

***In-Vitro* Anticoagulant Activity of Methanolic Extract of *Dialium ovoideum* (Thwaites): A Preliminary Study**

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Background: *Dialium ovoideum* (Thwaites) is an endemic medicinal plant widely used in indigenous medicine in Sri Lanka. However, its potential anticoagulant effect on human blood remains unexplored. Investigating its influence on blood coagulation pathways may provide preliminary evidence for its therapeutic application as a natural anticoagulant source.

Objective: To evaluate the *in vitro* anticoagulant activity of methanolic extract of *D. ovoideum* bark and screen its phytochemical constituents.

Method: This preliminary laboratory-based experimental study used fresh, healthy bark of *D. ovoideum*, authenticated at the Department of Plant and Molecular Biology at the University of Kelaniya. The barks were shade-dried, powdered, and extracted with methanol using a Soxhlet apparatus (40 g powder: 250 mL methanol; 1:6.25). The solvent was evaporated and reconstituted with Tris buffer (pH 7.4) to prepare four concentrations. Preliminary phytochemical screening was conducted using standard qualitative tests. Venous blood samples were collected from six healthy volunteers in sodium-citrate tubes and pooled plasma was separated by centrifugation at 3000 rpm for 15 minutes. Anticoagulant activity was assessed by Prothrombin time (PT) assay using a HumaClot Duo Plus coagulometer. Positive and negative controls were included. All tests were performed in triplicate. Statistical analysis was performed using one-way ANOVA in SPSS ($p \leq 0.05$).

Results: The methanolic bark extract yielded 13.82% (w/w). Phytochemical screening revealed phenolics, tannins, flavonoids, alkaloids, saponins, and glycosides. The extract prolonged PT in a concentration-dependent manner to 7.00 ± 0.12 , 13.07 ± 0.29 , 24.57 ± 0.40 , and 25.73 ± 0.12 seconds at 156.25, 312.5, 625, and 1250 $\mu\text{g/mL}$, respectively. All concentrations showed a significantly increased PT compared to the control (15.8 seconds) ($p < 0.05$).

Conclusion: These findings provide preliminary evidence that *D.ovoideum* bark extract exhibits concentration-dependent *in vitro* anticoagulant activity on pooled human plasma.

Keywords: anticoagulant activity, *dialium ovoideum*, methanolic bark extract, prothrombin time test, phytochemicals