Biological indicators of soil quality in natural and cultivated subtropical systems

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ABSTRACT

The objective of this work was to evaluate the effects of forest conversion into agricultural land on some biological indicators, and to assess their relationship in subtropical ecosystems. The experimental design consisted of randomized complete blocks, with four treatments: subtropical rainforest (F), yerba mate crops (I) (Ilex paraguariensis SH.), citrus crops (C) (Citrus unshiu Marc.) and tobacco crops (T) (Nicotiana tabacum L.). Soil samples were taken at 0-0.10, 0.10-0.20, 0.20-0.30m depths. The measured variables included: acid phosphatase activity (APA), clay content, pH, total nitrogen (N), available phosphorus (P), respiration (RE) and soil organic carbon (SOC). These soils showed acid reaction and their clay content was over 650 g kg⁻¹. Contents of SOC and N were higher in soils under the subtropical rainforest, intermediate under citrus crops, lower under tobacco and yerba mate crops. The highest APA was found in the subtropical rainforest and it decreased in the three depths. In all treatments, APA was higher in the superficial layer; the 76% of APA variability was explained by N and P. Acid phosphatase activity can indicate changes in soil quality, when comparing the subtropical rainforest to the agricultural systems. It does not indicate effects among soils under different crops. Our data suggest that acid phosphatase activity is closely associated with soil organic and nitrogen content as an energy source.

Keywords: Land-use changes; Acid phosphatase enzyme; Soil respiration; Soil organic carbon; Subtropical soil quality.
1. INTRODUCTION

Soil biological parameters are of great value as sensitive indicators of soil transformations occurring under different management practices [13]. Enzyme activity is crucial in soil biological transformations, enzymes act as sensors, since they integrate information, on the one hand, from microbial status, and on the other hand, from soil physico-chemical conditions [1].

Measurements of enzyme activity have been used with different purposes in topic-related studies: as productivity indicators; as indirect measurements of the microbial biomass; to compare effects in the rhizosphere; as indicators of the soil capacity to decompose different organic materials (e.g. compost, organic residues, activated mud); and as indicators of likely contaminations with heavy metals or pesticides [8].

Soils that have been managed to promote soil quality (e.g. minimum tillage, organic amendments, crop rotations, etc.) should have a higher biological activity and therefore greater enzyme production [3]. Soil organic matter contains a variety of organic P compounds, such as inositol phosphate, nucleic acids, and phospholipids; these compounds should be first converted to inorganic phosphate by soil enzymes before being used for plant growth. Phosphatase enzymes are produced by soil microorganisms, mycorrhizal fungi, or excreted by the roots of plants [12]. They play a key role in organic P mineralization of acidic soils from tropical and subtropical regions, typically poor in plant-available P and have become a promising variable to estimate soil quality [6]; [17]. They are inducible enzymes, and the intensity of their release by plant roots and microorganisms is determined by their requirement for phosphates [12].

Most studies on soil enzymology have particularly focused on surface soils, where enzyme activity is expected to be higher [27]; [24]. Very few studies however have examined the influence of soil depth on enzyme activity, such as the case of the one performed by [25], in Oxisols and Ultisols from India, under tea cropping and forestry culture.

Soil management practices influence soil microorganisms and microbial process through changes in the quantity and quality of plant residues in the soil profile [11]. Similarly [5] concluded that phosphatase activity, in kaolin clay Oxic Haplustalf, was affected by the addition of plant residues, enhancing enzyme activity in soils under conservation tillage compared to conventional tillage. This was associated with increased microbial biomass activity. [2], observed that acid phosphatase activity (APA) diminished as soil organic carbon (SOC) in forested soils from Costa Rica, with the highest total phosphorous concentration in tropical Rain Forest Soils [22]. The aim of this work was to evaluate the effects of forest conversion in to agricultural land on some biological indicators, and to assess their relationship in subtropical ecosystems.

2. MATERIALS AND METHODS

2.1. Study site

The assay was performed in Oxisols soils in the province of Misiones, northeast of the Argentine Republic, in four study areas: Dos Arroyos (27º36'38"S; 55º 17'W), Department of Alem; Oberá (27º28'43"S; 55º 07'W), Department of Oberá; Dos de Mayo (27º01'28"S; 54º 38'30"W), Department of Cainguás; and El Soberbio (27º14'47"S; 54º 20'26"W), Department Guarani.

The sampled sites are characterized by a humid subtropical climate, with an isohygric rainfall regime without a dry season. The soils were classified as Rhodic Eutrudox, pertaining to the fine clayey hyperthermic family; they are well drained, clayey and very deep soils that offer good physical conditions for root development.

2.2. Experimental design

Each study area was considered as a block. The treatments included subtropical rainforest (F), the cultivations of yerba mate (I), citrus (C) and tobacco (T).

The natural system corresponded to the subtropical rainforest treatment without anthropic alterations; the vegetation was tropical, mostly consisting of very tall trees, and a great deal of lianas and epiphytic plants. It as a taken was reference for high soil quality.

The I treatment corresponded to farmers´owned plots cultivated with yerba mate (Ilex paraguariensis, Saint Hill,) of 15 years with a density of 2,220 plants ha⁻¹, under conventional management practices and mechanical weed control using a disk harrow.

The C treatment corresponded to farmers´owned plots cultivated with Satsuma (Citrus unshiu Marc.), of 8 yr of age of the Okitsu variety, with a density of 660 plants ha⁻¹. Weed control was performed with weeding machines in the streets with herbicides in the rows. In this treatment, phosphate fertilizers were used (55 kg ha⁻³ of P per year).

The T treatment corresponded to farmers´plots cultivated annually with tobacco (Nicotiana tabacum L.), with under regional conventional management practices was described in [21]. All of them had a 15-to 20-yr history of previous agricultural use.

2.3. Soil sampling and variables measured

Three plots of 15 by 51 m were selected per block for each treatment. In each plot, three composite soil samples (by three individual samples) were collected to three depths: 0 to 0.10, 0.10 to 0.20, 0.20 to 0.30 m by using simple random sampling. The extracted soil samples were air-dried, manually ground in a mortar, sieved through a 2 mm mesh, and dried in an oven at 105°C for 24 h to determine gravimetric water content. Several edaphic variables were measured: APA by
colorimetric estimation of the p-nitrophenol released by phosphatase activity, when soil samples were incubated (37°C) with buffered (pH: 6.5) sodium p-nitrophenyl phosphate solution toluene; the p-nitrophenol released was extracted and determined using a spectrophotometer at 410 nm, according to the method of [20]. The enzymatic activity was expressed as micrograms of p-nitrophenol released by grams of air-dried soil during one hour of incubation. Clay content was measured according to Bouyoucos’ method [7], the pH was measured using a potentiometric method in 0.1M KCl solution (relation 1:2.5) [23], soil organic carbon as described by Walkley-Black (SOC) [4], total nitrogen [7], available phosphorus (P) by Bray Kurtz II [7], respiration (RE) [19].

2.4. Statistical Analysis

All data were analyzed through the analysis of variance (ANOVA), to assess the effects of land-use changes and soil depth. Comparisons between means for treatments and depth were performed using Duncan’s multiple range tests (p<0.05). To evaluate the relationship between variables, a simple correlation analysis between pairs of variables was performed, along with a multiple linear regression analysis, using the stepwise method to select the appropriate model. Selection criteria were the less mean square error (MSE) and Mallow’s Cp. The significance level for variable inclusion in the analysis was a P value of less than 0.13.

3. RESULTS AND DISCUSSION

The soils analyzed were rated as clay soils in terms of their texture, with a clay content of over 650 g kg⁻¹. The mean surface content was 740 g kg⁻¹ in the subtropical rainforest, 653 g kg⁻¹ in the citrus cropping, 671 g kg⁻¹ for the tobacco cropping and 779 g kg⁻¹ for the yerba mate cropping. These soils showed an acid reaction in the three assayed depths, with mean pH values between 3.66 and 4.36. The SOC and N contents were higher in soils under the subtropical rainforest, intermediate in soils under citrus cultivation and lower in soils under tobacco and yerba mate cropping (Table 1). Furthermore, these variables showed values that decreased with depth throughout the soil profile in all cases, showing significant differences (p<0.0001). Soil organic carbon (SOC) showed mean ranging from 18.9 to 40.2 g kg⁻¹ at the surface, which diminished across depths and reached mean values between 12.4 to 16.9 g kg⁻¹ in the 0.2-0.3 m depth (Table 1). Continuous cropping favoured fast mineralization of soil organic matter thereby altering the original condition.

Acid phosphatase activity was higher in the superficial layer for all the treatments and, as well SOC content, declined with depth. In the case of the subtropical rainforest, there were significant differences between the first depth and the rest. As to yerba mate and tobacco crops, there were highly significant differences among the three depths, in citrus crops, important differences were found between the surface and the third depth (Table 1). This was attributed to change in organic matter composition as a result of soil management practices, which greatly affected enzyme activity, in coincidence with reports by other authors [17]; [25]. The highest enzyme activity was found under the subtropical rainforest for the three depths, in comparison with the other agricultural uses, showing significant differences. The highest value of APA was found in the first 0.10 m depth of the subtropical rainforest. The root system in P-deficient soils enhances phosphatases secretion [15], which account for such differences, as a result of large amount of roots in the first 0.10 m of the subtropical rainforest, in comparison with soils under different crops.

The average soil respiration value was higher in the subtropical rainforest as compared with the cultivated soils (r=0.36; p<0.0227). The coefficient of variation was 40%, similar to the one found by [18], attributable to the great variability of such parameter-associated with other factors, including temperature and humidity. The higher respiration in subtropical rainforest soils shows there is more biological activity resulting from greater availability of substrat for soil flora and fauna. The forest conversion affected the soil respiration and acid phosphatase as reported by [26].

A significant direct correlation was found between APA-SOC and APA-N in the three depths assayed (Table 2). Acid phosphatase activity was closely linked to soil organic carbon content, since microbiological activity is directly related to organic matter content [25]. Similar results were obtained by [16], in Oxisols from Venezuela, where APA correlated significantly with SOC and with N, by [10] , in Oxisols of Brazil, and by [9], in Oxic Argudolls from the province of Chaco. A positive correlation was also found between APA and RE (Table 2). This is attributable to the fact that a higher availability of organic substrates enhanced biological activity [25], which is reflected in a higher respiration rate.

In the multiple linear regression analysis, the selected stepwise model (Table 3) produced the following equation:

\[ \text{APA}=1588.89 \times N - 34.07 - 3.96 \times P \ (R^2=0.76). \]

The 76% of APA variability was explained by total nitrogen, which is closely related to soil organic matter, and by available P, although total nitrogen showed a higher level of significance (p<0.0001). Similar results were obtained by [14], where available P deficiencies and N applications increased APA. Others authors [26] , suggest soil enzyme activity is partly controlled by soil C availability because soil microbial activity utilizes SOC as C source and releases CO₂ to the atmosphere, thus increasing C emission from the soil to the atmosphere and reducing soil C storage.

Therefore, the activity of this enzyme is not related solely P availability, it also depends on rapidly degradable energy sources soil N, attributing to soil N a positive effect on the increase of the synthesis of the enzyme phosphatase by soil microorganisms and plants [14].

4. CONCLUSIONS

4.1. Removal of the subtropical rainforest and the subsequent incorporation of soils to agricultural production led to a reduction in the organic content of these soils and to lower biological activity, resulting poorer soil quality.
4.2. Acid phosphatase enzyme activity is higher in the top 0.10 m soil layer, decreases along the profile depth, and is closely associated with soil nitrogen and organic carbon contents.

4.3. Acid phosphatase activity can detect changes in pristine conditions and in agricultural soils, but it fails to detect differences among the different agricultural uses.

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Means followed by the equal minuscule letters, in the row, and capital letters in the column, do not differ by Duncan test, at 5% of probability. *, **, *** Significant at 5, 1 0.01% probability for F value.

Data of C, APA the Tobacco were published in [21].

Table (1): Mean contents of soil organic carbon, available P, total nitrogen, respiration acid phosphatase activity of soils under subtropical rainforest, tobacco, citrus yerba mate croppings (1).

| Depth (m) | Subtropical rainforest | Tobacco | Citrus | Yerba mate | Variation Coefficient (%) | F Value |
|----------|------------------------|---------|--------|------------|---------------------------|---------|
| 0-0.10   | 40.2c                  | 21.6ab  | 23.5b  | 18.9a      | 15                        | 72.98***|
| 0.10-0.20| 21.6b                  | 16.2a   | 17.5a  | 17.2a      | 9                         | 28.40***|
| 0.20-0.30| 16.9d                  | 13.5b   | 12.4a  | 14.8c      | 7                         | 38.55***|

Available P (mg kg⁻¹)

| Depth (m) | Subtropical rainforest | Tobacco | Citrus | Yerba mate | Variation Coefficient (%) | F Value |
|----------|------------------------|---------|--------|------------|---------------------------|---------|
| 0-0.10   | 4.59a                  | 6.48a   | 12.67a | 3.82a      | 62                        | 10.44***|
| 0.10-0.20| 2.33b                  | 1.86b   | 0.98b  | 1.93b      | 42                        | 6.87**  |
| 0.20-0.30| 1.51c                  | 0.98b   | 0.44b  | 1.20bc     | 43                        | 12.06***|

Total nitrogen (g kg⁻¹)

| Depth (m) | Subtropical rainforest | Tobacco | Citrus | Yerba mate | Variation Coefficient (%) | F Value |
|----------|------------------------|---------|--------|------------|---------------------------|---------|
| 0-0.10   | 4.7c                   | 2.0a    | 2.4b   | 1.9a       | 18                        | 89.36***|
| 0.10-0.20| 2.7c                   | 1.5a    | 1.9b   | 1.6ab      | 15                        | 36.41***|
| 0.20-0.30| 2.0c                   | 1.3a    | 1.5b   | 1.4ab      | 11                        | 38.59***|

Respiration (kg ha⁻¹ of CO₂)

| Depth (m) | Subtropical rainforest | Tobacco | Citrus | Yerba mate | Variation Coefficient (%) | F Value |
|----------|------------------------|---------|--------|------------|---------------------------|---------|
| 0-0.10   | 47.32b                 | 38.31a  | 33.37a | 30.06a     | 40                        | 3.03*   |

Acid phosphatase activity (µg of p-nitrophenol g⁻¹ soil ha⁻¹)

| Depth (m) | Subtropical rainforest | Tobacco | Citrus | Yerba mate | Variation Coefficient (%) | F Value |
|----------|------------------------|---------|--------|------------|---------------------------|---------|
| 0-0.10   | 684.24bA               | 275.5aA | 270.2aA| 251.9aA    | 29                        | 48.86***|
| 0.10-0.20| 281.65bB              | 207.7aB | 221.6aA| 184.2aB    | 23                        | 7.95**  |
| 0.20-0.30| 192.26bB              | 176.7abB| 147.0aB| 144.7aC    | 20                        | 5.90**  |

CV F value (34) 48.9*** (12) 46.3*** (28) 13.0*** (17) 31.2*** - -

(1) Means followed by the equal minuscule letters, in the row, capital letters in the column, do not differ by Duncan test, at 5% of probability. *, **, *** Significant at 5, 1 0.01% probability for F value.

Data of C, APA the Tobacco were published in [21].
Table (2): Correlation matrix (coefficients, probabilities) between acid phosphatase activity with soil organic carbon (SOC), available P, total nitrogen respiration. N = 48.

| Depth (m) | SOC  | Available P | Total nitrogen | Respiration |
|-----------|------|-------------|----------------|-------------|
| 0-0.1     | 0.82*** | -0.18       | 0.87***        | 0.31*       |
| 0.1-0.2   | 0.48**  | 0.22        | 0.67***        |             |
| 0.2-0.3   | 0.32*   | 0.41        | 0.56***        |             |

*, **; ***Significant at 5, 1, 0.01% probability.

Table (3): Parameter estimates (St.) standard error (SE) of intercept of the multiple linear regression between acid phosphatase activity (APA) total nitrogen (N), available phosphorus (P). Stepwise selection. Total variables: 8; variables in the model: 2.

| Coefficient | St.  | SE    | p-value   | Cp Mallows |
|-------------|------|-------|-----------|------------|
| Intercept   | -34.07 | 44.74 | 0.4504    |            |
| N           | 1588.89 | 134.89 | <0.0001   | 137.75     |
| P           | -3.96   | 2.39  | 0.1051    | 4.70       |

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