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The first tropical ‘metal farm’: Some perspectives from field and pot experiments

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ABSTRACT

Agromining is the chain of processes that allows the phytoextraction of economically valuable elements by selected hyperaccumulator plants, and subsequent processing of biomass to produce targeted metals or commercial compounds of high value. Although substantial unrealized opportunities exist for economic nickel (Ni) agromining in the tropics, this technology has remained relatively unexploited. This study investigated the soil chemistry of a newly established tropical ‘metal farm’ and elucidated the elemental accumulation in prospective species to be used in a viable large-scale tropical Ni agromining program on ultramafic soils in Sabah (Malaysia). We found that a major portion of the site (>90%) had high total Ni concentrations ($>2000\,\mu g\,g^{-1}$) in a shallow Eutric Cambisol Magnesic. This study also recorded high phytoavailable soil Ni concentrations in the field plot, which is a desired property of soils intended for commercial Ni agromining. Moreover, the average soil pH of the field (pH 6.4) is ideal for maximum Ni uptake in the local candidate species. We recorded low concentrations of Ca, K and P, indicating the need for an improved fertilizer regime in sustainable agromining. The extraordinary shoot Ni concentrations (>2 wt%), coupled with the high purity of the ‘bio-ore’ of *Phyllanthus rufuschaneyi*, confirm its high potential for economic Ni agromining. The success of our first field trial is critical to provide ‘real-life’ evidence of the value of large-scale tropical ‘metal farming’. Research priorities include the need to intensify the search for candidate species, determine their agronomy, and to develop sustainable technologies to process the biomass to recover valuable products.

1. Introduction

Ultramafic rocks are mainly composed of ferromagnesian minerals obducted at continental margins (Brooks, 1987). Ultramafic soils, which cover >3% of the Earth's surface, are derived from igneous ultramafic rocks and have inherently low concentrations of essential nutrients particularly phosphorous (P), calcium (Ca) and potassium (K), but relatively high concentrations of magnesium (Mg) and nickel (Ni) (Echevarria, 2018; Guillot and Hattori, 2013; Proctor, 2003). These geochemical peculiarities offer an extreme edaphic environment for the local vegetation (Proctor et al., 1988). Some plants have evolved ecophysiological mechanisms to tolerate and accumulate trace elements, such as Ni, from these soils (Baker, 1981). Hyperaccumulators are plants that can accumulate specific trace elements to concentrations in their shoots far higher than other plants growing on the same substrates (Reeves, 1992). The hyperaccumulation threshold values are set at: $100\,\mu g\,g^{-1}$ for Cd, Se and Tl; $300\,\mu g\,g^{-1}$ for Co, Cr and Cu; $1000\,\mu g\,g^{-1}$ for As, Ni and Pb; $3000\,\mu g\,g^{-1}$ for Zn; and $10{,}000\,\mu g\,g^{-1}$ for Mn (van der Ent et al., 2013a). Globally, >700 hyperaccumulator plant species are now known, about 70% of which are Ni hyperaccumulators (Reeves et al., 2018). Notably, only 10% of the ~500 Ni hyperaccumulators reported to date are able to accumulate Ni to concentrations exceeding 1 wt% in the shoot (Nkrumah et al., 2016a, 2018a; van der Ent et al., 2015a) and are termed ‘hypernickelophores’ (Jaffré and Schmid, 1974).

Agromining is the chain of processes that allows the extraction of economically valuable elements from selected hyperaccumulator plants (‘metal crops’), and subsequent processing of biomass to produce com-

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mercial compounds of high value (Morel, 2013; van der Ent et al., 2015a; Vaughan et al., 2017; Zhang et al., 2014). As such, it is analogous to a traditional agro-food chain of processes, such as the production of wheat flour for bread or animal feed. Until recently, agromining has been developed in temperate countries, and primarily addresses Ni (Chaney et al., 2007; Kidd et al., 2018). It is based on the annual cropping of herbaceous or shrubby Ni hyperaccumulators selected from the Brassicaceae family and on the subsequent Ni recovery by hydrometallurgical processes (Bani et al., 2015; Barbaroux et al., 2012; Li et al., 2003; Simonnot et al., 2018; Zhang et al., 2016; Vaughan et al., 2017).

Although substantial unrealized opportunities exist for economic Ni agromining in the tropics, this technology has remained relatively unexplored (Nkrumah et al., 2016b; van der Ent et al., 2013b, 2015a). In the Asia-Pacific region (Indonesia, Malaysia, The Philippines, Papua New Guinea and New Caledonia), tropical Ni agromining can be a complementary process to existing mining operations in areas with low-grade Ni soils or Ni waste materials, as a part of a progressive rehabilitation process after conventional resource extraction (Erskine et al., 2018). Agromining could also replace existing marginal and abandoned agriculture on poor ultramafic soils, herein referred to as 'metal farm' (van der Ent et al., 2015a). The application of agromining is envisaged to provide opportunities for an income source for communities in Malaysia, Indonesia and the Philippines as an alternative to conventional agriculture or agroforestry ('farming for nickel') on these inherently severely infertile soils.

Recent laboratory, glasshouse and field experiments in Sabah (Malaysia) have demonstrated the great potential of growing plants to recover Ni from ultramafic soils (Nkrumah et al., 2017). Plants chosen for agromining in the tropics are shrubs or trees (e.g. *Phyllanthus rufuschaneyi* and *Rinorea* cf. *bengalensis*), discovered in the secondary forests developed on ultramafic substrates (e.g. in the Kinabalu National Park, Sabah) (Bouman et al., 2018; Nkrumah et al., 2018b, 2018c; van der Ent et al., 2016b). Nickel agromining could have high economic potential in the tropics, but large-scale demonstrations are needed to provide 'real-life' evidence of 'metal farms' for commercial operations. Selection of sites for 'metal farms' is a key consideration in developing a viable agromining operation (Nkrumah et al., 2016a). However, in the tropics, detailed information on the biogeochemistry of a typical 'metal farm' does not presently exist. Moreover, a higher soil Ni phytoavailability is always a desired property for any commercial agromining, but whether the plant-available soil Ni pool in the topsoil of tropical ultramafic substrates can sustain maximum Ni yields over the number of cropping years required for economic tropical 'metal farming' is unknown. This study investigated the spatial distribution of targeted elements (total and extractable concentrations) and soil pH in a typical tropical 'metal farm'. We also conducted a growth trial (in large pots using unfertilized ultramafic soils collected from a field that had been prepared for 'metal farming') to elucidate the elemental accumulation in *P. rufuschaneyi*. We highlight the potential development of tropical agromining and the subsequent needs for a general strategy, further scientific knowledge and the technological tools for a sustainable and profitable practice.

2. Materials and methods

2.1. Plant species

The tropical Ni hyperaccumulator species selected for this research project was *Phyllanthus rufuschaneyi* (Phyllanthaceae). This newly-described taxon is highly light-demanding and is known to be restricted to only two localities in Sabah (Bukit Hampuan and Lompoyou Hill; Bouman et al., 2018). The habitat is open secondary scrub that has been affected by recurring forest fires and excessive logging (van der Ent et al., 2015b). Considering that only limited populations exist, and these are highly prone to incessant disturbances, *P. rufuschaneyi* is classified as an endangered species according to IUCN criteria (Bouman et al., 2018). The soil type in the native habitat is young (eroded) (Hyper)eutric Chromic Cambisols Magnesic on strongly serpentinized bedrock. These soils are characterized by exceptionally high Mg/Ca quotients, *circum*-neutral pH and high diethylenetriaminepentaacetic acid (DTPA)-extractable Ni (van der Ent et al., 2017; Echevarria, 2018). *Phyllanthus rufuschaneyi* accumulates exceptional concentrations of Ni in the shoot tissues, with up to 2.5 wt% in the old leaves and 1.23 wt% in the twigs (van der Ent et al., 2015a, 2015b; van der Ent and Mulligan, 2015). The Ni concentrations in the phloem sap and the seeds are high, with mean concentrations of 8830 mg L^{-1} and 1.8 wt%, respectively (van der Ent and Mulligan, 2015). Considering the high shoot Ni concentrations, coupled with its multi-stemmed habit, rapid re-growth after coppicing, ease of propagation and pest resistance, *P. rufuschaneyi* makes an ideal candidate for use as a Ni 'metal crop' for tropical Ni agromining (Bouman et al., 2018).

2.2. Experimental design

The research program was carried out in two phases: i) a large field experiment in a previously unfertilized ultramafic area (Eutric Chromic Cambisol [Magnesic]) near Pahu village (335 m asl; 6°7′49″N; 116°46′22″E), Sabah, Malaysia, and ii) a pot experiment using soils obtained from the top 0 to 10–20 cm of the field plot. Fig. 1 shows an overview of the field plot. *Phyllanthus rufuschaneyi* wildlings collected from their native ultramafic habitats were planted manually into the field plot. The pot experiment was part of a large 12-month randomised block growth trial in large pots in Kinabalu National Park sub-station Monggis (altitude 345 m asl; latitude DMS: 6°12′1.58″N; longitude DMS: 116°45′8.03″E), Sabah. [See Nkrumah et al., 2018d for detailed experimental design, the local climatic factors and initial soil chemical analysis.] The pot experiment reported here included *P. rufuschaneyi* wildlings (collected from their native ultramafic habitats, all from a single location) potted in an 'untreated control' with five replicates. The wildlings (~10–15 cm height) were potted immediately in ultramafic soils after collection and allowed to acclimatize in the shade house for 2 weeks and then transplanted into pots of ultramafic soil. After transplanting, they were then allowed to establish for a period of 3 months before the start of the experiment.

2.3. Collection of plant tissue samples for bulk analysis

Plants from the pot trial were harvested in March 2017 after 12 months from the start of the experiment and then separated into various fractions: roots, lower stem, upper stem, old leaves and young leaves. The plant samples were then oven-dried at 70 °C for five days. Each sample was weighed, ground to a fine powder and subsequently packed for transport to Australia and gamma-irradiated at Steritech Pty Ltd. in Brisbane following Australian Quarantine Regulations. The samples were then digested using 4 mL HNO_3 (70%) and 1 mL H_2O_2 (30%) in a microwave oven (Milestone Start D) for a 45-minute program and diluted to 40 mL with ultrapure water (Millipore 18.2 MΩ cm^{-1} at 25 °C) before analysis by Inductively-Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) (Thermo iCap7400) for Ni, Co, Cr, Cu, Zn, Mn, Fe, Mg, Ca, Na, K, S and P.

2.4. Field collection of soil samples

Soil samples were taken from the ~1 ha 'metal farm' in Sabah. A total of 93 samples were collected from the field plot at a depth of 0 to 10–20 cm using a scoop (see Fig. 1b). They were then subsequently packed for transport to Australia and gamma-irradiated at Steritech Pty. Ltd. in Brisbane following Australian Quarantine Regulations. The

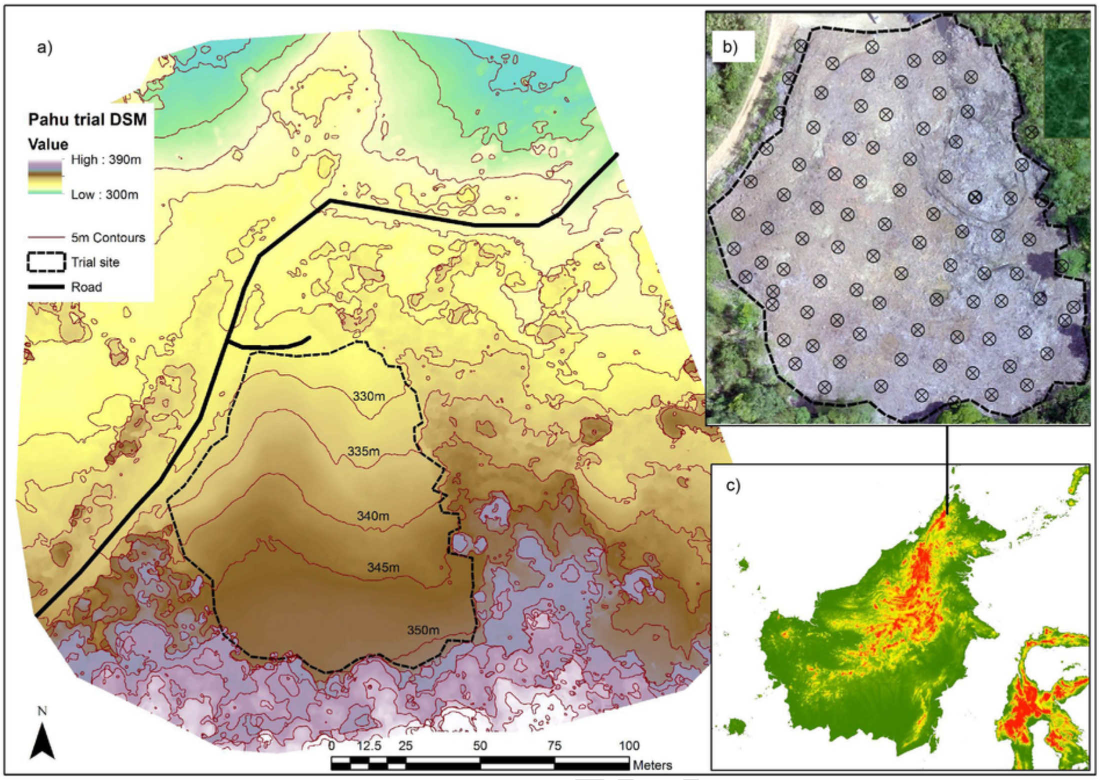

Fig. 1. a) Topography of the experimental 'metal farm' in Sabah, Malaysia, b) distribution of soil sample collection points across the field plot and c) location of the trial site on the Island of Borneo. (Adapted from Nkrumah et al. (2018a).)

soil samples were air-dried and sieved (<2 mm) and then sub-samples (~300 mg) digested using 9 mL 70% HNO_3 and 3 mL 37% HCl per sample in a microwave digester (Milestone Start D) for 1.5 h, and diluted to 40 mL with ultrapure water before analysis to obtain pseudo-total elemental concentrations. Soil pH was measured in a 1:2.5 soil:water mixture after 2 h shaking. Exchangeable trace elements were extracted in 0.01 M $Sr(NO_3)_2$ at a soil:solution ratio of 1:4 (10 gram soil with 40 mL solution) and 2 h shaking time (adapted from Kukier and Chaney, 2001). As a means of estimating potentially phytoavailable trace elements, diethylenetriaminepentaacetic acid (DTPA-extractant) was used according to the method of Becquer et al. (1995) which was adapted from the original method by Lindsay and Norvell (1978), with the following modifications: exclusion of triethanolamine (TEA), initial pH adjusted to 5.3, 5 g soil used with 25 mL extractant, and an extraction time of 1 h. The soil digests/extracts were analyzed by ICP-AES (Thermo iCap7400) for Ni, Co, Cr, Cu, Zn, Mn, Fe, Mg, Ca, Na, K, S and P. The ICP-AES accuracy for the concentrations of all the above-listed elements was checked by including certified reference plant materials that were digested in the same conditions as the plants after every 30–50 samples. The concentrations in the digestions solutions of these certified reference materials were always within the 5% error range for all the above-listed elements.

2.5. Spatial analysis

ArcGIS version 10.5 (ESRI, 2017) was used to plot the GPS location and values of the soil parameters. Interpolation of the soil values were calculated using an inverse distance weighted (IDW) technique available in the ArcGIS Geostatistical Analyst toolbox. IDW uses the measured values to predict surrounding values for any unsampled location, assuming that closer points are more alike than those that are farther apart and without explicit assumptions about the statistical properties of the input data.

3. Results

3.1. Spatial soil chemistry in the field plot

The spatial distribution of the total Ni concentrations of the field plot was not uniform, ranging from 1250 to 6550 $\mu g\, g^{-1}$ (Fig. 2a). However, a major portion of the site ($>90\%$) had exceptionally high total Ni concentrations ($>2000\, \mu g\, g^{-1}$). Furthermore, about 15% of the field plot had total Ni concentrations between 2500 and 3000 $\mu g\, g^{-1}$; 40% had 3500–4000 $\mu g\, g^{-1}$; 30% had 4000–5000 $\mu g\, g^{-1}$ and ~5% had $>5000\, \mu g\, g^{-1}$. The total Ni concentrations of the field plot had strong correlation with the total Fe concentrations (Table 1). The spatial distribution of the DTPA-extractable Ni concentrations of the field plot was not homogeneous (Fig. 2b). A large part of the site (~70%) recorded DTPA-extractable Ni concentrations $>150\, \mu g\, g^{-1}$; only about 20% of the field had DTPA-extractable Ni concentrations ranging from 100 to 150 $\mu g\, g^{-1}$. The remaining area had 30–100 $\mu g\, g^{-1}$. The relatively low DTPA-extractable Ni concentrations ($<100\, \mu g\, g^{-1}$) were recorded in waterlogged areas of the field. Interestingly, extremely high DTPA-extractable Ni concentrations ($>400\, \mu g\, g^{-1}$) were recorded in areas where natural populations of *Rinorea* cf. *bengalensis* had occurred prior to land clearing. The correlation in spatial distribution between the total and DTPA-extractable Ni concentrations of the field was not significant (see Table 1; Fig. 3). The distribution of the $Sr(NO_3)_2$-extractable Ni concentrations of the field plot was variable, ranging from 0.05–11.5 $\mu g\, g^{-1}$ (Fig. 2c). Extremely low $Sr(NO_3)_2$-extractable Ni concentrations were recorded in the waterlogged areas; these areas recorded the highest soil pH (up to pH 7.2). Very high concentrations ($>8.0\, \mu g\, g^{-1}$) were recorded again in areas where natural populations of *R.* cf. *bengalensis* had occurred. There was no correlation in spatial distribution of the total and $Sr(NO_3)_2$-extractable Ni concentrations (Fig. 3). However, areas that recorded high DTPA-extractable

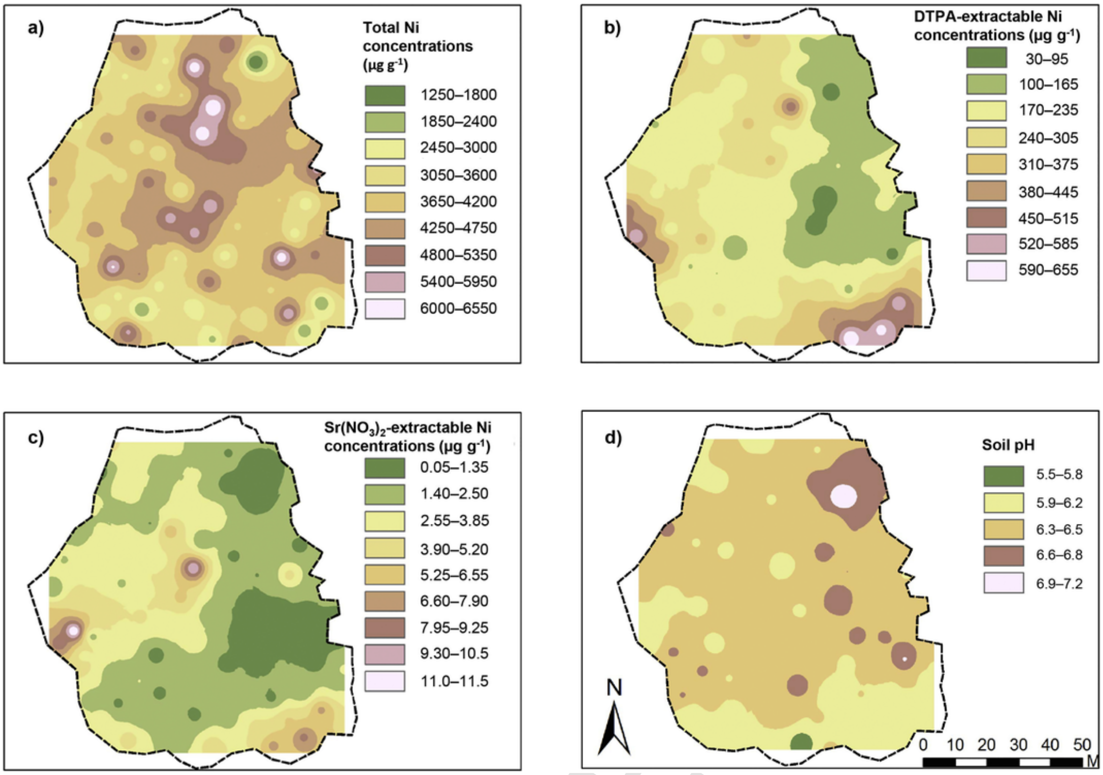

Fig. 2. Spatial distribution of a) soil total, b) DTPA-extractable and c) $Sr(NO_3)_2$-extractable nickel concentrations, and d) soil pH across the first tropical 'metal farm' in Sabah.

Table 1
Correlations of different soil properties.

Soil parameters	pH	Total-Ni	Total-Fe	DTPA-Ni
pH				
Total-Ni	0.150**			
Total-Fe	0.140**	0.825**		
DTPA-Ni	−0.600**	−0.145**	−0.155**	
DTPA-Mn	−0.400**	0.060**	0.080**	0.400**

n = 90.
** $p < 0.0001$.

Ni concentrations also had high $Sr(NO_3)_2$-extractable Ni concentrations, and regions with low DTPA-extractable Ni concentrations had extremely low $Sr(NO_3)_2$-extractable Ni concentrations because of the extreme mobility of $Sr(NO_3)_2$-extractable Ni (correlation DTPA-$Sr(NO_3)_2$-extractable Ni is R^2 0.50) (Fig. 3).

The spatial distribution of the soil pH was nearly uniform (Fig. 2d). The soil pH of the field ranged from pH 5.5–7.2, but a significant proportion of the field (~70%) had a soil pH between 6.3 and 6.5. The regions with relatively low soil pH (pH 5.9–6.2) corresponded to areas with relatively high extractable Ni concentrations (both DTPA and $Sr(NO_3)_2$) (Fig. 3). In contrast, areas with relatively high soil pH ($pH > 6.5$) had relatively low extractable Ni concentrations (Table 1). Soil pH had moderate correlations with DTPA-extractable Mn, compared to total Fe concentrations which had low correlation with soil pH (Table 1). The spatial distribution of soil pH did not correlate with the total Ni concentrations (Table 1; Fig. 3).

The exchangeable K of the field plot was nearly uniform (Fig. 4a). In general, the field had deficient concentrations of exchangeable K ($< 170\,\mu g\,g^{-1}$), which is characteristic of ultramafic soils. The total P concentrations of the field were relatively low, compared to the total Ni concentrations (Fig. 4b). The distribution of the total P concentrations of the field was not homogeneous, ranging from 100 to $850\,\mu g\,g^{-1}$. The majority of the field had total P concentrations $< 450\,\mu g\,g^{-1}$. The pattern in the spatial distribution for exchangeable Ca and exchangeable Mg concentrations was the opposite; regions with low exchangeable Ca concentrations recorded high exchangeable Mg concentrations (Figs. 3, 4c). Notably, significant areas of the field had very high Mg concentrations but had relatively low exchangeable Ca concentrations (Fig. 4).

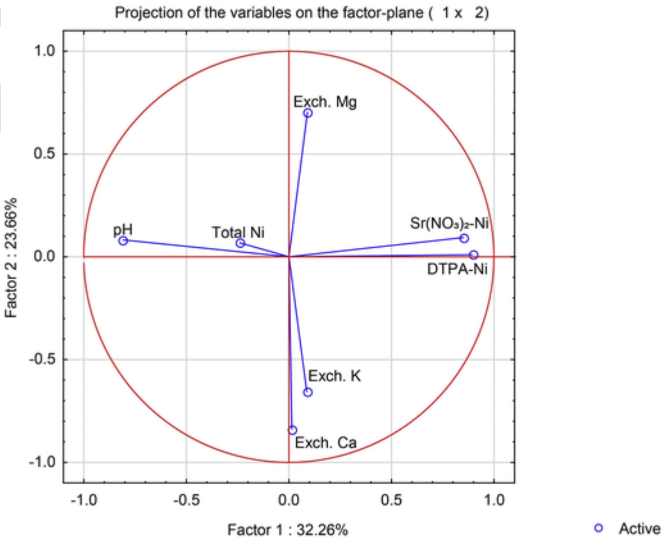

Fig. 3. Principal components and classification analysis of the soil physico-chemical properties of the Pahu field trial. There is high positive correlations in the DTPA- and $Sr(NO_3)_2$-extractable Ni concentrations (Factor 1: Soil pH vs. extractable Ni concentrations (DTPA and $Sr(NO_3)_2$) and Factor 2: Exchangeable Ca vs. Exch. Mg).

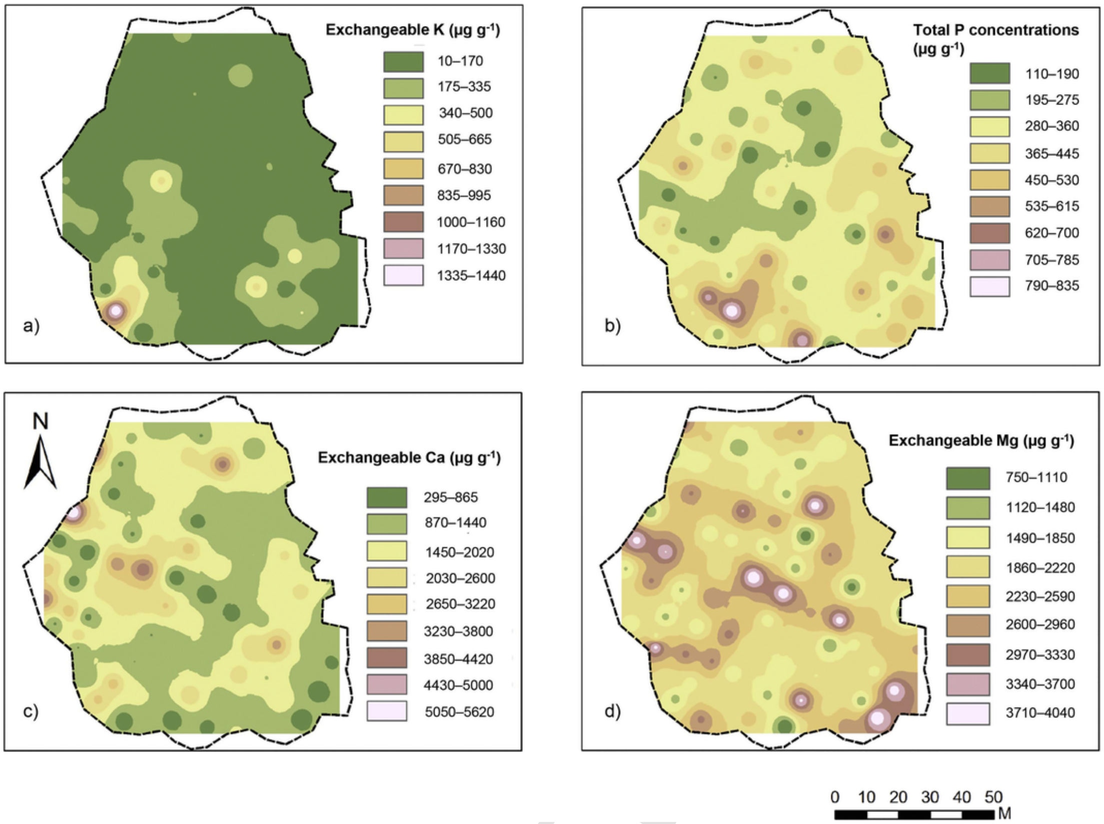

Fig. 4. Spatial distribution of soil: a) exchangeable K, b) total P, c) exchangeable Ca and d) exchangeable Mg concentrations across the first tropical 'metal farm' in Sabah.

3.2. Bulk elemental concentrations of Phyllanthus rufuschaneyi collected from the pot and field trials

The bulk elemental concentrations in the various fractions of pot grown *P. rufuschaneyi* are reported in Table 2. The concentrations of trace elements in all plant fractions are unremarkable, with the exception of Ni. The mean concentrations of Co, Fe, Mn and Zn in the shoots are $<200\,\mu g\,g^{-1}$. Cobalt is low in all plant parts, with mean concentrations $<100\,\mu g\,g^{-1}$. However, the concentrations of the major elements (Ca, K and P) in the various plant fractions are relatively high (Table 2) compared to the low concentrations of these elements in the soil (Fig. 4). Notably, the Ca/Mg quotients in the various plant fractions are high, despite the extremely low Ca/Mg quotients in the soil (Fig. 4). The Ni concentrations in the shoots are exceptional; a concentration as high as $28{,}400\,\mu g\,g^{-1}$ was recorded in the old leaves (Fig. 5). The old leaves, young leaves and upper stem had mean Ni concentrations > 1 wt%. Shoot Ni concentrations increased in the order: lower stem < upper stem < young leaves < old leaves (Fig. 5). The Ni concentrations in the lower stem fraction were also high (up to $7870\,\mu g\,g^{-1}$). In the field grown *P. rufuschaneyi*, Ni concentrations as high as 2.5 wt% and $8950\,\mu g\,g^{-1}$ were recorded in the leaf and stem fractions, respectively.

4. Discussion

This study recorded extremely high total Ni concentrations in the field plot used for the trial (as high as $6500\,\mu g\,g^{-1}$). Ultramafic soils are usually characterized by high concentrations of trace elements, including Ni (Echevarria, 2018; van der Ent et al., 2018a). Although the spatial distribution of the total Ni concentrations was heterogeneous, significant portions of the field recorded $>2000\,\mu g\,g^{-1}$. The range of total

Table 2
Bulk elemental concentrations in different plant tissues of *Phyllanthus rufuschaneyi* grown (over 12 months) in large pots containing unfertilized ultramafic soils collected from the field plot at Pahu (Sabah, Malaysia). The elemental concentrations are given in ranges and means in $\mu g\,g^{-1}$ (n = 5 for all plant fractions, except roots which had 2). The digest and extracts were analyzed with ICP-AES.

Plant fraction	Ca	Co	Fe	K	Mg	Mn	P	S	Ni	Zn
Old leaves	8230–11,300 [9140]	25–155 [65]	25–205 [120]	9420–24,200 [15,210]	2690–3700 [3220]	90–115 [105]	805–1090 [930]	3390–5080 [4100]	25,100–28,400 [26,400]	150–170 [160]
Young leaves	4490–9190 [7020]	30–85 [50]	70–100 [80]	14,400–26,100 [18,300]	2230–4380 [3300]	100–175 [120]	720–1320 [1030]	1840–3830 [2860]	17,500–23,200 [21,100]	135–155 [140]
Upper stem	6360–13,600 [8815]	10–25 [20]	45–80 [60]	19,800–20,000 [23,400]	925–3470 [1630]	50–75 [60]	1130–1890 [1600]	1420–3060 [2060]	8900–17,300 [11,800]	45–95 [65]
Lower stem	6290–11,800 [9440]	5.0–15 [10]	80–220 [175]	6640–13,500 [10,200]	545–990 [805]	35–60 [50]	600–1430 [870]	830–1570 [1270]	5270–7870 [6680]	40–55 [50]
Roots	9710–18,500 [9270]	10–25 [15]	165–445 [300]	5740–6290 [6020]	2020–2200 [2110]	75–110 [90]	605–745 [675]	995–1270 [1130]	5810–12,600 [9220]	70–80 [75]

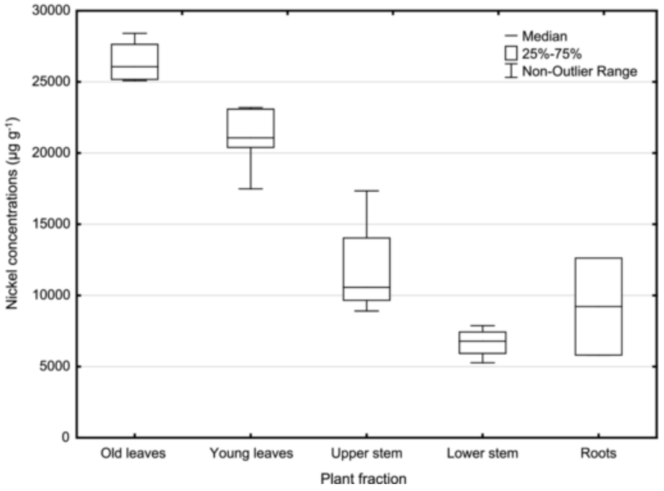

Fig. 5. Nickel concentrations in the various fractions of *Phyllanthus rufuschaneyi*. Key to symbols of boxplots: open squares are the ±mean, and whiskers are ±standard deviation (SD).

Ni concentrations recorded in this study is below the threshold required for conventional mining technologies, but adequate to supply Ni hyperaccumulators (van der Ent et al., 2013b, 2015a; Nkrumah et al., 2016a, 2016b, 2018a, 2018e). Ultramafic soils with total Ni concentrations >0.1% with high phytoavailable Ni pools are potentially suitable for Ni agromining operations (van der Ent et al., 2015a; Nkrumah et al., 2016a, 2016b, 2018a, 2018e). For a comparison, the average total Ni concentrations recorded in an agromining field on an ultramafic substrate in Albania was 3150 $\mu g\,g^{-1}$ (Bani et al., 2015).

We stress that a high phytoavailable soil Ni concentration is always a desired property of soils intended for commercial agromining (Bani et al., 2015; Chaney et al., 2007; Li et al., 2003; Nkrumah et al., 2016a, 2018a, 2018e; van der Ent et al., 2015a). This study recorded exceptionally high DTPA- and $Sr(NO_3)_2$-extractable Ni concentrations across the field, with the exception of waterlogged areas which had concentrations $<100\,\mu g\,g^{-1}$ DTPA-Ni and $2.5\,\mu g\,g^{-1}$ Sr-extractable Ni, respectively. In tropical ultramafic soils, nickel availability is usually high in clay mineral-rich young soils (Cambisols) and saprolite materials than in those dominated by goethite in strongly weathered ultramafic soils such as Ferralsols (Echevarria, 2018; Raous et al., 2013). Nickel associated to goethite in Ferralsols is contained in the crystal lattice of the Fe oxides and is thus less available to plants (Massoura et al., 2006). Therefore, Cambisols and saprolite substrates in Sabah are more useful for economic Ni agromining than Ferralsols. In nature, the occurrence of Ni hyperaccumulators in Sabah is restricted to Cambisols and Cambic Leptosols; the high Ni exchangeable pools appear to favor Ni hyperaccumulation by these specialized plants (van der Ent et al., 2016a, 2018a, 2018b). We found extreme values of extractable Ni concentrations in some areas of the field that previously had natural populations of the Ni hyperaccumulator species, *R.* cf. *bengalensis*. This finding confirms the contribution of leaf litter of Ni hyperaccumulator plants to the phytoavailable Ni in local ultramafic substrates (Estrade et al., 2015; Zelano et al., 2018). Future study needs to investigate the biogeochemical cycling of Ni (and other elements) in the native habitats of tropical hyperaccumulator plants. During agromining operations, the biomass of Ni-rich shoots will be removed by harvesting, but the subsoil Ni may be sufficient to supply the next crop (see Nkrumah et al., 2018a). The extremely high extractable Ni concentrations have major implications for economic Ni agromining. The extensive occurrence of Cambisols and saprolite materials in Sabah and other regions in Southeast Asia suggests that there are substantial potential sites that could be harnessed for large-scale agromining purposes. However, there is high pedodiversity of ultramafic soils in Sabah (van der Ent et al., 2018a), hence, it is important to assess the phytoavailable soil Ni concentrations of every substrate to be used for agromining. Moreover, future studies must assess the soil Ni-bearing phases of soils intended for economic Ni agromining as Ni availability is mainly influenced by the soil Ni-bearing phases (Massoura et al., 2006).

It is of note that we recorded low extractable Ni concentrations in the waterlogged areas of our field plot. This indicates that there is possible loss of soluble Ni in the affected areas through run-off. If soils remain anaerobic below the very surface layer, roots cannot penetrate the anaerobic layer, limiting access to soil Ni. Future study is required to investigate the depletion rates of extractable soil Ni in these areas: i) to measure the Ni concentrations of the run-off water along the flow direction, and ii) to elucidate the Ni concentration gradient in the sediment of the downstream river. The relatively high soil pH in the waterlogged areas could possibly explain the low Ni extractability, but this needs to be investigated in future studies. The low extractable Ni concentrations in the waterlogged areas imply that poorly drained soils may not support economic Ni agromining. Moreover, evidence from the pot trial suggests that poorly drained soils also have adverse growth effects on *P. rufuschaneyi* (Nkrumah et al., 2017). In temperate regions, poorly drained soils adversely affect the growth of *Alyssum* Ni hyperaccumulator species, and plants may die before normal harvest time (Chaney et al., 2007). These authors have demonstrated in field trials that ridge tilling and drainage channels can improve root aeration.

Soil pH plays a major role in Ni uptake in Ni hyperaccumulator plants. The average soil pH of the field plot was pH 6.4 and the range was 5.5 to 7.2 (Fig. 2d). We found that areas with relatively low soil pH (5.9–6.2) corresponded to areas with relatively high extractable Ni concentrations. Moreover, areas with relatively high soil pH (>6.5) recorded low extractable Ni concentrations (Figs. 2, 3). It is currently unknown whether low soil pH (corresponding to high extractable Ni concentrations) will yield maximum Ni uptake in the local 'metal crops' to be used in our field. Studies in temperate ultramafic soils have shown that reducing soil pH did not increase Ni uptake in *Alyssum* Ni hyperaccumulator species (Li et al., 2003; Kukier et al., 2004; Chaney et al., 2007). These authors found that shoot Ni concentrations were highest at pH 6.5 in their test soil with 22% Fe. The pH of the native ultramafic soils where the *Alyssum* species occur were typically 6.5–8. It is possible that the average soil pH of our field (6.4) is ideal for maximum Ni uptake in local candidate species because, in nature, most Ni hyperaccumulators in Sabah are restricted to *circum*-neutral soil pH (van der Ent et al., 2016a). This is probably why the Ni-bearing phases are of high importance to define the potential for Ni phytoextraction by hyperaccumulators, as neutral pH values limit Ni solubility; a large available Ni reservoir then compensates the limited Ni concentration in the soil solution (Bani et al., 2015). Notwithstanding this, both pot and field trials are required to ascertain the Ni uptake by local 'metal crops' over a wide range of soil pH; soils can be acidified and limed to include pH from strongly acidic to mildly alkaline using soils with low and high Fe levels. These investigations are critical because of the wide pH range among ultramafic soils in tropical regions (e.g. 4.4–6.9, 5.3–6.3, 4.3–5.5 and 3.8–9.7 in New Caledonia, Indonesia, Philippines and Malaysia respectively) (van der Ent et al., 2018a).

Our study recorded deficient soil concentrations of essential macronutrients (P, Ca, N and K), which is typical of unfarmed ultramafic soils. In ultramafic soils in Kinabalu Park (Sabah), van der Ent et al. (2018a, 2018b) recorded lower concentrations of potentially plant-available P (Mehlich-3 extract), relative to that in local non-ultramafic soils (~4-fold). These attributes of the soils pose edaphic constraints to local vegetation (Echevarria, 2018). Evidence from our pot trial revealed that candidate species respond strongly to nutrient additions, despite the marginal concentrations in the local habitats of these Ni hyperaccumulator plants (Nkrumah et al., 2018d). This implies that incorporating an improved nutrient regime into tropical agromining operations may ensure sustainable production. The high Mg/Ca quotients recorded in this study have been observed in previous studies to play a critical role for the vegetation that occurs on ultramafic soils (Walker et al., 1955; Echevarria, 2018). Calcium and S additions are important to replenish depleted Ca and S concentrations during agromining operations (Chaney et al., 2008). Moreover, Ca and S additions enhance Ni tolerance in *Alyssum* Ni hyperaccumulator species (Brooks et al., 1981; Chaney et al., 2008), and *P. rufuschaneyi* (Nkrumah et al., unpublished), thereby increasing Ni yields in these species. Hence, Ca and S fertilization in the form $CaSO_4$ is an important consideration during commercial agromining operations and so standard inorganic fertilization is essential for sustainable tropical agromining operations.

For commercial Ni agromining operations, both high biomass production and shoot Ni concentrations are important prerequisites (Bani et al., 2015; Chaney et al., 2007; Nkrumah et al., 2016a, 2016b, 2018a, 2018e; van der Ent et al., 2015a). Our study has shown that *P. rufuschaneyi* has the high shoot Ni concentrations necessary for viable Ni agromining operations. Notably, the high shoot Ni concentrations recorded in the pot grown plants (Table 2) are similar to those recorded in field grown plants (~2.5 wt%) and naturally occurring ones in the native habitats (Bouman et al., 2018; van der Ent and Mulligan, 2015). Moreover, it was evident in our nutrient dosing experiments that the biomass production of *P. rufuschaneyi* could be improved with no significant shoot-dilution effect under appropriate agronomic management (Nkrumah et al., 2018d). The high Ni concentrations in the stem fraction (~1 wt%) of *P. rufuschaneyi* (Bouman et al., 2018) will be important when considering coppicing management of the crop. It is of note that the green shoot and leaf biomass of *P. rufuschaneyi* comprise ~50% of the total harvested biomass, which is indicative of its high potential for profitable agromining. The high purity of the 'bio-ore' obtained from *P. rufuschaneyi* (low in Fe and Mn oxides, silicate and other materials which interfere with recovering Ni) will also enable efficient recovery of Ni (Vaughan et al., 2017). For large-scale tropical agromining employing *P. rufuschaneyi* considering a biomass of 200 g per plant, 2.5 wt% shoot Ni concentrations, 5 plants/m^2, one harvest per year, we can achieve a Ni yield of about 250 kg ha^{-1}, the greatest to be recorded across the globe, however, this remains to be confirmed by field data. Fig. 6 shows the progress of the first tropical 'metal farm', employing *P. rufuschaneyi*. Nickel yields of 120 and 200 kg ha^{-1} have been achieved in Albania and the USA, respectively using *Alyssum* species (Bani et al., 2015; Li et al., 2003). We have previously performed a detailed economic analysis of an annual Ni agromining crop under two main production systems as demonstrated in Albania and USA (see Nkrumah et al., 2016a, 2018e). The system in Albania mainly employs manual labour in its operation with relatively low production cost, whereas in the USA the system is fully mechanized with a relatively high cost of production. We have shown that Ni agromining is highly profitable under the respective systems. The production system in Sabah is similar to that of Albania, and we estimate the Ni yield to exceed that of Albania by a factor of two-fold, making Ni agromining in Sabah more economically lucrative.

5. Conclusion and outlook

This study reports on the first tropical 'metal farm' employing a local woody 'metal crop' (the endemic species *P. rufuschaneyi*), which could serve as a model for future commercial agromining operations in tropical regions. Although large expanses of ultramafic soils exist in the tropical Asia Pacific region (Galey et al., 2017), the high local pedodiversity (van der Ent et al., 2018a) calls for detailed biogeochemical studies prior to site selection for viable large-scale commercial agromining. The current agromining trial plot has high extractable Ni concentrations and *circum*-neutral pH, which are desirable properties for

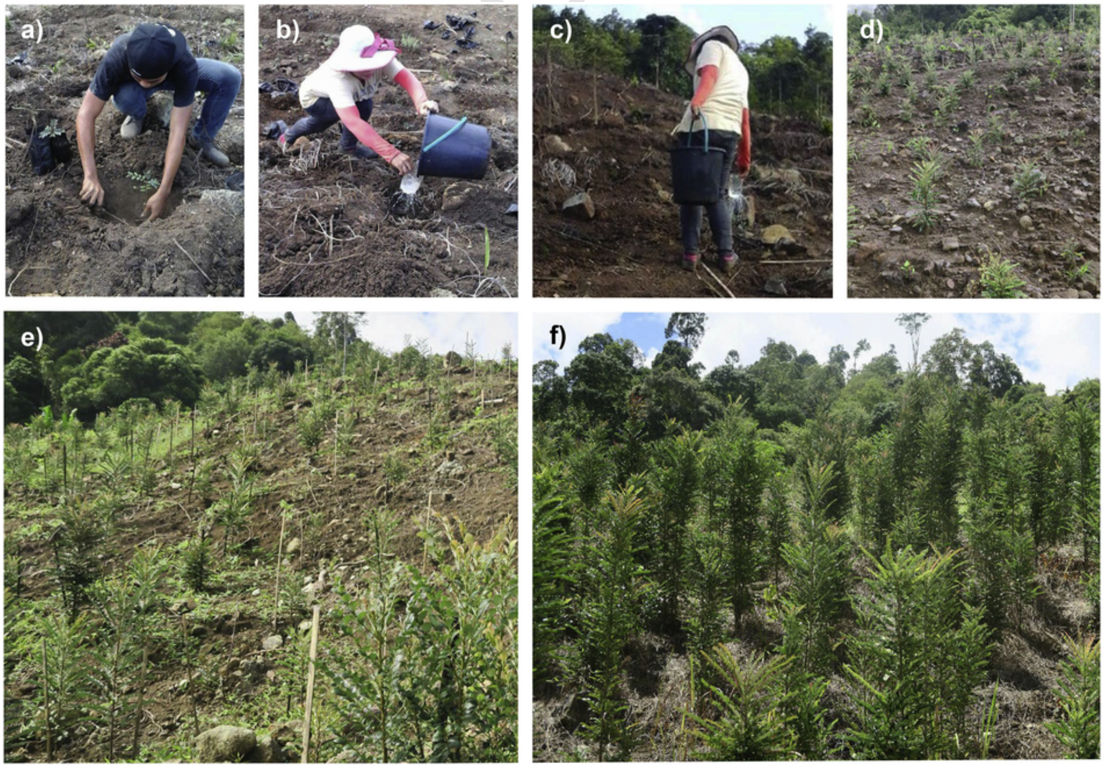

Fig. 6. Progress of the first large-scale demonstration of tropical Ni agromining in Sabah, Malaysia: a, b, c: manual planting and watering of *Phyllanthus rufuschaneyi* wildlings, d, e: initial growth stages after establishment (6 and 9 months respectively), and f: current status of the field trial, 2 years after planting.

soils intended for economic agromining on account of the favorable Ni hyperaccumulation responses to these types of soils (van der Ent et al., 2016b). However, the low concentrations of major plant nutrients in the trial plot, which is an inherent characteristic of ultramafic soils, indicates the need for an effective fertilizer regime. Soil physical properties may also play an important role in ensuring sustainable economic Ni agromining. The drainage conditions, rocky nature of some substrates and topography of the site may require conditioning prior to usage. Efforts are underway to explore more potential sites for implementation of large-scale commercial agromining in Sabah.

The discoveries of a range of different hyperaccumulator species in Sabah, some with characteristics amenable to utilization as 'metal crops' (van der Ent et al., 2015b, 2018b), shows the need to undertake further systematic screening of the Sabah flora considering the high plant diversity that occurs on ultramafic soils in this part of the world. The use of state-of-the-art portable X-Ray fluorescence spectroscopy (XRF) instruments could overcome the drawbacks of conventional screening approaches and enable rapid non-destructive analyses of large existing herbarium collections (Gei et al., 2017). This method is fast (<30 s/sample) and hence makes it possible to perform systematic screening of entire phylogenetic lineages, which can lead to discoveries of taxa with unique properties. This approach has already led to the discovery of the novel Ni hyperaccumulator *Antidesma montis-silam* in Sabah, which has a number of highly desirable traits (high biomass and up to 32,700 $\mu g\,g^{-1}$ foliar Ni), for application in agromining (Nkrumah et al., 2018b). The selection of suitable candidate species depends primarily on biomass and Ni yields. Optimum pH and soil nutrient supplies need to be established to promote high yields of high Ni crops for any prospective agromining species. This study shows that *P. rufuschaneyi* has immediate practical use as 'metal crop' for viable commercial agromining operations in Sabah. However, the current approach of propagation of *P. rufuschaneyi* using wildlings collected from native ultramafic habitats is not sustainable as only few natural populations are known to occur in Sabah. Although perennial crops could help sustain population numbers, there is the need to undertake possible mass propagation via cuttings and seeds. Cuttings seem immediately practical, but considerable research is required in producing viable seeds and raising seedlings. Ordinary cultivation methods, without appropriate agronomic systems, may not be sufficient to make Ni yields from tropical 'metal crops' economically viable. Evidence from a nutrient dosing experiment suggests that *P. rufuschaneyi* responds strongly to moderate inorganic fertilization (Nkrumah et al., 2018d). Optimum plant density and weed control measures will minimise competition for vital resources such as water and nutrients. Biomass harvesting methods have not been tested for *P. rufuschaneyi*, but the most effective method may be annual collection of foliage by coppicing before leaf fall. Moreover, multi-year biomass collection appears to be a possibility but needs to be tested in the field. Innovative extraction methods are currently being explored to efficiently recover Ni from harvested biomass that take into account the comparatively remove location of agromining farms in Sabah.

We have demonstrated that commercial-scale Ni agromining is feasible in Sabah (Malaysia), and the future holds opportunities for its implementation in other Asia-Pacific regions with similar settings, including Indonesia, the Philippines, Papua New Guinea and New Caledonia. Tropical agromining presents economic and socially desirable prospects in Sabah, but a detailed life cycle analysis is a research priority. Considering the untapped potential in other tropical regions such as Africa and South America, agromining may be possible in these regions, and could follow the same general approach reported here.

Uncited reference

Repin, 1998

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