BRIEF REPORT

Differences in total iron content at various altitudes of Amazonian Andes soil in Ecuador [version 1; peer review: 2 approved]

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Abstract

Although iron is not contained by chlorophyll, it is indispensable for plants as it plays an essential role in the biosynthesis of chlorophyll. It is a component of many important plant enzyme systems, e.g. cytochrome oxidase, which is responsible for electron transport. Therefore, examining iron content of soils, particularly ionic forms of iron (Fe^{2+} and Fe^{3+}) is important for fruit growers. In this article, we disclose the total iron content determined in soils (Hyperalic Alisol soil) at three altitudes of Amazonian rainforest in Ecuador. We examine how different altitudes impact the pH and total iron content in the selected study area. We found that total iron content significantly decreases ($R^2=0.966$) at lower altitudes. For future studies, the authors recommend that along with Fe ion content one should determine calcium, microbial biomass, and microbial activity to better understand iron mobility and dynamics of iron uptake in the area.

Keywords

Iron, total iron in soil, Hyperalic Alisol, Amazonian rainforest, Ecuador

This article is included in the **Agriculture, Food and Nutrition** gateway.

Open Peer Review

Approval Status

	1	2
version 1		
20 Feb 2020	view	view
1. Zsolt Kotroczo , Szent István University, Budapest, Hungary		
2. Ankit Singla , Ministry of Agriculture and Farmers Welfare, Government of India, Ghaziabad, India		
Any reports and responses or comments on the article can be found at the end of the article.		

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Author roles: **Mendoza B:** Investigation, Methodology, Project Administration, Writing – Review & Editing; **Guananga N:** Conceptualization, Investigation, Methodology, Project Administration, Resources, Supervision, Validation, Visualization, Writing – Original Draft Preparation, Writing – Review & Editing; **Melendez JR:** Investigation, Methodology, Writing – Review & Editing; **Lowy DA:** Investigation, Methodology, Supervision, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

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Introduction

Total iron concentration of soils mainly depends on pH (Colombo *et al.*, 2014; Jelic *et al.*, 2010) and moisture content; and is also affected by root respiration, soil microbial activity, leaching, and erosion (Spectrum Analytic, Inc. 2020). Given that iron deficiency is a regular problem for various crops, it is essential to determine the total iron content of soils (Mengel *et al.*, 2001), particularly in orchards (Simon & Szilágyi, 2003).

In a highly cited review paper, Bünemann *et al.*, (2018) identify the most frequently used soil quality indicators under agricultural land use: organic matter, pH, available phosphate, and water storage. Soil quality evaluation should specify targeted soil threats, functions, and ecosystem services. The authors of the review recommend developing increasingly interactive assessment tools.

Recently, several studies have been undertaken on the effects on soil quality exerted by various minerals contained in the soil, such as ammonium lactate-soluble potassium and phosphorus content (Jakab, 2020; Li *et al.*, 2020). Also investigated was the impact of various soil cultivation methods on some microbial soil properties (Beni *et al.*, 2017; Sándor *et al.*, 2020; Sándor, 2020; Veres *et al.*, 2015).

In this article, we report the variations with altitude of the total iron content measured in intact soil in the Amazonian rainforest (in an uncultivated and uninhabited area). Considering that

orchards are the most sensitive to iron deficiency, our results are aimed to support local farmers, when they select new areas for fruit plantations. An intact area was chosen as the control for soil samples, which will serve as the reference for future studies initiated in the nearby agricultural region.

Methods

Soil sampling

A total of 15 soil samples were collected from three altitude levels: 420, 1000, and 1600 m.a.s.l. (meters above sea level) near Tena, Ecuador, on December 10, 2019, from the upper layer (top 20 cm) of Hyperalic Alisol (Ultisols in US Soil Taxonomy) soil (Table 1).

Determination of soil properties

We measured pH in distilled water for soil/water ratio of 1:25 (w/w) using a glass electrode (Model Seven2Go Advanced Single-Channel Portable pH Meter, Mettler, Toledo). Soil moisture content was determined gravimetrically; drying the soil samples at 105°C for 24 h and weighing the mass loss. We measured allophane using 10.0 ± 0.5 g soil/water (1:2, w/w), soil/water plus 20. mL 1.0 M NaF, soil/water (1:2.5, w/w) + 25 mL 1.0 M NaF, soil/water (1:2.5, w/w) + 25 mL 0.50 M NaF, as described by Singla *et al.* (2018).

We determined total iron (all ionic forms) according to modified Blakemore 1981 method described in Singla *et al.*, 2018. Briefly, 50 mL of ammonium oxalate monohydrate (Spectrum

Table 1. Soil sampling points along with main physical-chemical soil properties.

Nº Soil Sample	Latitude	Longitude	Altitude (m.a.s.l.)	pH (H ₂ O)	Allophane	Moisture content (%)
1	4.628247894396525	-74.95615214109422	420	5.34	Volcanic	54.04
2	4.628247894396525	-74.95615214109422	420	5.11	Volcanic	50.48
3	4.628247894396525	-74.95615214109422	420	5.98	Volcanic	53.11
4	4.628247894396525	-74.95615214109422	420	5.01	Volcanic	55.52
5	4.628247894396525	-74.95615214109422	420	5.55	Volcanic	52.22
6	4.641091761957411	-75.02968892455102	1000	4.99	Volcanic	45.45
7	4.641091761957411	-75.02968892455102	1000	5.42	Volcanic	45.19
8	4.641091761957411	-75.02968892455102	1000	5.90	Volcanic	44.01
9	4.641091761957411	-75.02968892455102	1000	5.65	Volcanic	46.04
10	4.641091761957411	-75.02968892455102	1000	5.34	Volcanic	43.94
11	4.65093587318055	-75.09377360343935	1600	5.28	Volcanic	58.86
12	4.65093587318055	-75.09377360343935	1600	5.13	Volcanic	60.23
13	4.65093587318055	-75.09377360343935	1600	5.04	Volcanic	60.45
14	4.65093587318055	-75.09377360343935	1600	5.45	Volcanic	66.56
15	4.65093587318055	-75.09377360343935	1600	5.43	Volcanic	64.14

Chemical) (0.20 M, pH 3) was added to 1 gram of soil sample. The mixture was shaken with a Model NB-101M Medium Orbital Shaker (N-Biotek, Inc.) in orbital mode, for 4.5 h at 150 rpm. In total 12 hours later, samples were centrifuged for 15 min at 3500 rpm (using Hermle Z400, Hermle, AG, Germany). Double filtration was performed (Whatman ^{no}42 filter). A calibration curve was determined from the extracted solution (oxalate ammonium acid 0.20 M) according to Singla *et al.* (2018). The solution was measured with a Model 240Z Atomic Absorption Furnace Spectrophotometer (Agilent) at a wavelength of 392 nm and with a slit width of 0.2 nm.

Data analysis

We applied simple linear regression (Z-test) for statistical analysis, using SPSS (version 26) to reveal possible relevant differences in pH values and total iron content at different altitudes.

Results and discussion

Examined soil samples in the chosen area were strongly or moderately acidic, with pH values in the range from pH 4.95 ± 0.05 to pH 5.95 ± 0.05 (Table 1). We did not find any meaningful correlation between altitude and pH values, or between pH and total iron content. Moisture content is the highest at 1600 m.a.s.l. Allophane was detected in all samples, which supports the volcanic nature of the sampling area (Fiedles & Perrot, 1986) (Table 1).

Total iron content significantly decreases ($R^2=0.966$) at lower altitudes (Figure 1). No significant changes in pH were found, and we can explain this by the following:

- (i) vegetation at lower lying areas receive less light, so it absorbs a greater quantity of iron ions; so far, there

is no relevant literature data on the effect of light intensity on the iron uptake of plants (Borowski, 2013).

- (ii) there is a greater concentration of iron-reducing bacteria in the lower lying areas, which seems to be verified by a prior study (Fiedler *et al.*, 2007). This finding is, however, unusual, because such bacteria are typically present in sea water (Bae *et al.*, 2001) and paddy soils (Singla & Inubushi, 2013), rather than in Hyperalic Alisol soils.

High moisture content of the soil and organic matter accumulated on the soil surface can make air circulation difficult, hence, anaerobic conditions can develop in lower lying areas.

Our results (from 400 m.a.s.l. to 1000 m.a.s.l.) are comparable with a prior study performed in the same region (Singla *et al.*, 2018), in which the authors report a decrease in iron content for lower lying areas. The main difference between our assessment relative Singla and colleagues' results is that they observed a radical decrease in iron content above 1000 m.a.s.l., while we found greater iron concentrations at this altitude. Our results are comparable in magnitude to other study findings (Fageria & Stone, 2008) carried out in South American Hyperalic Alisol soils in which high iron content was found at depths of 0–20 cm.

Conclusions

Total iron content significantly decreases ($R^2=0.966$) at lower altitudes. Genomics studies could detect possible iron consuming

Figure 1. Correlation between total iron content (mg/kg) and m.a.s.l in soil samples from Amazonian Andes soil in Ecuador.

bacterial strains. For future studies, we recommend that in addition to Fe²⁺ and Fe³⁺ content one should determine calcium, microbial biomass, and microbial activity. Altogether, this approach would enable a better understanding of iron mobility and dynamics of iron uptake in the area.

Data availability

Underlying data

Figshare: Raw data for “Differences in total iron content at various altitudes of Amazonian Andes soil in Ecuador”, <https://doi.org/10.6084/m9.figshare.11833554.v2> (Guananga, 2020).

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Reviewer Report 28 February 2020

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Ankit Singla

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Summary of the content:

It should be mentioned that Kotroczo and co-workers present novel research findings considering that currently there is no iron concentration-related study published in the literature in the sampling area that examines differences in soil iron ion concentrations at different altitudinal levels. As mentioned in the manuscript, this is especially useful for orchard-growers. The sampling area was well selected because of its specific location, as it is an inherit area, that is close to cultivated lands. The presented results may serve as control results for further agri- and/or horticultural studies. Also unexpected findings about the iron decrease may awaken researchers' attention, and/or inspire a more complex research (i.e. genomic studies) in the examination area.

Abstract: abstract is well written and informative.

Keywords: I agree with Reviewer 1 Dr. Zsolt Kotroczo that Keywords should not include those words that also could be found in the title. Please consider to replace the following keywords: *Iron*, *total iron*, *Ecuador*, providing higher possibility for search engines to find the article.

Introduction: One of the most challenged part of the evaluation process in case of Brief Report or Short Communications is the Introduction. Despite it is short, I believe that it consists relevant, and up-to-date research findings of the topic (mainly 2019-2020 content).

Methods: Methods are well written, well referenced. I agree with Reviewer 1 about it would be more fortunate to provide detailed information about the sampling area. However, it is required to clarify soil/water ratio for pH measurement was 1:25 or 1:2.5? Please check first line of determination of soil properties in the methods section.

Results and Discussion: Brief and straightforward. Consists of statistical results, supporting data is available as well.

Conclusions: Summarizes and concludes from research findings correctly.

Is the work clearly and accurately presented and does it cite the current literature?

Yes

Is the study design appropriate and is the work technically sound?

Yes

Are sufficient details of methods and analysis provided to allow replication by others?

Yes

If applicable, is the statistical analysis and its interpretation appropriate?

Yes

Are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions drawn adequately supported by the results?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Soil Microbiology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Reviewer Report 24 February 2020

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Zsolt Kotroczo

Department of Soil Science and Water Management, Szent István University, Budapest, Hungary

Comments and suggestions

Authors: Benito Mendoza, Nelly Guananga, Jesus R. Melendez; Daniel A. Lowy

Title: Differences in total iron content at various altitudes of Amazonian Andes soil in Ecuador

Summary of the manuscript:

The work contains many novelties. The objectives are well founded but I miss some well-formulated hypotheses. This is very important in such well-established and well-planned research.

The topic chosen increases the significance of the research.

The manuscript is the original research paper of the authors. The subject of the manuscript is interesting and current.

The theme of the manuscript's scope fits the aims of the journal. Most probably the manuscript will be of great interest, contributing to the reading of the journal. It is suitable for indexing in *F1000Research* journal. The basic idea of the experiment was carried out precisely, the study provided large amount of high quality data.

The **Abstract** is quite concise and sums up the essence of the manuscript appropriately. The abstract properly introduces the topic, summarizes the methods used and the results obtained. It is summarizes the essence of the manuscript.

Keywords: I suggest replacing some of the key words. The keywords that are included in the manuscript title should be replaced.

Introduction

The chapter is well structured. It fits in well with the manuscript theme, and establishes and complements the whole research topic. This chapter comprehensively supports the whole manuscript with literature. Appropriate and timely references are built in the introduction chapter. The chapter detailed and well processed.

Methods

The methods chapter are appropriate and sufficiently detailed. In my opinion, soil sampling and soil analysis methods are sufficiently detailed, but the research area description is inadequate. For better identification and understanding of the results, I suggest a brief description of the sampling area.

Results

The interpretation of results is generally proper. Results chapter is detailed. These chapters are well-structured and properly constructed. This chapters provide detailed and perfectly summarizes the new and novel results. It contains a number of useful experience and findings. The authors show the results using a graph. Evaluation of these results is appropriate and draws realistic conclusions. On the other hand, it make useful and interesting findings that may be interested.

Conclusions

The chapter summarizes and well sums up the essence of the manuscript. This chapter explain and justified by the data. I think that the allegations are supported by the data and results.

The manuscript is suitable for indexing after an above-mentioned minor revisions.

Is the work clearly and accurately presented and does it cite the current literature?

Yes

Is the study design appropriate and is the work technically sound?

Yes

Are sufficient details of methods and analysis provided to allow replication by others?

Yes

If applicable, is the statistical analysis and its interpretation appropriate?

Yes

Are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions drawn adequately supported by the results?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Soil biology, soil chemistry

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

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