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## Liquid–liquid extraction method developed for thymoquinone from seed powder of *Nigella Sativa*, characterized it by UV-spectrophotometer

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**N**igella Sativa Linn. (Ranunculaceae) (*N. sativa*), commonly known as black seed or black cumin, is an herbaceous plant. Liquid–Liquid extraction method developed, the seed powder *Nigella sativa* was packed in a muslin cloth and placed in a beaker containing sufficient quantity of methanol for 72 hrs. Thereafter the methanolic extracts were filtered through Whatman filter paper no. 42 and the resultant filtrates were concentrated under reduced pressure using rotary evaporator. Characterize by UV-Spectrometer. The  $\lambda_{max}$  of thymoquinone is found 254 nm and confirm it by pure compound of thymoquinone. The accuracy of recovery studies by the standard addition technique was carried out by adding 50, 100 and 150% of the thymoquinone concentration in the sample. The % recoveries of the three concentrations were found to be (99.95–101.68) & % RSD (1.63-1.02). The precision method was assessed by analyzing thymoquinone in three different concentrations as 10, 25 and 40

$\mu\text{g mL}^{-1}$  of thymoquinone. Repeatability (intra-day) was assessed by analyzing thymoquinone in three different concentrations (10, 25 and 40  $\mu\text{g mL}^{-1}$ ) three times a day the % RSD (1.83-0.91). Intermediate precision (inter-day) was established by analyzing three different concentrations (10, 25 and 40  $\mu\text{g mL}^{-1}$ ) of thymoquinone for three different days. % RSD (1.85-0.93). The low values of % RSD for repeatability and intermediate precision suggested an excellent precision of the developed UV spectrophotometric method. The optical, linear regression and validation data of UV spectrophotometry for the quantification of thymoquinone in methanol-Optical characteristics  $E_{1\%}^{1\text{cm}}$  (201.82 $\pm$ 2.43) Regression analysis-Slope (0.0204 $\pm$ 0.0002), Intercept (0.0062 $\pm$ 0.0012), Regression coefficient ( $R^2$ ) (0.9984 $\pm$ 0.0003), Validation- Range (5-50  $\mu\text{g mL}^{-1}$ ), Detection limit (DL) (0.99  $\mu\text{g mL}^{-1}$ ), Quantitation limit (QL) (2.89  $\mu\text{g mL}^{-1}$ ).

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