European Journal of Soil Biology 87 (2018) 17–24

Contents lists available at ScienceDirect

European Journal of Soil Biology

journal homepage: www.elsevier.com/locate/ejsobi

Phosphorus fractions in subtropical soils depending on land use

Menuka Maharjan[a,b,*], Deejay Maranguit[a,c], Yakov Kuzyakov[d,e,f]

[a] Department of Soil Science of Temperate Ecosystems University of Göttingen, Göttingen, Germany
[b] Institute of Forestry, Tribhuwan University, Hetauda, Makwanpur, Nepal
[c] Department of Soil Science, Visayas State University, Baybay, 6521-A Leyte, Philippines
[d] Department of Agricultural Soil Science, University of Göttingen, Göttingen, Germany
[e] Institute of Environmental Sciences, Kazan Federal University, 420049 Kazan, Russia
[f] Agro-Technology Institute, RUDN University, Moscow, Russia

ARTICLE INFO

Handling editor: Yakov Kuzyakov

Keywords:
Land-use change
Phosphorus fractions
Carbon-to-phosphorus ratio
Acid phosphatase activity

ABSTRACT

Land-use change from forest to agriculture, which is driven by the demands of sustaining the growing global population, affects nutrient dynamics and availability in soil. Although phosphorus (P) is one of the main limiting nutrients in agricultural production, little is known about the influence of soil microorganisms on the dynamics of P cycling in subtropical land use systems. The objective was to assess the impacts of land use (organic farming, conventional farming and forest) on forms and distribution of P in soil.

After conversion of forest, the P stock significantly increased by 373% and 170% in soil under organic farming at 0–10 and 10–20 cm depth, respectively. In conventional farming, the P stock increased by 64% and 36% at 0–10 cm and 10–20 cm depth, respectively compared to forest. The larger (up to 4 times) fraction of organic P (Po) than inorganic P (Pi) implies that total P is regulated by organic P. Easily-available P fractions (microbial biomass P, $NaHCO_3$-Pi and Po), moderately available P (NaOH-Po) and non-available P (HCl-Pi and Po) were much higher in organic farming than conventional farming and forest, especially at the 0–10 cm depth. Compared to organic farming, the higher (> 100) C_{org}: Po ratio that soils under conventional farming and forest are P limited which correspond with higher (2–8 times) activity of acid phosphatase in conventional farming and forest. Concluding, land use and management practices i.e. crop rotation, residue input and farmyard manure application significantly increase different fractions of P in organic farming.

1. Introduction

Land-use change, such as conversion of forest to intensively managed agriculture, is the largest global change of the last two centuries due to the increasing demands of feeding the growing human population [1]. During the period of 1980–2000, approximately 50% of the new arable land came from intact forest, while 28% came from disturbed forest in the tropics [2]. Furthermore, land-use change significantly alters the physical, chemical and biological properties of soil, affecting soil fertility and ultimately reducing the capacity of land for sustainable crop production [3].

Land-use change has substantial effects on phosphorus (P) availability for plant uptake by increasing P losses or P transfer into recalcitrant pools, leading to significant alterations in P distribution and availability [4]. The main sources of soil P are either parent material or application of mineral and/or organic fertilizers [5]. Other than N, P is considered as the largest globally limiting nutrient for food production [6]. P availability may be limited due to 1) inherent characteristics of the parent material 2) strong sorption of PO_4^{3-} to Al and Fe (hydro) oxides or 3) a low input of inorganic and organic fertilizers [7]. Plants and microorganisms have developed a broad range of mechanisms to enhance the acquisition of P, e.g., production of phosphatase enzymes, which are responsible for hydrolyzing recalcitrant forms of organic P to make it available to plants [8]. Plants can only produce acid phosphatases whereas microbes have capacity to produce both acid and alkaline phosphatases [9]. Microorganisms play a vital role in P mineralization from various organic sources [10] and transformations of soil organic P [11]. Thus, soil microorganisms are a key pool not only of C and N, but also of P. Furthermore, a majority of the P held in microorganisms can be released quickly and be readily available for plant growth [12]. The activity of phosphatase is negatively correlated with availability of inorganic P [13]. Previous studies have examined the effects of land-use change on P forms, distribution along soil depth, availability for plants and long term stability as a consequence of forest conversion to monoculture plantations [4] mineral and manure fertilization [14], tillage system [15] and cropping/vegetation systems

* Corresponding author. Department of Soil Science of Temperate Ecosystems University of Göttingen, Göttingen, Germany.
E-mail address: menuka48maharjan@gmail.com (M. Maharjan).

https://doi.org/10.1016/j.ejsobi.2018.04.002
Received 21 December 2017; Received in revised form 18 April 2018; Accepted 18 April 2018

[16]. These studies highlight that the status and distribution of P depends on land use and management practices. Although the effects of management practices on P fractions are intensively discussed, knowledge of the linkage between microbiological processes and P fractions is limited.

The study site in "Chitwan district" lies in the Terai region, a plain in southern Nepal. Most of the studies in Chitwan focused on the effects of land-use change on above and below ground carbon content and stocks [17]. However, studies focused on various P pools and linking the response of microbe-induced P availability to different land use systems is absent. Beside, phosphatase activity which is highly influenced by land use system plays key role for mobilizing the organically bound P. Thus, this calls for evaluation on the effects of land use on P forms and distribution in subtropical soils. The objective was to assess the impacts of three land use systems, i.e. organic farming, conventional farming and forest (litter are collected by villagers), on the forms and distribution of soil P. We hypothesized that land use affects the availability of P fractions (inorganic and organic) and acid phosphatase activity.

2. Materials and methods

2.1. Site description

The study was carried out in Chitwan district (27°35′N 84°30′E) of Nepal. The climate is subtropical with annual rainfall of 1763 mm. The mean temperature is 22 °C and annual average temperature is 30 °C. Three land-use systems were selected (Table 1): forest, organic farming and conventional farming. Both farming sites were located in Fullbari Village Development Committee (VDC) and the forest site in Patihani VDC. The soils are Gleyic Cambisols (organic farming and forest) and Eutric Cambisol for the conventional farming site [18]. The soil texture at all sites is sandy loam. The organic farming site has been under organic farming practices for 15 years. The agricultural site has been under conventional farming practices for 25 years. The crop rotations are maize + rice + vegetables/mustard, and maize + rice + wheat/lentils for the organic and conventional farms, respectively (Table 1). The organic farm was under vegetable farming during soil sampling, while the conventional farm was fallow with remaining rice stubbles. About 10 ton ha^{-1} $yr^{-1}$of farmyard manure and vermicomposting are applied in organic farming, whereas urea (60 kg ha^{-1} yr^{-1}), potassium (15 kg ha^{-1} yr^{-1}), and diammonium phosphate (94 kg ha^{-1} yr^{-1}) are added in conventional farming. Forest is dominated by the broad leaf *Shorea robusta* commonly known as Sal. The leaves of Sal are collected by local people for performing social and religious activities. Pesticides are applied only in conventional farming.

2.2. Soil sampling and preparation

To assess the effects of land use on P fractions, soils from three land use systems (with four replication for each land use) were sampled from 0-10 and 10–20 cm depth. Plant remains, debris and roots were removed using tweezers. The samples were kept cold (at 4 °C) prior to analysis.

22 grams of air-dried soil from each land use system was placed into 100-ml jars. The soil was adjusted to 70% of the water holding capacity (WHC) and pre-incubated for 14 days at 22 °C prior to sequential extraction to restore equilibrium following the disturbance of drying and sieving [19].

Table 1
Description of soil properties (Ap/Ah horizon).

Land use	pH (H_2O)	Carbon (mg C g^{-1})	Nitrogen (mg N g^{-1})
Organic farming	7.5	21	1.9
Conventional farming	5.0	15	1.2
Forest	5.5	9	0.7

2.3. Sequential P fractionation

Various organic and inorganic P fractions were determined in soil *via* the Hedley et al. [19] sequential fractionation method with the modifications of Tiessen and Moir [20]. This method uses a sequence of strong extractants that remove labile Pi and Po, followed by the stable P forms Tiessen et al. [21] (Fig. S1). Hedley fractionation is based on the analysis of inorganic phosphorus (Pi) and organic phosphorus (Po) fractions of different availability and chemical binding ability by utilizing extractants of increasing strength [19] [22]. The following fractions are extracted (i) readily available P for plants, i.e., $NaHCO_3$-Pi ii) easily mineralizable P, i.e., $NaHCO_3$-Po and microbial biomass P (iii) strongly adsorbed P by aluminum (Al) and iron (Fe) oxides i.e. NaOH-P (iv) P associated with calcium (Ca-P), considered as non-available P to plants i.e. HCl-P.

Microbial biomass P (MBP) was determined by chloroform fumigation-extraction [23] [24] modified by Yevdokimov and Blagodatskaya [25]. Briefly, approximately 3 grams of soil was placed into a 50-ml centrifuge tube filled with 30 ml of deionized water for both fumigated and non-fumigated samples. For fumigated samples, 300 µl of chloroform was added to the sample. Both fumigated and non-fumigated samples contained one anion exchange membrane (AEM) strip and were kept for 24 h on an end-to-end mechanical shaker. After shaking, AEM strips were removed and washed three times by gently submerging the strip into deionized water. The AEM strips were subsequently immersed into centrifuge tubes filled with 45 ml of 0.25 M H_2SO_4 and were shaken for three hours. Finally, phosphate was measured in the extracts *via* the malachite green colorimetric method of D'Angelo et al. [26] modified by Yevdokimov and Blagodatskaya [25]. Following MBP extraction, the soils were further extracted sequentially for the rest of the P pools. Briefly, 3 grams of soil was placed into a 50-ml centrifuge tube and extracted with the following extractants in sequential order: (i) 30 ml 0.5 M $NaHCO_3$ at pH 8.5, which extracts relatively labile Pi and Po (ii) 30 ml 0.1 M NaOH, which extracts Fe and Al bound P iii) 30 ml 1 M HCl, which extracts Ca bound P. After addition of extractants, samples were shaken for 16 h and the soil suspensions were centrifuged at 3500 rpm for 15 min. The resulting supernatants were filtered using Whatman no. 42 filter papers and stored in small vials at 4 °C for P measurement.

2.4. Determination of phosphate

For Total P, 5 ml aliquots of each extract were digested by ammonium-persulfate and H_2SO_4 to oxidize dissolved Po to Pi forms. TP was determined as the concentration of soluble reactive P [27]. For Pi, 5 ml aliquots of each extract were digested by H_2SO_4. Po was calculated as the difference between the TP and Pi.

For the measurement of Pi, a 150 µl aliquot from each extract was added to a disposable 96-well polystyrene microplate. 30 µl of Reagent 1 (14.2 mmol L^{-1} ammonium molybdate tetrahydrate + 3.1 M H_2SO_4) was subsequently added to each sample and the microplate was shaken for 10 min. After shaking, 30 µl of Reagent 2 (aqueous polyvinyl alcohol + deionized distilled water + MG carbinol hydrochloride) was added and the microplate was shaken for an additional 20 min. After shaking, microplates were incubated at 40 °C for 40 min. Incubated microplates were read on a *Victor* microplate reader at 630 nm. The microplate was again read after 12 h for evaluation of the stability of measurements. Simultaneously, standards were prepared with the same extractants as used for extraction of phosphate i.e. $NaHCO_3$, NaOH, HCl and H_2SO_4. The standard varied depending on the P concentration range (D'Angelo et al. [26] modified by Yevdokimov and Blagodatskaya [25]). Values for the residual P fractions and the Po fraction of soil under organic farming at 10–20 cm were below the detection limit.