

Nickel Accumulation in Soybean and Melastoma on Contaminated Soil

Netty Syam

Indonesia Muslim University (UMI), Makassar, South Selatan, Indonesia

Email: netty.said@umi.ac.id; nettysyam@gmail.com

Elkawakib Syam'un and Hidrawati

Department of Agronomy, Hasanuddin University, Makassar, Sulawesi Selatan, Indonesia

Email: {elkawakib, Hidrawati}@yahoo.com

Abstract—The Ni-contaminated soil was obtained from post-mining sites in Sorowako, South Sulawesi in which Soybean (*Glycine max*) and Melastoma were grown together in a pot experiment. The design of treatment was factorial design of two factors i.e. urea dose (0, 100, 200 and 300 kg/ha) and kind of compost (without compost, *Chromolaena* compost, *Glyricidia* compost and Husk compost 20 ton/ha). Biomass production and accumulation of nickel in the shoots (parts of the plant above ground) and roots (underground plant parts) were analyzed to determine the ability of plants to accumulate nickel. The values of Bioconcentration Factor (BCF) of the plant was calculated to analyze bioremediation of the plants. The results showed that urea and compost applications, especially *Chromolaena* and *Glyricidia* compost increased dry weight of soybean and Melastoma. In addition, application of urea and all types of compost resulted in decreased nickel accumulation in both soybean and Melastoma. Increased urea dose up to 100 kg/ha without compost caused BCF value increased to 0.24 in soybean (moderate accumulator category).

Index Terms—contaminated-soil, heavy metal, soybean, *Melastoma*, urea, compost

I. INTRODUCTION

In recent years, Nickel (Ni) contamination occurs upon our ecosystems due to various human and natural activities. Ni could be a serious environment pollutant [1] that could migrate contaminants into non-contaminated sites as dust or leachate through the soil, and the spreading of sewage sludge is examples of events that contribute towards contamination of our ecosystems. Excessive accumulation of heavy metals (such as Ni) in agricultural soils through wastewater irrigation, may not only result in soil contamination, but also lead to elevated heavy metal uptake by crops, and thus affect food quality and safety [2].

Remediation of soil contaminated by heavy metals has become a hot topic in the world, and the most commonly used biological method is phytoremediation, which uses plants to absorb heavy metals. It is efficient and economical. In recent years, as the repair material plant

has been used more and more, we find that the use of heavy metals and hyperaccumulators tolerant plants is the key to phytoremediation [3]. However, the success of phytoextraction depends upon the identification of suitable plant species that tolerate and accumulate heavy metals and produce large amounts of biomass using established agricultural techniques.

Phytoextraction is the accumulation of sufficient heavy metals concentration by plants, translocate and store such metals at the aboveground parts [4] thereby reducing the heavy metals concentration in the soils to regulatory levels with relatively few repeated cropping. Plants for phytoextraction, i.e. metal removal from the soil, should have the following characteristics: (i) tolerant to high levels of the metal, (ii) accumulate reasonably high levels of the metal, (iii) rapid growth rate, (iv) produce reasonably high biomass in the field and (v) profuse root system [4], [5].

Several researches proposed to use cultivated species that produce a large biomass, and simultaneously accumulate metals in non-edible parts, in phytoremediation [6], [7]. Netty *et al.* 2016 [8] conducted field research to study the response of soybean planting with *Melastoma malabathricum* (Fig. 1) in pot experiment (polyculture) on nickel contaminated soil. The study revealed that there are increasing biomass production and Ni accumulation which were higher than those in monoculture.

The major problem hindering plant remediation efficiency is that some of the metals are immobile in soils and their availability and phytoextraction rates are limited by solubility and diffusion to the root surface [4]. Inorganic and organic agents, including EDTA, citric acid, elemental sulfur or ammonium sulfate, and urea have been applied to soils to improve phytoextraction potentials of different plant species [7]. One such agent involves soil amendment with organic substances, which exerts a complex effect on soil properties [9]. Tejada *et al.* [10] also pointed out a positive role of organic substance in neutralizing the adverse effect of nickel on soil enzymatic activity. These authors found that under laboratory conditions, the negative effect of soil contamination with nickel on the activity of urease, BBA-protease, alkaline phosphatase, β -glucosidase and

arylsul-phatase may be alleviated by the application of poultry dung and cotton burr compost. This study aimed to determine the ability of nickel accumulation from Soybean and Melastoma by using urea and compost (organic fertilizer) on nickel contaminated post mining.

II. MATERIALS AND METHODS

This research was a field experiment that using *Melastoma* and soybean planted in a pot. *Melastoma malabathricum* is a shrub plant that grow and develop around the mining area in Sorowako (Fig. 1). Melastoma was planted about 4 weeks before soybean. The soil (soil obtained from the post nickel mining area in Sorowako, East Luwu regency of South Sulawesi) was dried, sifted (5 mm), mixed with compost used in the treatment and filled into plastic pots 10 kg per pot.

This experiment was adjusted to Randomized Block Design with Factorial two factors. The first factor was *Chromolaena*, *Glyrisidia*, Husk and control and second factor was urea 100 kg/ha, 200 kg/ha, 300 kg/ha and control, repeated 3 times. The number of pot used 4 pots per treatment and there were 144 pots.

Seedlings of *Melastoma malabathricum* with uniform size (2-4 foliages) collected from the post-mining lands were planted on media adaptation for 2 weeks. Soybean seeds *Tanggamus* variety were planted 3 to 5 seeds and a week later selected and planted into pot beside *Melastoma*. Plants were watered daily and each pot is added 100 kg / ha SP-36 and 100 kg / ha KCl as basic fertilizer one week after planting. Urea was applied twice, in the time of soybean planting and 6 weeks after planting.

Soil analyzed for pH (H₂O and KCl), organic C (Walkley and Black), N (Kjeldahl), P (Olsen), Ca, Mg, Na, K (1 N NH₄OAc pH 7.0) and soil texture (hydrometer method). The soil samples were dried in an oven for 6 h at 105°C, crushed, and 100 mg dissolved in 2 ml of HNO₃ (65%). This solution was heated in an oven at 200 °C for about 14 h, until the sample dissolved completely; the extract was made up to 50 ml, which was used to determine Ni concentrations [11].

Triplicate of soil and plant samples were analyzed and their means with standard error (SE) are presented. Characteristics of the soils and Ni content are presented in Table I.

Harvest crops were washed with tap water mixed with 3% HCl, and then rinsed twice with deionized water. The harvested plant stem, leaf and root were dried at 65 °C for 24 h. For Ni analysis, 100 g of dried sample of each parts of plant species were grounded and digested in a mixture of 2 ml of HNO₃ (65%). This solution was heated in an oven at 200 °C for about 14 h until the sample dissolved completely [11]. Extract was made up to 50 ml used to determine Total Ni and available Ni in soil samples by employ Inductively-Coupled Plasma, Optical Emission Spectroscopy (ICP-OES).

The ability of plants to accumulate nickel was determined by Biological Concentration Factor (BCF) that was calculated as nickel concentration ratio of plant shoot to soil [12]. BCF value of 1 to 10 indicates

hyperaccumulator plant, BCF values of 0.1 to 1 indicates moderate accumulator plant, BCF value of 0.01 to 0.1 indicates low accumulator plant, and BCF value of <0.01 indicates non-accumulator plant.



Figure 1. Melastoma (*Melastoma malabathricum* L.).

TABLE I. SOIL PROPERTIES before TREATMENT. (VALUES ARE MEAN $\pm$ STANDARD ERROR, N=3)

Soil Characteristics	Value
Clay (%)	58.67 (± 3.33)
Silt (%)	29.33 (± 1.33)
Sand (%)	12.00 (± 2.00)
pH (H ₂ O)	6.27 (± 0.05)
pH (KCl)	5.92 (± 0.06)
CEC (me100 g ⁻¹ soil)	8.55 (± 0.54)
C-Organik (%)	2.90 (± 0.25)
N-Total (%)	0.10 (± 0.01)
P ₂ O ₅ (mgkg ⁻¹)	7.00 (± 1.61)
K ₂ O	55.00 (± 3.97)
Ca (me100 g ⁻¹ soil)	3.88 (± 0.93)
Mg (me100 g ⁻¹ soil)	2.99 (± 0.21)
Na (me100 g ⁻¹ soil)	0.41 (± 0.05)
K (me100 g ⁻¹ soil)	0.12 (± 0.01)
Ni-Available (mgkg ⁻¹) *	88.91 (± 2.28)
Ni-Total (mgkg ⁻¹) *	7457.33 (± 293.51)

III. RESULTS AND DISCUSSION

A. Biomass Production

Reduction in plant biomass as a result of heavy metal stress appears to be an almost universal finding [13]. However, that finding do not occur on soybean. Planting soybeans along with Melastoma in pots has a good effect on soybean growth without toxicity on nickel contaminated media, resulting soybean can produce higher shoot biomass than Melastoma (Fig. 2) However, the application of urea and compost to nickel contaminated soil produces a biomass of soybean and Melastoma was not significant. Urea applications without or with compost in nickel contaminated soil increased shoot and root biomass production in both planted species.

In contrast, application compost to contaminated soil resulted in decreased shoot and root biomass in soybean, except on Chromolaena compost (Fig. 2 and 3). Higher biomass yields in soybean shoot because soybean is a symbiotic plant species with rhizobia that helps the plant in N fixation. In addition, the nitrogen metabolism in soybeans is influenced by the use of nickel in the change to form an existing urea (NO₃- or NH₄+) [14]. Nickel acts as an urease enzyme that hydrolyzes the CO (NH₂)₂ urea into ammonium and carbon dioxide, resulting in increased biomass in soybeans.

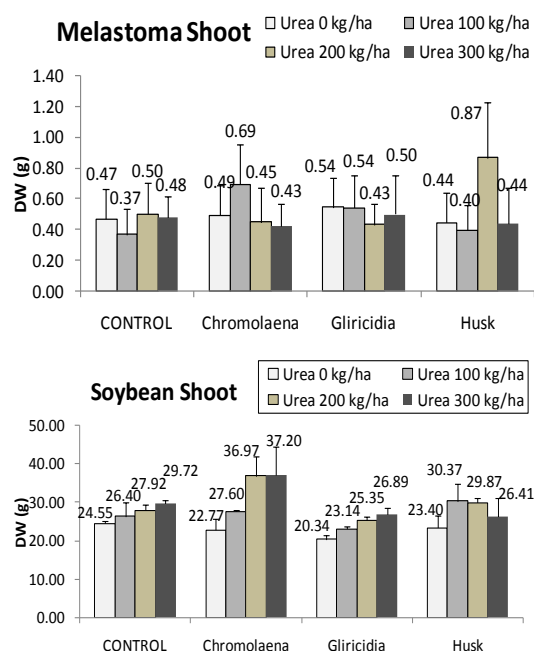


Figure 2. DW of soybean (above) and melastoma (under) shoot. Vertical bars represent SE.

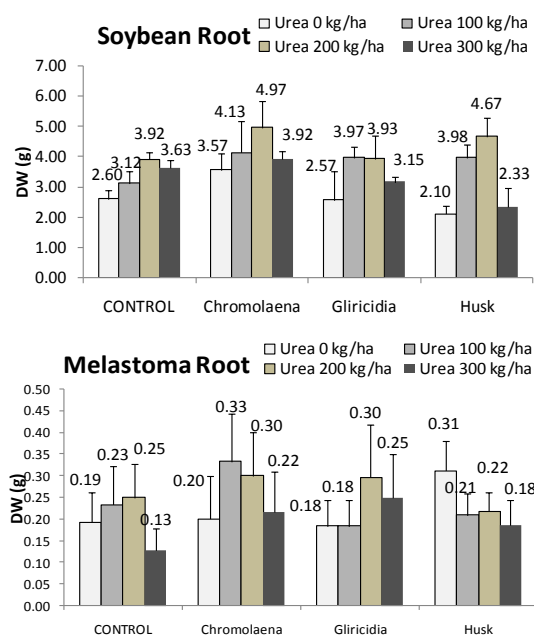


Figure 3. DW of soybean (above) and melastoma (under) root. Vertical bars represent SE.

B. Nickel Content in Soil and Plant

The concentrations of available and total nickel of the soils under the study. These soils were classified as a type Oxisol. The available Ni and total Ni content of the collected soil samples before treatment were 88.91 mg/kg (SE ± 2.28) and 7457.33 mg/kg (SE ± 293.51) respectively (Table I). Analysis of nickel content available on contaminated soils that have been treated with urea and compost showed an increased in the amount of nickel available compared to nickel available before the

treatment application. However, the amount of nickel available on the soil among all treatments tested were tends to be almost as much. Only nickel available in the Gliricidia compost which applied without urea was higher than the other treatment of 806.66 mg / kg (Table II).

A similar trend also occurs in the results of a total nickel analysis which contained in contaminated soil after treatment applied. The highest nickel content was shown in the compost of Gliricidia without urea and the lowest in Gliricidia compost with urea 300 kg /ha, i.e. 3694.99 mg/kg and 1936.09 mg/kg respectively.

TABLE II. Nickel TOTAL and AVAILABLE in SOIL after TREATMENT. (Values Are Mean $\pm$ Standard Error, N=3)

Ni Available in Soil after treatment (mg/kg)				
Urea	Control	Chromolaena	Gliricidia	Husk
0	748.53 (± 52.03)	767.22 (± 56.54)	806.66 (± 2.88)	728.37 (± 105.39)
100	607.71 (± 94.95)	661.07 (± 39.39)	758.41 (± 47.46)	706.10 (± 3.46)
200	659.90 (± 52.06)	769.13 (± 32.46)	652.09 (± 129.23)	653.47 (± 47.42)
300	750.02 (± 49.63)	666.82 (± 54.85)	774.92 (± 23.84)	754.67 (± 41.99)
Ni Total in Soil after treatment (mg/kg)				
Urea	Control	Chromolaena	Gliricidia	Husk
0	2679.38 (± 288.52)	2544.43 (± 17.93)	3694.99 (± 533.58)	2798.12 (± 574.71)
100	2709.26 (± 525.64)	2524.27 (± 143.31)	2201.05 (± 605.69)	2565.71 (± 228.45)
200	2512.90 (± 463.52)	2739.36 (± 177.18)	1893.60 (± 51.72)	2550.70 (± 111.13)
300	2325.31 (± 104.00)	3421.72 (± 155.25)	1936.09 (± 225.37)	2633.26 (± 186.34)

Analysis of Ni content in Soybean shoot showed significant results on the interaction between kind of organic matter and urea dose on nickel contaminated soil. The application of urea without organic matter / compost on nickel contaminated soil resulted in higher nickel accumulation in shoot compared with urea application with compost in soybean. Increased doses of urea greater than 100 kg / ha without compost application also suppressed the amount of nickel in soybean canopy (Fig. 4).

Furthermore, compost applied without urea was likely make low nickel in soybean shoot because nickel in the soil was bound by compost or organic material applied as a treatment. The factor which contributes to the nickel content in soils is the presence of organic matter. The high content of organic matter can also reduce nickel toxicity by removing this metal as a chelate complex [14].

The addition of urea and compost in nickel contaminated soil resulted in decrease of the amount of nickel content in soybean tissue both in the shoot and the root. In contrast, the application of urea without or with compost showed higher nickel content in the Melastoma shoot than nickel content in soybean shoot. The application of Chromolaena and Gliricidia compost was able to increase the nickel content in melastoma canopy compared to without compost and husk compost. The highest nickel content in the Melastoma canopy was

shown on applied of Chromolaena compost and urea 300 kg / ha that was equal to 634,53 mg/kg (Fig. 4).

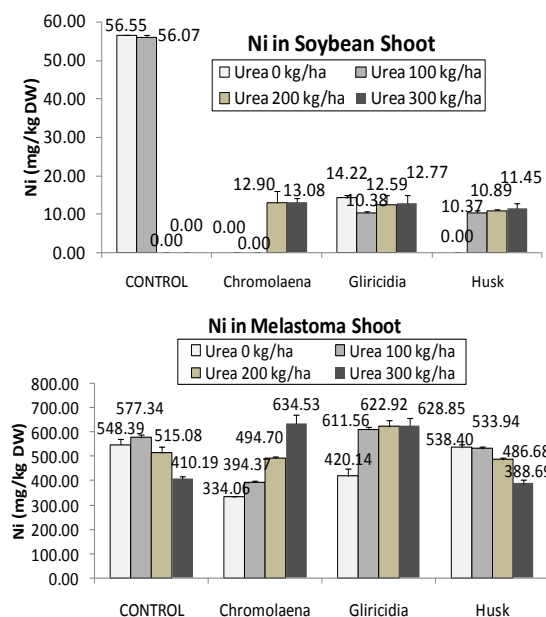


Figure 4. Ni accumulation of soybean (above) and melastoma (under) shoot. Vertical bars represent SE.

Similar to soybean canopy, the nickel content in the roots of soybean and Melastoma were also higher when urea was given without compost and nickel content decrease when urea application was given with compost on nickel contaminated soil (Fig. 5). This fact shows that exudate compounds can increase the absorption of Ni in intercropping Melastoma with soybean or other kind of nuts [11], [15]. The results of this study indicate that intercropping allows to be an option in the phytoremediation practice of nickel contaminated soil.

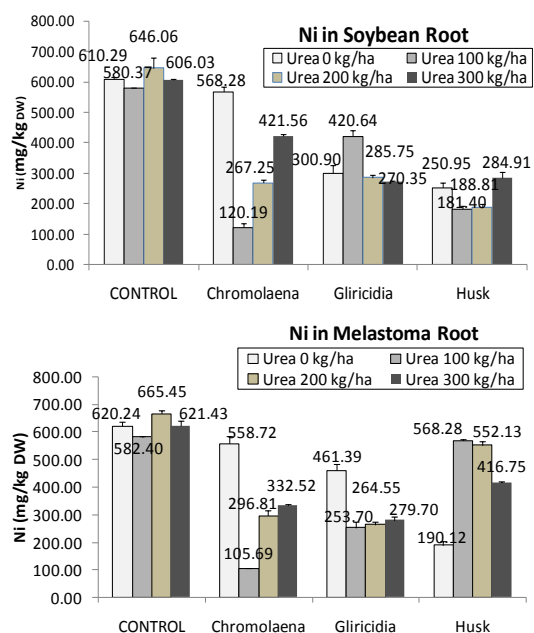


Figure 5. Ni accumulation of soybean (above) and melastoma (under) shoot. Vertical bars represent SE.

C. Bioconcentration Factor (BCF)

The potential for plant remediation, known as Bioremediation or Phytoremediation, is the ability of plants to reduce contaminated land or soil. Bioremediation can be calculated through Bioaccumulation or Bioconcentration Factor (BCF) which indicates the ability of plants to absorb heavy metals from the soil. BCF is calculated by comparing the content of the plant metal with the metal content in the soil [12]. To obtain the value of BCF Ni in Soybean and Melastoma plants, each Ni content value in the crown and root of both plants is multiplied by the biomass or dry weight of each part of the plant. Similarly, the Ni content available (available) in the soil is multiplied by the dry weight of the soil used as the planting medium. Both values obtained are used to calculate BCF [8].

Grouping of crops based on their ability to absorb heavy metals and their sensitivity to heavy metal contamination is divided into 4 groups of plant species [16] namely: BCF value between 1-10 is called high accumulator plant (Hyperaccumulator); 2) BCF 0.1-1 are called moderate accumulators; 3) 0.01-0.1 are called low accumulator plants and 4) <0.01 are called nonaccumulator plants.

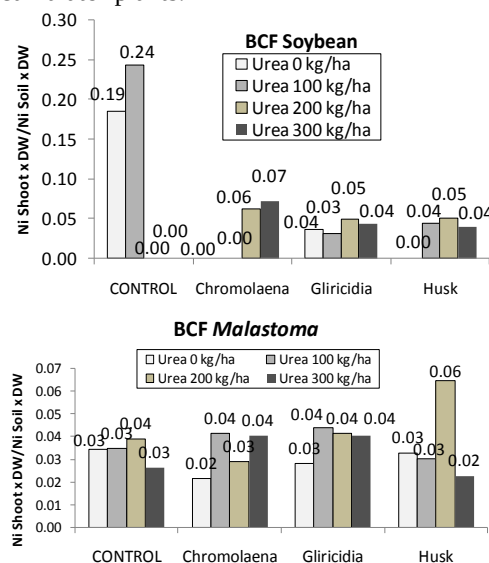


Figure 6. Bioconcentration Factor (BCF) on of soybean (above) and melastoma (under).

Soybean BCF values obtained from urea and compost applications on nickel contaminated soil produce BCF which belongs to low accumulator category, except without urea, 100 kg / ha urea and no compost which includes BCF moderate category accumulator. Melastoma BCF values obtained from urea and compost applications on nickel contaminated soil produce BCF which all belong to the low accumulator category (Fig. 6). The highest BCF value was shown on the application of urea 100 kg / ha without compost that is 0.24 which belongs to moderate accumulator group.

The results of this study indicate that soybeans and Melastoma are able to remove heavy metals from soil and then translocated from root to plant canopy only by urea

application. Therefore, cultivation of crops on contaminated soil makes it possible to do by using compost to reduce heavy metal absorption in planted crops.

IV. CONCLUSION

The study concluded that:

- 1) The application of urea and compost, especially Chromolaena and Glyricidia compost increased the dry weight of soybeans and Melastoma.
- 2) The application of urea and all types of compost resulted in decrease of nickel accumulation in both soybean and Melastoma.
- 3) The application of urea 100 kg /ha without compost gives BCF value of 0.24 on soybean and was categorized as moderate accumulator

ACKNOWLEDGEMENT

The authors wish to thank to Agency and Agricultural Development - Ministry Of Agriculture which has funded the Cooperation of National Agricultural Research and Development (KKP3N) activities in 2014. KKP3N is a program of agricultural research and development cooperation with universities and government research institutions.

REFERENCES

- [1] K. S. Sajwan, W. H. Ornes, T. V. Youngblood, and A. K. Alva, "Uptake of soil applied cadmium, nickel and selenium by bush beans," *Water, Air and Soil Pollution*, vol. 91, pp. 209-217, 1996.
- [2] M. Muchuweti, J. W. Birkett, E. Chinyanga, R. Zvauya, M. D. Scrimshaw, and J. N. Lester, "Heavy metal content of vegetables irrigated with mixture of wastewater and sewage sludge in Zimbabwe: Implications for human health," *Agriculture, Ecosystem and Environment*, vol. 112, p. 41e48, 2006.
- [3] M. A. Jiafang, M. E. N. G. Guangtao, H. E. Liping, and L. I. Guixiang, "Research progress of artificial forest in the remediation of heavy metal contaminated soils," *IOP Conf. Series: Earth and Environmental Science*, vol. 51, 2017.
- [4] L. Marchiol, P. Sacco, S. Assolari, and G. Zerbi, "Reclamation of polluted soil: Phytoremediation potential of crop-related Brassica species," *Water Air Soil Pollut.*, vol. 158, pp. 345-356, 2004.
- [5] M. A. Eissa, M. F. Ghoneim, A. Galal, El-Gharably, and M. A. El-Razek, "Phytoextraction of nickel, lead and cadmium from metals contaminated soils using different field crops and EDTA," *World Applied Sciences Journal*, vol. 32, no. 6, pp. 1045-1052, 2014.
- [6] A. J. Baker, S. P. McGrath, R. D. Reeves, and J. A. Smith, "Metal hyperaccumulator plants: A review of the ecology and, physiology of a biological resource for phytoremediation of metal-polluted soils," in *Phytoremediation of Contaminated Soil and Water*, N. Terry, G. Banuelos, and J. Vangronsveld, Eds., Boca Raton, FL, USA: Lewis Publisher, 2000, pp. 85-107.
- [7] M. Poniedzialek, A. Sękara, E. Jędraszczyk, and J. Ciura. Phytoremediation efficiency of crop plants in removing cadmium, lead and zinc from soil. *Folia Horticulturae Ann.* [Online]. 22/2 (2010): 25-31. Available: <http://www.ptno.ogr.ar.krakow.pl/Wydawn/FoliaHorticulturae/Spisy/FH2010/PDF22022010/fh2202p04.pdf>
- [8] S. Netty, T. Wardiyati, M. D. Maghfoer, and E. Handayanto, "Effect of accumulator plants on growth and nickel accumulation of soybean on metal-contaminated soil," *Agriculture and Agricultural Science Procedia*, vol. 9, pp. 13-19, 2016.
- [9] E. O. Nwaichi and O. P. Dhankher, "Heavy metals contaminated environments and the road map with phytoremediation," *Journal of Environmental Protection*, vol. 7, pp. 41-51, 2016.
- [10] M. Tejada, J. L. Moreno, M. T. Hernandez, C. Garcia, "Soil amendments with organic wastes reduce the toxicity of nickel to soil enzyme activities," *Eur. J. Soil Biol.*, vol. 44, p. 129, 2008.
- [11] S. Netty, T. Wardiyati, E. Handayanto, and M. D. Maghfoer, "Nickel accumulating plants in the post-mining land of Sorowako, South Sulawesi, Indonesia," *Journal of Tropical Agriculture*, vol. 50, no. 1-2, pp. 45-48, 2012.
- [12] M. Ghosh and S. P. Singh, "Comparative content and phytoremediation study soil induced chromium by accumulator and high biomass species," *Applied Ecology and Environmental Research*, vol. 3, no. 2, pp. 67-79, 2005.
- [13] J. Kucharski, E. Boros, and J. Wyszowska, "Biochemical activity of nickel-contaminated soil," *Polish J. of Environ. Stud.*, vol. 18, no. 6, pp. 1039-1044, 2009.
- [14] D. L. Eskew, R. M. Welch, and W. A. Norvell, "Nickel, in higher plants. Further evidence for an essential role," *Plants Physiology*, vol. 76, pp. 691-693, 1984.
- [15] T. Watanabe and M. Osaki, "Mechanisms of adaptation to high aluminum condition in native plant species growing in acid soils: A review," *Communication in Soil Science and Plant Analysis*, 2002.
- [16] B. E. Malayeri, A. Chehregani, N. Yousefi, and B. Lorestani, "Identification of the hyper accumulator plants in copper and iron mine in Iran," *Pakistan J. Biol. Sci.*, vol. 11, no. 3, p. 490, 2008.



Netty Syam is a permanent lecturer at the Faculty of Agriculture of Muslim University of Indonesia, Makassar since 1988. An undergraduate degree in Agriculture was obtained from the Faculty of Agriculture of Hasanuddin University in 1987. In 1996 he obtained a Master's degree from Bogor Agricultural Institute and in 2013 obtained his Doctor of Agricultural Science from Brawijaya University. Some of her researches in the field of agronomy and phytoremediation of post nickel mines have been published in various national and international journals and a book has been produced alongside other authors entitled Phytoremediation and phytomining of heavy metals soil contaminants.