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ANNOUNCEMENT OF THE MASTER'S EXAMINATION

of

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in defense of her Master's thesis

MORPHO-PHYSIOLOGICAL RESPONSES, SECONDARY METABOLITE PROFILES AND ANTIOXIDANT ACTIVITY OF *ORIGANUM VULGARE* L. EXPOSED TO VARYING AMMONIUM SULFATE LEVELS

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MORPHO-PHYSIOLOGICAL RESPONSES, SECONDARY METABOLITE PROFILES AND SELECTED BIOACTIVITIES OF *Origanum vulgare* L. EXPOSED TO VARYING AMMONIUM SULFATE LEVELS

Medicinal plants are consumed by approximately 80% of the people in developing countries. To increase shoot production, ammonium-based (NH4+-N) fertilizers are commonly applied. This is so since primary metabolism, observed as plant growth, is enhanced when the amount of nitrogen (N) is sufficient. However, Carbon/Nutrient Balance (CNB) hypothesis states that the production of plant secondary metabolites, such as carbon-based phenolic compounds known to exhibit antioxidative properties, is enhanced when N supply is limiting. Excess N application, therefore, may be considered as an avertible production practice cost. Moreover, there are also associated environmental costs of fertilizer production and ecosystem changes by NH4+-N or its derivative, nitrate (NO3--N).

This study investigated the effects of increasing NH4+-N concentrations on morphophysiological parameters and secondary metabolite production of Origanum vulgare L. through the application varying amounts of ammonium sulfate in a modified Hoagland solution with fixed amount of NO3--N (NH4+-N : NO3--N of 0, 1, 2.5, 4, 5.75, 7.75). Based on the results, the concentrations of chlorophyll b, carotenoids and anthocyanin, which are important for photosynthesis, showed significant decreasing trends as NH4+-N : NO3--N increases ($p < 0.01$-0.05). There was a significant increasing trend for the chlorophyll a to chlorophyll b ratio ($p < 0.01$), which was up to the NH4+-N : NO3--N of 5.75. The results further revealed very strong correlations among concentrations of photosynthetic pigments ($p < 0.01$-0.05) and between concentrations of chlorophylls and leaf area ($p < 0.05$), probably demonstrating the plants' coordinated responses to optimize photosynthesis, However, there was no significant effect of increasing the NH4+-N : NO3--N level on change in leaf biomass, indicating that plants were under stress when supplied with higher NH4+-N amounts. Moreover, there was a very strong correlation between concentrations of chlorophylls and TPC ($p < 0.05$), pointing that allocation of energy and carbon from photosynthesis favored secondary metabolism over primary metabolism. Nonetheless, more energy and carbon may have been spent to alleviate NH4+-N toxicity, hence validating the CNB hypothesis. Further, the antioxidative property, measured as DPPH scavenging activity ($p < 0.01$), and the total phenolic content ($p < 0.05$) had significant decreasing trends as NH4+-N : NO3--N increases, showing a very strong correlation between the two parameters ($p < 0.01$), but not between DPPH scavenging activity and total flavonoid content (TFC). This would indicate that phenolic compounds, besides flavonoids, have contributed to the DPPH scavenging activity of the O. vulgare leaf extract.

Overall, the results of the study demonstrated that higher NH4+-N levels did not significantly affect shoot growth, instead it significantly decreased the concentrations of chlorophyll b, carotenoid and anthocyanin, as well as significantly decreased the TPC and DPPH scavenging activity in O. vulgare leaves. Further studies may consider the following recommendations: application of more NH4+-N : NO3--N levels to better understand the NH4+-N dose-response of O. vulgare; testing the effects of NO3--N as alternative N source; and employing mass spectrometry to better identify and quantify the bioactive secondary metabolites of interest.