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Impact of an Introduced Forage Legume and Grazing on Soil Fertility in Native Pastures of the Humid Tropics of Mexico

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1. Introduction

About 45 %of the world's humid tropics are located in tropical Latin American. The majority (81 %) of soils in this region are acidic, classified as Oxisols, Ultisols and Dystropepts that are deficient in phosphorus (90 %), exhibit aliminium toxicity (73 %), often under drought stress (53 %) and have low nutrient reserves (50 %) (Sánchez, 1987). These constraints in soil fertility are a barrier to high and sustainable productivity and a risk factor that could lead to irreversible soil degradation. Recent research has identified several management options to overcome fertility limitations or prevent the rapid process of soil degradation in such tropical regions (Crowder and Chheda, 1982).

Agricultural practices adopted to improve livestock production, to meet the demand for food of the growing population, have led to an inadequate management of agroecosystems such as native pastures (rangelands). As a result, changes in physical, chemical and biological soil properties have ocurred, to the detriment of soil nutrients and plant growth.

In this Chapter we address the problem of soil fertility in tropical rangelands, through the use of an introduced forage legume (*Arachis pintoi*) and grazing management strategies. This legume was chosen due to its benefits for soil fertility (e.g., N_2 fixation) and good yield potential, mentioned by Kerridge (1994). Several experiments were carried out, in vegetated pots grown under glasshouse conditions and at the field-scale, including grazing experiments on pastures with native plants alone (*Paspalum* spp, *Axonopus* spp, mainly) or a mixture of native plants and the introduced forage legume *A pintoi*. The field experiments were conducted under a rotational grazing regime, in trials that lasted from 12 to 36 months.

Introduced forages contribute strongly to forage quality and productivity in pastures when they are grown in association with grasses, because these take advantage of the N_2 fixed by the legume (Rao and Kerridge, 1994). We believe that sustainable improvement of native rangelands in the Mexican humid tropics could be attained through the use of persistent

legumes that increase animal production by raising the nutritional quality of the domestic ruminant diet. These legumes should also have beneficial effects on soil properties and overall soil fertility.

According to this reasoning, we tested the benefits of legumes related to N_2 fixation, considering that N soil deficiency is a major limitation to pasture productivity in tropical grass-based rangelands. In recent years several new tropical forage legumes have been tested to improve pasture productivity. Some of these genuses are (in alphabetical order) *Arachis, Centrosema, Desmodium* and *Stylosanthes.* As has been mentioned in a previous sentence, about the potential of *Arachis pintoi* (Kerridge, 1994), other authors (Argel, 1994; Cárdenas *et al*., 1999) have suggested this species as promising for the humid tropics of Latin America.

The effect of soil acidity on legumes growing in tropical agricultural systems, including rangelands has scarcely been evaluated (Davidson and Davidson, 1993). The development of soil acidification in pastures containing legumes has been attributed to causes such as: an accumulation of organic matter and an increase in soil cation exchange capacity, to an imbalance in carbon and nitrogen cycles, particularly removal of alkalinity in farm products, net losses of nitrogen through nitrate leaching, and to the release of H+ by legume roots due to excess uptake of cations over anions during N_2 fixation (Tang, 1998). As legumes (by nitrification process) releases H⁺, according to the following soil-plant reaction: NH₄+ + 3/2 O² - + H2O+ + 2H+, we tested the hypothesis that the introduction of the forage legume *A. pintoi* CIAT 17434 in a tropical native rangeland will increase soil acidity.

There are few studies that evaluated soil fertility as impacted by grazing and/or livestock stocking rates under tropical conditions (Cadisch *et al*., 1994; Thomas, 1995; Veldkamp *et al*., 1999). In an innovative experiment, we tested the effect of increased stocking rates on selected soil biological, chemical and physical properties by measuring soil N mineralization, organic matter, C/N ratios, plant-available N, P and K concentrations, bulk density in native pastures, with and without the introduced forage legume *A. pintoi* CIAT 17434.

In all cases, the data were statistically analyzed, using appropriate methods, considering the particularities of each experiment. Results showed that is possible to make a positive impact on fertility of soils, applying technologies that are available and affordable to farmers, on lands that are devoted to livestock grazing.

2. Biological nitrogen fixation by the forage legume *Arachis pintoi* **in a native pasture system in Veracruz, Mexico**

Introduction of persistent legumes leads to improved soil fertility, which enhances dry matter yield and nutritive value of the rangelands. Legume intake improves the diet of the animal, leading to higher milk yields and better reproductive performance. *Arachis pintoi*, a pasture legume native to South America, has been shown to be persistent under heavy grazing, has a high nutritive value, is palatable to cattle, and can improve soil fertility. Agronomic and grazing trials conducted in Veracruz State, Mexico indicated that *A. pintoi* CIAT 17434 was a promising legume for the hot-humid climate and acid, infertile soils of the research station (Argel, 1994; Valles *et al*., 1992; Vos, 1998).

2.1 Experimental site characteristics

The experiment was conducted at the Center for Teaching, Research and Extension in Tropical Animal Husbandry (Centro de Enseñanza, Investigación y Extensión en Ganadería Tropical –CEIEGT) of the Faculty of Veterinary Medicine and Animal Science of the National University of Mexico. The Center is located near to the town of Martinez de la Torre in the North-Central region of Veracruz State, at 20° 03' N, 97° 04' W, at at 112 m.a.s.l. The climate is warm and humid, with an annual rainfall of 1991 ± 392 mm. The average daily mean temperature is 24 ± 0.47 °C. Three seasons can be distinguished. "Dry" from March to June (30.6 \pm 1.7 °C and 19.9 \pm 1.7 °C of maximum and minimum temperatures, respectively, and 477 \pm 343 mm of rainfall). "Rainy" from July to October (31.2 \pm 1.3 °C and 20.5 \pm 1.8 °C of maximum and minimum temperatures, respectively, and 1032 \pm 612 mm of rainfall) and, "Winter" from November to February (25.0 \pm 2.1 °C and 15.1 \pm 1.3 °C of maximum and minimum temperatures, respectively, and 482 ± 268 mm of rainfall)

The soil has been classified as Ultisol and has moderate drainage. The *A* horizon has a maximum depth of 30 cm. There is no *B* horizon. Horizon *C* is an acid hardpan formed with material that was transported deposited and compacted. Due to the microrelief, soils are prone to waterlogging during the wet and northern wind periods and thus horizon *A* exhibits gleyization (Hernández, 1988). A chemical description of the soil in the experimental site is shown in Table 1.

¹N(%): Nitrogen in soil >1.0=very high, 0.5-1.0=high, 0.2-0.5=medium, 0.1-0.2=low, <0.1=very low (Landon, 1984).

² Natural enrichment of the ¹⁵N isotope expressed as δ ¹⁵N ($\frac{0}{00}$). Values could vary from -6 to + 16 δ ¹⁵N. 3 C (%): Total soil carbon.

 48 PDB carbonate standard: Isotopic composition of ¹³C. The range in soils is -9 to -32 ($0/_{00}$). The values reflect the relative contribution of plant species (C3 plants: -32 to -22 $^0/\omega$, or C4 plants: -9 to -17 $^0/\omega$.) (Boutton *et al*., 1998).

 $5P$, Bray I (µg/g soil): A range of 2.5 to 11.4 µg/g soil is considered as critical level for tropical legumes (CIAT, 1984).

6 Soil pH (1:2.5 H2O): 4.5-5.0 very strongly acidic, 5.0-5.5 strongly acidic, 5.5-6.0 moderately acidic (Landon, 1984).

Table 1. Chemical description of the native pasture soil.

2.2 Rangelands used

Paddocks were composed of native pasture-*Arachis pintoi* from an ongoing grazing experiment at the research station in Veracruz, Mexico. Paddocks with native pasture alone

(mixed grasses, mostly *Paspalum* spp, and *Axonopus* spp,) were included as a control. The paddocks described in this chapter, are nested within one of seven permanent divisions of the mentioned native pasture-legume association and were originally planted to study the establishment and persistence of the accessions CIAT 17434, 18744 and 18748 under grazing. Three replicates of each ecotype were located within the pasture, and native grasses grew in mixtures with the legume ecotypes. These plots have been under rotational grazing (1-day grazing and 20-day rest periods), by sectioning the divisions into three temporary subdivisions. There have been 5 "fixed" cows per treatment and in all treatments, three additional cows per treatment were introduced in order to consume the extra forage produced in the rangelands during the rainy season. Thus, stocking rates were 2 cows/ha when only fixed cows grazed and 3.2 cows/ha when both fixed and additional cows grazed the experiment. These rangelands were not fertilised with mineral or organic fertilizer for at least five years prior to the planting of the legume, nor after legume planting. These grasses grew mixed in the ecotypes plots. Introduced legumes and adjacent native grasses were sampled at the beginning and at the end of each one of the three seasonal periods (dry, rainy and winter) during 1999.

2.3 N2 fixation assessment and statistical data management

Dry matter yield of shoots was measured in legumes, as well as in grasses, and analysed for N (%) and δ ¹⁵N ($0/_{00}$) using a Europa 20-20 isotope ratio mass spectrometer (Europa Scientific, Crewe, UK), coupled to an automated Roboprep C/N analyser (Europe Scientific, Crewe, UK).

The percentage of atmospheric N_2 fixed by the legumes (P) using the natural ¹⁵N abundance technique was calculated using the following equation (Bergersen & Turner, 1983; Ledgard *et al*., 1985):

$$
P = \frac{\delta^{15} N \ ref - \delta^{15} N \ leg}{\delta^{15} N \ ref - B} \times 100
$$
 (1)

where *B* is the δ ¹⁵N of the legume grown with atmospheric N₂ as the only N source, δ ¹⁵N ref and δ ¹⁵N leg are the δ ¹⁵N units ($0/_{00}$) for reference plant and legume, respectively. *B*-values of shoots corresponding to each ecotype from the "Isotope fractionation" were used. These values were (⁰/00) 0.08, -0.18 and –0.29 for *A. pintoi* CIAT 17434, 18744 and 18748, respectively.

The total N2 fixed for every *A. pintoi* ecotype was calculated using the following equation:

Total N₂ fixed
$$
(kg/ha) = Dry
$$
 matter yield $(kg/ha) x (%) / 100) x$
 $(%N2 fixed / 100) x (%) (% legume in positive / 100)$ (2)

In each pasture treatment, botanical composition of each component (%) was estimated using the dry weight rank (DWR) method of Mannetje & Haydock (1963) four times a year: March-April, June-July, September-October and December-January.

The experiment was analysed as a completely randomized design with two rangelands (native pasture (NP) and the association (NP+*Ap*) each with three replicates. The comparison among the *A*. *pintoi* ecotypes was done also as a completely randomized design with three treatments (the ecotypes) and two replications.

2.4 Results

2.4.1 N2 fixation by *Arachis pintoi* **ecotypes**

Figure 1 shows the percentage of N_2 fixed by the three *Arachis pintoi* ecotypes using either *Axonopous* spp (Figure 1a) or *Paspalum* spp (Figure 1b) as reference plant, averaged for each climatic period. The analysis of variance did not find statistical differences in N_2 fixation for ecotype or season within by each reference plant, and *Ap* CIAT 18744 averaged 86 % of fixed N2, compared to reference plants. When data were analysed considering both reference plants, statistical differences among ecotypes were found. *Ap* CIAT 18744 and 18748 were statistically similar ($P \le 0.05$) but different to the ecotype *Ap* CIAT 17434, e.g. %N fixed was 86 ± 2.3 , 77 ± 3.1 and 69 ± 4.5 , respectively.

2.4.2 Botanical composition

The highest proportion of *A. pintoi* (on average, 29%) was measured in the native pasture during the dry season, with less prominence (16-19%) on other sampling dates. The average legume content for the whole year was 20 %, while native grasses were 64 % of the stand. Weeds and a low proportion of native legumes (3%), and bare soil (13%) occupied the remaining area.

2.5 Discussion

The best N2 fixation estimates (%) were obtained with *Arachis pintoi* CIAT 18744, regardless of the reference plant used (Figure 1); although the ecotype CIAT 18748 fixed a similar proportion of N_2 as ecotype CIAT 18744. The proportions of N_2 fixed were higher than the values reported by Peoples *et al*. (1992) for a variety of the grain legume *A. hypogaea*, in Australia. They estimated a N_2 fixation in two years of 22 to 31 % and 44 to 48 %, respectively using a non-nodulating peanut genotype as a reference plant. Likewise, Cadisch *et al*. (2000) reported percentages of N2 fixed in *A. hypogaea* of 45 to 54 % in the first year and 21 to 16 % in the second year, also with a non-nodulating groundnut and maize as reference plants.

In Brazil, Behling-Miranda *et al*. (1999) reported that four non-inoculated *Stylosanthes* species fixed (on average) more N_2 using *P. plicatulum* as reference plant (on average, 80 %) than with *B. decumbens* (71 %). In our experiment, no major difference in N_2 fixation was calculated due to the choice of reference plant, e.g. *A. pintoi* ecotypes gave similar estimates of N2 fixed whether using *Axonopus* spp or *Paspalum* spp were used as the non-N2 fixing reference plant. In Colombia, Suarez *et al.* (1992) evaluated the N₂ fixation of non-inoculated *Arachis pintoi*, using the isotope dilution method and found that the legume derived 63 % of its total N from the air, which is lower than the results shown in this Chapter. In our case, although the legumes were not inoculated, the high levels estimated of N_2 fixation, mainly with the ecotypes *Arachis pintoi* CIAT 18744 and 18748 showed that the native *Rhizobium* population was effective in N_2 fixation.

A rough estimation of kg/ha/day of N2 fixed by *A. pintoi* ecotypes could be obtained if we consider a DMY of 25 kg/ha/day for this legume under field conditions (average from two years of data collected by Valles *et al*. (1992) in Veracruz, Mexico). Using this assumption and considering the measured percentages of N_2 fixed by the *A. pintoi* ecotypes as well as the proportion of the legume in the associated pasture, the estimated amounts of N_2 fixed are shown in the Table 2.

The estimated N_2 fixation during the rainy season, considering 71, 93 and 77 % of N_2 fixed by *A. pintoi* ecotypes and using data from Table 2 for N % and presence of the legume (%) resulted in 140, 230 and 148 g N/ha/day for Ap 17434, 18744 and 18748, respectively. This is within the same range as Thomas *et al*. (1997), who reported for *A. pintoi* CIAT 17434 (23 % of the pasture stand and deriving 79 % of its N from N_2 fixation) a figure of 11.4 kg N/ha/8 weeks, e.g. approximately 200 g N/ha/day. Our values are also similar to Cadisch *et al*. (1989) for eight forage legumes in Colombia, e.g. on average 463 and 202 g $N_2/ha/day$ for legumes that received or no P+K fertiliser, respectively.

2.6 Conclusions

The ecotypes of *Arachis pintoi* CIAT 18744 and 18748 had the highest % N derived from N² fixation. This represents a significant input of N to the pasture systems in the tropics. Although the ecotype CIAT 17434 showed a reasonabe percentage of N_2 fixation, its estimated N contribution to the pasture was lower, mainly due to its low presence in the sampled plots. On the other hand, all other ecotypes seemed to tolerate intensive grazing and competed successfully with aggressive grasses and weeds, e.g. maintained a population of about 20% of the stand in the native pasture.

Fig. 1. Percentage of nitrogen fixed by three *Arachis pintoi* ecotypes considering a) *Axonopus* spp or b) *Paspalum* spp as reference plant, during three climatic periods in Veracruz, Mexico. Lines upon bars are standard error of means.

¹Estimations in the paddocks of *Ap* ecotypes during February 1999.

2 Average values from Figure 1.

Table 2. Estimation of N2 fixed by three *Arachis pintoi* ecotypes under field conditions in Veracruz, Mexico.

3. Changes in soil properties after the introduction of *Arachis pintoi* **in native rangelands in the humid tropic of Mexico**

In the Mexican humid tropics, native rangelands compose from 25 to 75 % of the pastureland, and are the main feed source for cow-calf and dual-purpose production systems. In practice, these lands are not fertilized or sown with productive legumes to add nitrogen (N) to the system. Since N is the element driving pasture growth, decades of use without adequate N fertilization (either from nutrient inputs or N_2 fixation) have led to considerable degradation of soil and grassland fertility. Legumes are well known as soil building plants. They enhance soil quality by adding organic matter, and improve soil structure and water infiltration. Earthworm populations are usually greater in fields planted to perennial forages than in fields planted to annual row crops, which may further enhance nutrient recycling in these pastures.

Arachis pintoi CIAT 17434 is a legume that has induced increases in milk and meat production when planted in association with introduced grasses like African stargrass (*Cynodon plectostachyus*, *C. nlemfuensis*) and *Brachiaria* spp (González *et al*., 1996; Hernández *et al*., 1995), and is known to raise soil N and C inventories (Ibrahim, 1995). The present study documented the effect of the perennial forage legume *Arachis pinto*i CIAT 17434 when introduced into native grasslands on some soil variables, considering soil physico-chemical properties important for pasture sustainability.

3.1 Experimental details

3.1.1 Climate and soil in the experimental site

The experiment was carried out at the Center for Teaching, Research and Extension Center in Tropical Animal Husbandry (Centro de Enseñanza, Investigación y Extensión en Ganadería Tropical - CEIEGT) of the Faculty of Veterinary Medicine and Animal Science (FMVZ) of the Universidad Nacional Autónoma de México (UNAM). The CEIEGT is located on the Gulf of Mexico coastal plain, 40 km west of the coast (20° 02' N, 97° 06' W) at 112 m above sea level. Climate (1980-2000) is hot and humid with rains year round, and an average annual rainfall of 1931 \pm 334 mm. Mean monthly rainfall is highly variable (161 \pm 132 mm), but is generally sufficient for pasture growth as dry periods are occasional and short. Average daily temperature during the study was 23.9 ± 6.4 °C, with average monthly maximums (29.2 \pm 3.3 °C) and minimums (18.6 \pm 3.9 °C) that are reasonably uniform from

year to year. There are three seasons: rainy fromJuly to October, with high rainfall and temperature; winter or "northwind" from November to February, with rain and lower temperatures; and dry from March to June. Low winter temperatures and high evapotranspiration during the dry season do not favor forage production, because the shallow soils of the experimental site do not store much moisture.

Soils in the experimental fields are acidic, heavy clay, Ultisols (Durustults) with low P (3.5 ppm by Bray, 2.0 ppm by Olsen), S, Ca and K concentrations, low cation exchange capacity (10.5 meq/100 g) and aluminium saturation levels below the level toxic to plants (Arscott, 1978; Toledo, 1986). There is an impermeable soil layer from 0 to 25 cm depth that causes inadequate drainage during the rainy season and winter. Soil samples taken in the experimental area in 1998 showed that C content decreased from 1.02 ± 0.44 % at 0 to 15 cm, to 0.78 ± 0.15 % at 15 to 30 cm, and that N content also dropped, from 0.13 ± 0.06 at 0 to 15 cm, to 0.10 ± 0.04 % at 15 to 30 cm.

3.1.2 Treatments and soil variables assessed

Two treatments were applied, a native grass pasture (NG) and a NG associated to *A. pintoi* CIAT 17434 (NG+Ap); each one nested in a larger field (F1 and F2, respectively). Each field was divided into seven permanent sections measuring 22 m wide by 165 m long. These were then temporarily subdivided into three paddocks of 22 m by 55 m (21 paddocks per field), to allow a rotation grazing of 1 d of grazing and 20 d of recovery. Stocking rate (SR) was 2 cows/ha during the low forage production period and 3.2 cows/ha during the rest of the year. The SR was reduced when standing dry matter before grazing was less than 2,500 kg/ha in any paddock.

Soil samples were taken in April 1999 and September 2000 in transects located in the center and passing through the entire length of each of the seven divisions. Six soil samples were taken along each transect at even intervals, for a total of 336 soil samples. Samples were analyzed for the variables described below.

To estimate apparent density (AD, $g/cm³$), a metal cylinder (50 mm long, 50 mm internal diameter) was inserted into the bare soil to collect a soil sample, which was dried at 65 °C until constant weight was reached (Anderson & Ingram, 1993). Samples for other variables were dried at room temperature, cleaned of all roots and other organic matter and then ground until they passed through a 2 mm mesh. A potentiometer was used to measure pH in a soil: distilled water (2:1) suspension. Organic carbon (C) was analyzed according to Walkley & Black (1934), and organic matter (OM, %) was calculated assuming a 58% C content in the OM (Anderson & Ingram, 1993). Total C for the 0-20 cm soil layer was calculated using Ct = Vs x DA x C/100, where: Vs is volume in 1 ha at 20 cm depth, AD is apparent density (kg/m³) and C is as defined above. It was assumed that AD in the 0-5 cm layer was representative of the entire 0-20 cm layer. Nitrogen (N, %) was estimated with the Kjeldahl technique calculating total N (Nt, kg/ha) similar to Ct by substituting C for N.

Field 1 was evaluated every three months to determine the contribution of *A. pintoi* to the botanical composition. Samples for this analysis were taken from two paddocks, each within the three divisions chosen visually as the most representative of the experimental area. Sections b1, d1 and f1 had higher *A. pintoi* content compared to the other three (i.e. b3, d3 and f3), and were consequently classified as having high and low legume content, respectively.

3.1.3 Data statistical management

The division was used as the experimental unit and thus variation between sampling sites within a transect was not considered. Variance analysis was done separately for F1 and F2. The resulting linear model was:

$$
Yijk = M + Tj + Di(Tj) + Ak + (T x A)jk + Eijk,
$$
\n(3)

where: Yijk is the response variable, recorded in the transect corresponding to division i, in treatment j, in year k; M is the general mean, common to all observations; Tj is the effect of treatment j (j: NG and NG+Ap); Di(Tj) is the variation between divisions within treatment j, used as an error to test the treatment effect; Ak is the effect of year k (k: 1999 y 2000); (T χ A)jk is the combined effect, or interaction, of treatment by year; and, Eijk is residual variation, used as an error to test the effect of the year and interaction. The annual means were not included because the samples were taken in different seasons each year, and to concentrate on the effects of treatments.

3.2 Results and discussion

3.2.1 Effect of rangelands on soil variables

The T \times A interaction did not affect (P>0.05) any response variable, meaning the effects of T and A were independent. The effect of T on pH and AD in F1 was not significant, was close to significant for the C:N ratio (P=0.0563) and was significant for all other variables (P<0.05). In contrast, T had no effect on any variable in F2 (Table 3). The different effect of treatments between can be explained by the length of time the legume had been growing in each field, meaning this legume needs a medium to long term to start manifesting its beneficial effects on the soil. Fields 1 and 2 were established in November 1996 and 1999, respectively.

Legumes are reported to reduce soil compaction at higher stocking rates (Alegre & Lara, 1991), though *A. pintoi* had no effect on compaction during the three-and-half year period of this study. In Costa Rica, Ibrahim (1994) found that an increase in stocking rate from 2 to 3 animal units/ha in a *Brachiaria* spp/*A. pintoi* association only slightly increased AD from 0.76 to 0.85 $g/cm3$ in the 0-5 cm layer.

Previous research (Thomas *et al*., 1997; Valles, 2001) indicates that *A. pintoi* CIAT 17434 derives from 65 to 85% of its N_2 from fixation, suggesting N accumulation in the soil, probable $NO₃$ leaching losses and soil acidification (Haynes, 1983). In the present study, however, *A. pintoi* had not been planted long enough to influence pH. Valles (2001) found no signs of difference in pH in samples of the soil profile to 1 m depth in both treatments in F1, one year after *A. pintoi* had been established. However, in native grasslands associated with this legume for five years, the pH was significantly (P<0.05) lower at all depths than in grasslands without it. The pH values for the NG+Ap were 5.05 (0-5 cm), 5.07 (5-15 cm) and 5.52 (15-30 cm), and for the NG they were 5.66 (0-5 cm), 5.44 (5-15 cm) and 5.72 (15-30 cm). This suggests the possibility of a decrease in pH over time in the soils with the *A. pintoi*/native grasslands association.

a, b Means with different letters within a row are significantly different (P<0.05).

Table 3. Effect of pasture on soil variables in the two, grazed paddocks with natural grass (GN) or natural grass and the introduced forage *A. pintoi* (GN+Ap) in Veracruz, Mexico. Soil sampling en Field 1 was done 16 months after the establishment of *A. pintoi*; and in Field 2, sampling was 20 months before the establishment of the legume. Values are the mean ± standard error.

Accumulation of OM in the soil of pasture agroecosystems is due to continuous addition of roots and plant litter, coupled with a chronic N deficiency (Huntjes and Albers, 1978). Although in Field 1, N deficiency probably was not evidente, because the supply of this element by the N_2 fixation from the legume, changes in organic C and total N was probably due to the build-up of organic matter process, done by soil microorganisms on grassland vegetation.

The calculated initial (1998) Ct value for both treatments was 24.7 t/ha. Average annual Ct increase to the year 2000, by linear regression, was 3.7 t/ha/yr for NG and 4.8 t/ha/yr for NG+Ap, an acceptable differential increase. This coincides with observations reported by Fisher *et al.* (1994) for the Colombian savannah and by Ibrahim (1994) for the Costa Rican humid tropics. The C:N ratio is a soil quality indicator with an ideal value of 10:1 or less. In F1, the NG was above the ideal value, and the NG+Ap was slightly below it (Table 3). In F2, both treatments were higher than the ideal ratio. This difference is likely due to the different amounts of time the legume had been established in the two fields. Data from Costa Rica (Ibrahim, 1994) show a C:N ratio in a *Brachiaria* spp/*A. pintoi* association of 12.5:1, which is

comparable to the present F1 control treatment but lower than the F2 results. This suggests that *A. pintoi* remains productive in this association and needs time to alter the soil C:N ratio.

3.2.2 Contribution of *Arachis pintoi* **to soil C and N stocks**

The contribution of *A. pintoi* to the botanical composition increased linearly in sections of Field 1 with high and low initial content of the legume. In the sections with high legume composition, its abundance continued to increase at a rate of 13.3% per year, whereas the sections with low initial legume content showed an increase of 7.8% per year (Figure 2). The respective contribution of the legume to the soil C and N stocks tended to be greater in sections with high legume content than those with lower legume presence, although not significant due to high variation (Table 4). An important aspect of productivity in tropical rangelands is to maintain or increase the stock of soil C and N in response to increases in biomass induced by the biological N_2 fixation, which implies long-term sequestration of $CO₂$ and improved soil fertility. There are also negative effects to biological fixation of N_2 , such as soil acidification, that are only apparent over the long term (Haynes, 1983).

3.3 Conclusions

Introduction of *Arachis pintoi* CIAT 17434 to native rangelands increased soil organic C and total N concentrations, but had a marginal effect on the apparent density and soil pH. This indicates that this legume can improve soil fertility, particularly from the perspective of building soil organic matter, thus maintaining a sustainable dual-purpose cattle system in the tropics.

Fig. 2. Increase in the contribution of *Arachis pintoi* to botanical composition in Field 1 over a three year period (1997-2000). Field 1 was a grazed paddock with a mixture of natural grasses and introduced *A. pintoi*, located in Veracruz, Mexico.

Table 4. Contribution of *Arachis pintoi* to botanical composition, and amounts (kg/ha) of soil organic carbon and total nitrogen at 0.2 m depth, 9 months after the introduction of this forage legume to a grazed paddock in Veracruz, Mexico. Values are the mean ± standard error- $(n = 6)$.

4. Evidences of topsoil acidification by the forage legume *Arachis pintoi*

Soils in tropical Latin American include 45 % of the world humid tropics, with a high proportion of acid soils. Oxisols, Ultisols and Dystropepts cover 81 percent of these areas. They are characterised as deficient in phosphorus (90 %), aluminium toxicity (73 %), drought stress (53 %) and low nutrient reserves (50 %) (Sanchez, 1987).

These constraints in soil fertility are a barrier to high and sustainable productivity and a risk factor that could lead to irreversible soil degradation. Recent research highlights several management options in order to overcome fertility limitations or prevent the very fast process of soil degradation. The introduction of legume-based rangelands is considered as one of the most desirable option to reduce the fertiliser requirements in animal production systems.

4.1 Implications of soil acidity

Basically there are two acidity sources: the atmosphere and the soil. In the soil, the decomposition of organic matter by mineralization, nitrification and leaching releases H⁺ ions. In agricultural systems, the use and abuse of N fertilisers, particularly those containing ammonium ions, make large contributions to soil acidity. Fortunately for plants, there are buffering materials in soils that consume H⁺ ions and maintain the soil pH at a more or less constant level.

4.2 Acidification by legumes

The soil acidity induced by an intensive use of legumes in animal production systems and other agricultural systems has been scarcely evaluated under tropical and sub-tropical conditions. In Australia, Davidson & Davidson (1993) cited reports of this problem dated since 1939 and Haynes (1983) makes mention of references since 1954. Considering the previous reasoning, we set up this experiment to determine if there is a relationship between pasture age and soil acidification under legume covers.

4.3 Soil sampling and rangelands used

The experimental fields were located at the Center for Teaching, Research and Extension in Tropical Animal Husbandry (CEIEGT) of the Faculty of Veterinary Medicine and Animal Science of the National University of Mexico. The soils were classified as Ultisols. Additional details about this site were described previously in this Chapter.

During the period June-August 1997, soil samples were taken at four sites containing native rangelands associated with *A. pintoi* CIAT 17434 of different ages (1, 5, 8 or 11 years) and corresponding controls (Table 5). The control areas, all adjacent to *A. pintoi* (*Ap*) sites, consisted of soils with native vegetation, which were under rotational grazing regime at the time of sampling, except the control site for the pasture 8 years *Ap* that remained without grazing The 11 years control site remained under grazing until 2 years before sampling.

The total size of each site was 7.5, 10.0, 0.5 and 0.25 ha for 1, 5, 8 and 11 year old *Ap* sites, respectively. In the first site, 7.5 ha of native rangelands were assigned to carry out a grazing experiment of which 5 ha remained unchanged and 2.5 ha were used to introduce the legume (1 year *Ap*). In the 5 years old *Ap* site, half of the site were used to introduce the legume mixed with the native pasture or associated with African star grass -*Cynodon nlemfuensis*. On the other sites (8 and 11 year old *Ap*) *Arachis pintoi* was planted to establish seed banks for further spread of the legume. No fertilisers were added during the establishing period, nor during the last five years.

1 .- nd= not determined.

Table 5. Chemical description of four control soils in rangelands located in Veracruz, Mexico, in 1997.

At the time of soil sampling, these areas were divided each into four portions to obtain a more uniform sampling. In each quadrat, samples were taken using an auger at several depths . At the time of soil sampling, these areas were divided each into four portions to obtain a more uniform sampling. In each quadrat, samples were taken using an auger at several depths . All sites were sampled at 0 - 5, 5 - 15 and 15 - 30 cm depth, and the 1 year old site was additionally sampled at 30 - 60 and 60 - 100 cm depth.

All sites were used to estimate changes in soil fertility and acidity. The samples were immediately identified and air dried for 3 - 4 days before stored in black plastic bags until further analysis. In all cases, soils were passed through a 2 mm sieve.

4.4 Soil pH and statistical analysis

Measurements of pH were made by weighing 10 ± 0.1 g air-dried soil (< 2 mm) in a test tube and adding 25 ml of deionised water. Tubes were stirred for one minute, left to settle for one hour and stirred again for one minute. A pH meter with a glass electrode and previously calibrated for acid soils (pH=4) was used. Between each pH reading value, the electrode was washed with water and gently dried with a tissue. Every ten measurements, the pH meter was checked to ensure correct pH readings. Data were statistically analysed performing analysis of variance and differences among means were established by least significant differences, using the Genstat statistics program (Lawes Agricultural Trust, 1996). Regressions using "age" as a dependent variable were also considered but very low *R²* (determination coefficients) were obtained.

4.5 Results

4.5.1 Changes in soil pH

Changes in soil pH for every one of the paired sites (with and without *A. pintoi*) was analysed statisticall (Figure 3). There were no significant differences in pH between the 1 year old *Ap* based pasture and the control pasture in the topsoil (5.2), although the control soil was less acid at 15-30 cm. The 5 years old Ap pasture showed lower soil pH ($P \le 0.001$) than the control site at all sampling depths. Larger differences in pH were observed between the two rangelands at 5 cm depth. Likewise, the soil pH increased significantly (P≤0.001) with increasing depth in the soil profile, average values being 5.3, 5.3 and 5.6 for the depths 0-5, 5-15 and 15-30 cm, respectively. The pH of the 8 year old *Ap* pasture (pH=5.3) was statistically different ($P \le 0.001$) from the control site (pH=5.6) ($P \le 0.001$).

The acidity level (soil pH) in the *Ap* pasture decreased from 5.4 at 15 - 30 cm to 5.2 at 0 - 5 cm depth while in the control pasture the values were similar with depth. In the case of 11 years old *Ap* pasture differences in soil pH between the two sites were not found, except for depth and the interaction site/depth ($P \le 0.001$). The pH at the lowest soil depth was higher in the legume based pasture.

Acidity was less pronounced at 15-30 cm of soil depth compared to the other depths; but the sites *5* and *8 years* under *A. pintoi* rangelands showed lower pH values with respect to the rest of the sites (5.4). In the case of the *control* site (native pasture) differences among pH values with soil depth were less evident than in sites with *Arachis pintoi.* The averages for each depth were 5.52, 5.67 and 5.67, respectively.

4.6 Discussion

The soil pH results suggested that apparently an acidification process occurred in most of the sites covered by the forage legume *A. pintoi* compared with nearby control sites. This

effect was most evident in the first 5 cm of the soil profile and in pastures with 5 year old and 8 year old *A. pintoi* forage. However, the site covered by the legume for 11 years did not show an even greater increase in soil acidity.

Fig. 3. Changes in soil pH with depth in four native rangelands mixed with *Arachis pintoi* at different ages, compared to nearbycontrol sites, located in Veracruz, Mexico

In all cases, the lowest pH values were found in the first five centimetres depth. This fact could be related with the largest presence of legume roots in this layer (CIAT, 1991), and because the addition of litter and excreta from grazing animals. Consequently, the exchange of cations/anions is more intense at this depth. Soil pH values at 15-30 cm did not show a consistent trend in acidity. The highest pH value (6.0) was found under "11 years *Ap* site" site. Marschner *et al*. (1986) mentioned that rhizosphere pH may be different (higher or lower) than the pH of the bulk soil. The results suggested that the magnitude and direction of pH change depended mainly on soil depth. Marschner *et al*. (1986) mentioned that shifts in the cation/anion uptake ratio with the age of root zones, and different rates of depletion and replenishment of various cations and anions could be presumably the main factors responsible for these pH differences. Comparing the control sites data with the *Ap* rangelands in each site (Figure 3) soil acidification was evident mainly in the cases of 5 and 8 years old *Ap*. Soil pH values of 5.6 for native rangelands at 0-5 cm depth were also reported

by Bosman *et al*. (1990) for this region, including a site in the same research station, which are in accordance with our observations.

Several researchers have reported an acidifying effect of legumes. Noble *et al*. (1997) assessed acidification in Australian soils under *Stylosanthes* spp. They found the highest rate of acidification in an irrigated *Stylosanthes* seed production system. In a pot experiment, Yan *et al*. (1996) studied soil pH changes produced by field beans (*Vicia faba*). In a period of 45 days they observed that soil pH decreased significantly from 6.00 to 5.64.

Nyatsanga & Pierre (1973) mentioned that acidity developing in the soil from N_2 fixation will depend on whether all or only a part of the crop is removed in the harvest. If only the grain of any leguminous crop is harvested, the N_2 fixed would have a relatively small effect on the soil acidity because most of the N_2 fixed is in the seed. But in the case of a legume used as green manure or a grazed legume-based pasture, N_2 fixation would result in larger increases of acidity because the N_2 fixed will be acid-forming when completely nitrified in the soil. This could partially explain the low acidification under the 11 year old *A. pintoi* pasture since this was extensively harvested for planting material.

Considering the importance of tropical forage legumes to animal production systems, the acidifying effect is an important subject. Several options have been proposed to overcome this problem. Application of lime is probably the most popular recommendation, but in economically depressed areas, this strategy may not be a viable option, even if transportation difficulties were overcome. In Veracruz, Mexico, farmers known that lime application has only short-term effects on soil acidity because of high rainfall and high temperatures. More investigation into the subject is necessary.

4.7 Conclusions

The presence of *Arachis pintoi* in grazed rangelands resulted in a decrease of soil pH in several rangelands. This acidification was strongest in the topsoil (0-5 cm). Also, soil acidification was most pronounced in 5 and 8 year old *Ap* rangelands. Because of the limitations in the experimental design, these are preliminary results and further investigation will be necessary to establish the acidification problem and devise possible solutions for legume-based tropical rangelands.

5. Effect of stocking rate on soil properties in a native pasture of the humid tropic of Veracruz, Mexico

5.1 Alternatives to intensification of pasture management

In the search of alternatives for improving animal production, intensification of the system is proposed, in order to meet the demand for food of animal origin for the population. Unfortunately, this intensification has led to inadequate management of agroecosystems , resulting in deterioration of physical, chemical and biological soil properties that affect the productivity of the system to the detriment of the animals reliant on soil nutrients for plant growth (Lal, 2000).

The stocking rate as grazing strategy has been developed to control the use of pasture, in order to obtain an optimal response of vegetation to grazing (Mousel *et al*., 2005). Thus, a more

efficient management of the stocking rate could significantly improve the nutrient use efficiency in rangelands, resulting in an improvement in system productivity (Dubeux, 2005).

In the humid tropic of Mexico, studies concerning the effect of stocking rate on physical and chemical characteristics of soil are scarce.

5.2 Details of the site and experimental area

The work began in September 2005 and ended in August 2007, in the Center for Teaching, Research and Extension in Tropical Animal Husbandry (CEIEGT) of the Faculty of Veterinary Medicine and Animal Science of the National University of Mexico. Details about this site had been described previously in this Chapter.

We used a native pasture (*Paspalum* spp and *Axonopus* spp) area; since February 2002, it has been under rotational grazing with a pattern of use-rest of 3-27 days and a stocking rate of 2, 3 and 4 cows/ha. The treatments were stocking rates of 2 (low), 3 (average) and 4 (high) cows/ha, each housed in 5.0, 3.3 and 2.5 ha, where each area is divided into 10 paddocks.

5.3 Variables measured

In 100 quadrats of 0.25 m², we estimated, by direct observation, the percentage of bare soil. Two samples per paddock (replicates) in two cycles of grazing per season, within two years of evaluation were considered, which produced a total of 144 samples for the three treatments.

The length of roots ($mm/cm²$) and root density ($mg/cm²$) was measured in the last two cycles of grazing periods of rainy, dry and north season of the second year (Sept-2006/Jun-2007). We used a metal cylindrical tube of 10 cm of diameter and 20 cm long (Rowell, 1997). Eighteen samples were obtained by repetition, randomly at depths of 0-10 cm and 10-20 cm, respectively. After sampling, the soil was placed in trays to wash with water and retrieve the root in a 2 mm mesh sieve. Subsequently, the fresh root sample was placed in an oven for forced air drying at 60 ° C for 72 h, and then weighed to obtain the dry weight of the sample. Values are expressed in terms of organic matter of roots. The root length was determined by the linear intercept method (Tennant, 1975). The data were expressed in terms of mg of root root/cm3 (density) and mm/cm3 (length). The number of samples per repetition was 12, at two soil depths (0 - 10 and 10 to 20 cm), in the three seasons of the year, resulting in a total of 432 samples. Bulk density (BD, g/cm^3) was calculated at a depth of 0 -5 cm, using a cylindrical metal auger of 5 cm in diameter. Ten samples were taken for treatment in each of the two repetitions, in the three seasons and for two years, making a total of 360 samples. The samples were weighed fresh and then dried at 100 ° C for 72 h, to obtain the dry weight of soil, and with the volume of the cylinder to calculate the BD (weight/volume). To determine soil compaction (kg/cm2), 400 measurements were made randomly per repetition by stocking rate, in a single sampling in the rainy season (October) 2007 before grazing, using a penetrometer (Dickey-John Soil Compaction Tester), which produced a total of 2400 observations.

The rate of soil nitrogen mineralization was determined by anaerobic incubations of soil samples, collected at the season of the "nortes" (January 2007) at depths of 0-5 and 5-15 cm, using the technique of Waring & Bremner (1964) with slight modification, where we use 10 g of soil instead 5 g; as well as, a solution 3 M KCl to rinse the test tubes, replacing 2 M solution.

5.4 Experimental design and statistical data management

The experimental design was completely randomised. The analysis of variance was done considering the effect of treatment, repetition and the season. We used the PROC MIXED procedure of SAS (1999) for repeated measures and considered the covariance structure of symmetrical components, as the best fit to the data (Little *et al*., 1998). The comparison of treatment means was performed using LSMEANS (SAS, 1999). In all cases, the variation between replicates was used to generate the experimental error. In the case of the N mineralization rate, we considered the effect of treatment, repetition and soil depth; and likewise, the data of soil compaction $\frac{\text{kg}}{\text{cm}^2}$ were analyzed per treatment and repetition, with the PROC GLM (SAS, 1999).

5.5 Results and discussion

5.5.1 Root density and root length

The density and root length were not affected by the stocking rate (P> 0.05) at any level of soil depth (Table 6). However, the season affected (P <0.05) the density and root length in both soil depths (P <0.05). Thus, the dry season was detrimental to both soil depths, compared with the other seasons. Results of Chen *et al*. (2006) showed that root density decreased with increasing stocking rate, but the presence of moisture encouraged root growth. This coincides with the results obtained here, where the seasons wetter roots generated higher densities.

The root length was not affected by stocking rate for any of the two soil depths, but in the first 10 cm, there was a trend of greater length in the highest stocking rate, compared with the low stocking rate. On the coast of Texas in a pasture with common Bermuda grass and Coastal Bermuda grass, Rouquette, and Florence (1986) reported that the increase in stocking rate (high, medium and low) in terms of forage allowance affected the root mass in these grasses at 350, 460 and 477 kg / ha of roots. Also, no difference in root length, by effect of stocking rate could be related to the presence of hardpan at shallow depths (<30 cm), which could have affected the expansion (length and density) of roots in the soil.

Different letters for season or stocking rate within column are statistically different ($P\leq 0.05$).

Table 6. Effect of the stocking rate and year season over root density and root length (mean \pm standard error) in grazed native rangelands, in the humid tropics of Mexico.

5.5.2 Soil physico-chemical changes

The experiment did not detect any significant change in the soil physical and chemical properties between the beginning and end (Table 7), except the organic matter. The soil organic matter increased from the low to medium stocking rate, and decreased in the high stocking rate. Beare *et al*. (2005) mentioned that the SOM does not necessarily increase with increasing dry matter production, which is consistent with the results observed here.

Means with different letters within rows are significantly different ($P\leq 0.05$).

Table 7. Effect of the stocking rate over chemicals and physical variables, in a soil covered by native rangelands, in the humid tropic of Mexico.

5.5.3 Bulk density and soil compactation

In both periods, we observed that increasing the stocking rate tended to increase the bulk density, averaging 1.27 ± 0.01 , 1.3 and $1.3 \pm 0.02 \pm 0.01$ for 2, 3 and 4 cows/ha, although we found no effect on it throughout the experiment. The pooled bulk density data were fitted to the linear equation y = $0.019x + 1.23$ (R² = 0.83), which indicates that every unit increase in stocking rate (x) increased the bulk density (y) to 0.019 g/cm3. Castillo *et al*. (2003), who measured the effect of native grass, alone and associated with *A. pintoi*, three and a half years of its establishment, under similar soil and climatic conditions, reported an average value of bulk density of 1.21 g/cm^3 , slightly lower than reported in this trial for any stocking rate or season.

5.5.4 Bare soil

The evaluation of bare soil indicated significant differences due to the stocking rate (P \leq 0.0001) and season (P \leq 0.0001), as well as their interaction (P \leq 0.0001, Table 8). The proportion of bare soil increased as the stocking rate increased. With increased stocking rate, the return of the animal at a given point of the pasture is more frequent (Stewart, 2003). Another aspect that could explain this difference is related to the increased presence in the rainy season of introduced grasses, which may favor the bare soil due to its erect growth habit, compared with stoloniferous species (Barrios *et al*. , 2004).

5.5.5 Soil nitrogen mineralization rate

A trend of increased mineralization as the stocking rate increased was observed (Table 9), but the effect was not significant (P > 0.05) for this variable.

The equation $y = 68.09 + 19.59 \ln(x)$ (R² = 0.51), described the change in the mineralization of N (y) in the 0-5 cm soil depth, which increased with the stocking rate (x) , while in the 5-15 cm soil depth, the equation was $y = 21.29 + 9.55 \ln(x)$ (R² = 0.77). The highest rate of soil N mineralization was observed from 0-5 cm, and decreased with soil depth. These results match those of Valles *et al*. (2008) who found the highest rate of soil N mineralization within the first 5 cm of soil depth, although their values were lower $(5.8 - 18.7 \text{ mg NH}_4\text{-N g/soil})$ than the values measured in this study.

In this experiment, the increase in the rate of N mineralization with increasing stocking rate was consistent with other reports. Dubeux *et al*. (2006), who evaluated the effect of three levels of N from litter of *Paspalum notatum* (40, 120 and 360 kg N/ha) with three stocking rates (1.3, 2.7 and 4.0 animal units/ha), also found that the N mineralization rate increased with increasing stocking rate.

Different letters for the season x stocking rate interaction indicate significant differences ($P\leq 0.05$).

Table 8. Bare soil (%, mean ± standard error) in a native rangeland during three climatic seasons and three stocking rates, in the humid tropic of Mexico.

Table 9. N Mineralization rate (μ g NH₄-N g soil/day, means ± standard error) in a native pasture soil, at three stocking rates and two depths in the humid tropic of Mexico.

5.6 Conclusions

There was no effect of stocking rate on most variables measured. Soil compaction did not change due to stocking rate. The length and root density did not change, however, the trend of increased root length coincided with the increasing trend in the rate of soil N mineralization with increasing stocking rate, suggesting a more dynamic nutrient cycling in pastures with high stocking rate. It is possible that the absence of differences indicate variability in the response of native rangelands to cattle grazing and trampling activities. There may be sufficient, inherent adaptation in native rangelands to offset the negative effect of increased stocking rate at the levels evaluated in this study.

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