE*
OCW	42.3–69.6	0.976	1.44	48.9–77.2	0.973	1.60
LDF	33.7–60.2	0.969	1.56	41.0–67.4	0.965	1.70
CP	6.14–9.60	0.879	0.33	4.44–8.12	0,915	0.32

High performance calibration equations for predicting fibre (OCW, LDF) and CP of both whole crop silage and stover silage were obtained (Table 2).

The prediction of feed composition of one group was obtained using calibration equations developed from the rest of the samples: samples from all the other groups were tested. When the precision of prediction is not high, it can be assumed that the spectral data of the group is not similar to those of different groups. Low precision was shown in the cases of grain type, Sudangrass and the bmr, bm group (Table 3).

Validation between groups

Next, prediction of LDF of one group using calibration equations developed from another group were tested. The prediction of LDF of group 7, using calibration equations developed from group 6 showed that the precision of prediction is not high on the stover silage. On the whole crop silage, the precision was high compared with stover silage. The grain mixed with stover might reduced the difference based on fibre between the bmr, bm group and the Bmr, Bm group [Table 3, Figures 2(a)–5(a)].

Table 3. Validation with LDF on stover silage.

	R	*SEP*	Bias	Slope ad.	(See figure)
Applied to calibration equations developed from the rest of the samples[a]					
group (1)	0.938	1.860	0.876	1.146	—
group (2)	0.980	1.640	0.482	1.010	Figure 2(a)
group (3)	0.968	1.720	–1.180	0.854	—
group (4)	0.950	1.220	0.699	1.311	—
group (5)	0.820	1.340	1.010	0.965	—
group (7)	0.891	2.500	3.090	1.249	Figure 3(a)
Applied to calibration equatiosn developed from another group					
g. (7) –cal (6)	0.930	2.020	2.330	1.394	Figure 4(a)
g. (5)– cal (6)	0.775	1.480	0.773	0.967	—
g. (5) –cal (7)	0.819	1.350	0.724	0.535	Figure 5(a)

[a]The rest of the samples means all other groups' samples.

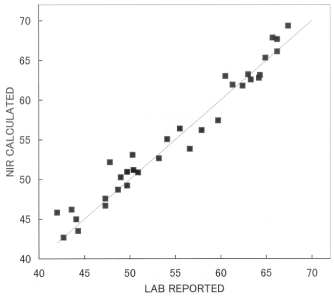

Figure 2(a). Prediction of LDF of group 2 by calibration equations developed from other samples.

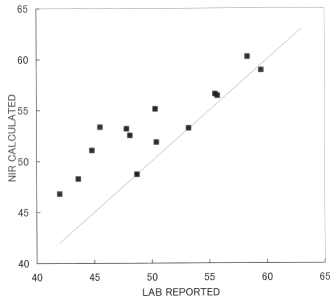

Figure 3(a). Prediction of LDF of group 7 by calibration equations developed from other samples.

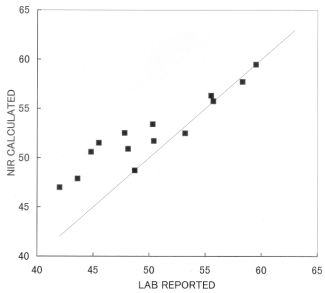

Figure 4(a). Prediction of LDF of group 7 by calibration equation developed from group 6.

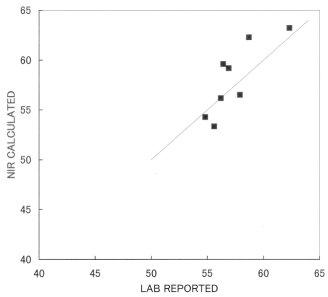

Figure 5(a). Prediction of LDF of group 5 by calibration equation developed from group 7.

The prediction of fibre components of Sudangrass was made using a calibration equation developed from the rest of the samples; all other groups' samples had a low precision. A similar prediction, using a calibration equation developed from group 6, shows high precision, but in the case of group 7, low precision was observed.

Classification by PCA

As with the above validation, a group of samples was plotted in a 3-dimensional space which consisted of three major components calculated from samples from all other groups. When the samples of one group are independent in space and don't intermingle with other samples, it can be assumed that the spectral data of the group are not similar to those of different groups. A high degree of separation in 3-dimensional space was shown in the cases of grain type, Sudangrass and the bmr, bm group when the space consisted of three components connected with fibre. With regard to the grain type, separation of samples in the space was seen not only on fibre but also on CP.

Distribution of samples in 3-dimensional space was illustrated by using the best angle in which the separation of samples would be recognised [Figures 2(b)–5(b)].

The next validation plotted a group of samples in 3-dimensional space which consisted of three major components calculated from another group. When the samples of the bmr, bm group are plotted in 3-dimensional space calculated from the Bmr, Bm group, in the case of stover silage, separation was recognised.

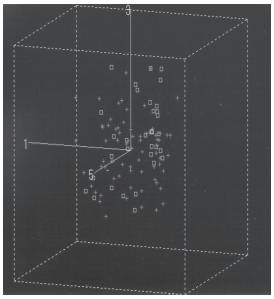

Figure 2(b). Group 2 samples in three principal components space consisting of all other samples.

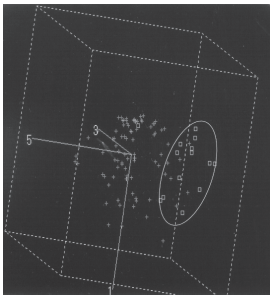

Figure 3(b). Group 7 samples in three principal components space consisting of all other samples.

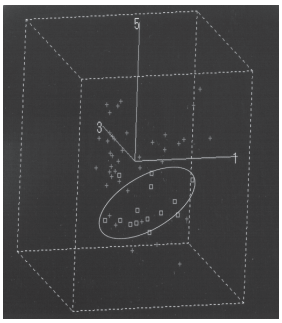

Figure 4(b). Group 7 samples in three principal components space consisting of group 6 samples.

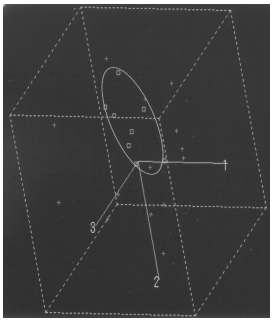

Figure 5(b). Group 5 samples in three principal components space consisting of group 7 samples.

Samples of Sudangrass were plotted in 3-dimensional space calculated from the Bmr, Bm group or the bmr, bm group. Though separation was recognised in both case, the latter showed a more remarkable separation.

Conclusion

From the results of multiple regression and PCA of NIR spectral data, three groups of forage sorghum, i.e. the bmr, bm group, Sudangrass and grain type were classified into groups. In making calibrations for the prediction of chemical components of forage sorghum, it is desirable to utilise samples of all types or groups. In particular, without samples of the bmr, bm group, Sudangrass and grain type, it would be impossible to make a calibration that could accurately predict the components of these groups.

The differences between the groups of forage sorghum were observed mainly on their fibre. Tissue of Sudangrass may be lignificated considerably.

References

1. Y. Masaoka and I. Tarumoto, *J. Jpn Grassl. Sci.* **27,** 87 (1981).
2. D.E. Akin, W.W. Hanna and L.L. Rigsby, *Agron. J.* **78,** 827 (1986).