Associative effects *in vitro* of mixtures of tropical fodder trees

M.Rosales[1],M. Gill[2], C.D. Wood[2] and A. W. Speedy[3]
[1]*Centro para la Investigación en Sistemas Sostenibles de Producción Agropecuaria, AA 20591, Cali, Colombia*
[2]*Natural Resources Institute, Central Avenue, Chatham Maritime, Chatham,Kent ME4 4TB, UK*
[3]*Department of Plant Sciences, University of Oxford, South Parks Road,Oxford OX1 3RB, UK*

Introduction. Previous work has focused on single feeds and assumed additivity during *in vitro* fermentation. In the tropics, farmers are likely to feed mixtures of feeds, including tree fodders, which may not be simply additive in nutritional terms. There is little information about the nutritional interactions between tropical feeds. The objective of this research was to test the existence of associative effects of mixtures of tropical fodder tree leaves.

Materials and methods. Additivity was tested by comparing observed gas production of a mixture (50:50 ratio) with expected values calculated from fermentation of individual components. Using the pressure transducer technique, samples of five fodder tree species were fermented alone and in all possible combinations of two species. Mixtures were fermented in triplicate but six replicates of the single species controls were fermented, to increase the precision of the predicted value. Fermentation was carried out for 70 hours at two levels of nitrogen (from urea) in the medium: 0 and 60 mg N/l. Student's t tests were used to test for associative effects on the cumulative gas production from fermentation of the different mixtures at 12, 24, 45 and 70 hours. The value of t for testing the null hypothesis was calculated and compared against a tabulated point of the t-distribution (5%) with 12 degrees of freedom.

Results. The student's t tests showed significant differences between the sum of the individual components and their mixture for some substrates. In other words, associative effects of mixtures of fodder tree leaves were observed. The difference between the observed and expected values for each mixture is expressed as a percentage in Table 1. Positive and negative associative effects of mixing tropical fodder tree leaves *in vitro* were proved to occur. These associative effects varied with time and with the level of nitrogen. Most of the negative associative effects were related to the presence of condensed tannins in at least one component of the mixture.

Table 1 *Associative effects (%) of mixtures of fodder tree leaves under two levels of nitrogen.*†

	Time (hours)			
Nitrogen-free medium	**12**	**24**	**45**	**70**
Trichanthera gigantea:Gliricidia sepium	18.1**	12.1***	2.9n.s	0.5n.s
Trichanthera gigantea:Inga sp‡.	-5.5n.s	4.9n.s	2.0n.s	1.2n.s
Trichanthera gigantea:Leucaena leucocephala‡	9.0n.s	8.0n.s	3.6n.s	1.2n.s
Trichanthera gigantea:Erythrina edulis	6.0n.s	9.4*	2.9n.s	1.5n.s
Gliricidia sepium:Inga sp‡.	15.5*	9.3**	7.5**	5.8*
Gliricidia sepium:Leucaena leucocephala‡	12.3n.s	7.1n.s	2.6n.s	-0.2n.s
Gliricidia sepium:Erythrina edulis	6.3n.s	0.7n.s	-0.9n.s	-1.7n.s
Inga sp‡.:*Leucaena leucocephala‡*	0.6n.s	3.3n.s	0.8n.s	-1.1n.s
Inga sp‡.:*Erythrina edulis*	-6.3n.s	1.3n.s	3.1n.s	2.7n.s
Leucaena leucocephala‡:Erythrina edulis	-3.0n.s	-2.7n.s	-1.5n.s	-2.3n.s
Level of 60 mg N/l from urea	**12**	**24**	**45**	**70**
Trichanthera gigantea:Gliricidia sepium	5.1n.s	5.2n.s	0.4n.s	0.7n.s
Trichanthera gigantea:Inga sp‡.	0.3n.s	6.5n.s	8.1***	7.9***
Trichanthera gigantea:Leucaena leucocephala‡	7.2n.s	8.2*	8.0***	6.8***
Trichanthera gigantea:Erythrina edulis	0.0n.s	6.3n.s	4.8**	4.4***
Gliricidia sepium:Inga sp‡.	10.1n.s	9.4**	9.7***	9.0***
Gliricidia sepium:Leucaena leucocephala‡	-16.0n.s	-3.3n.s	1.6n.s	1.3n.s
Gliricidia sepium:Erythrina edulis	1.1n.s	1.1n.s	0.6n.s	0.2n.s
Inga sp.‡:*Leucaena leucocephala‡*	-17.8*	-6.6n.s	1.3n.s	2.9n.s
Inga sp.‡:*Erythrina edulis*	-16.4n.s	-6.5n.s	1.0n.s	3.0n.s
Leucaena leucocephala‡:Erythrina edulis	-1.1n.s	4.3n.s	5.2**	5.2**

†Calculated as: (Observed-expected/observed)x 100. ‡Tannin containing species.

Conclusions. The magnitude of associative effects found to be significant in this *in vitro* study varied from 4.4 to 18.1%. It is difficult to predict the consequences of associative effects observed *in vitro* on animal production. However, if effects of the same magnitude found in this study are reflected *in vivo*, the consequences could be important. For example, if the highest positive associative effect of 18% is found *in vivo*, it may mean that the animals will receive almost one-fifth more potentially fermentable material with the mixture than they receive when fed with the single components. This may represent a major benefit to small holder farmers.