APPLICATION OF LIQUID CHROMATOGRAPHY WITH UV DETECTION FOR DETERMINATION OF PLANT GROWTH REGULATOR IN COMMON PLANT FERTILIZERS

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INTRODUCTION

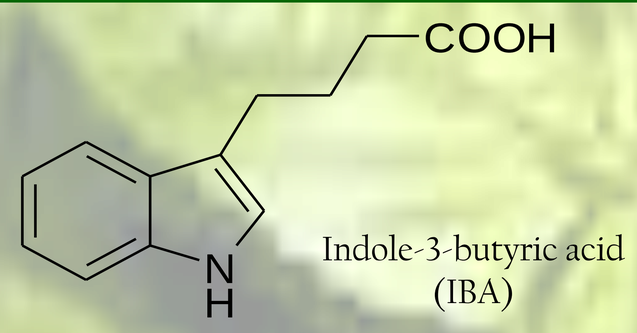
A simple and rapid HPLC-based method was developed for the determination of plant growth regulator in plant fertilizer samples. Plant hormones including indole-3-butyric acid (IBA) are structurally diverse compounds that play an important role in a variety of processes related to plant growth and development. As a minor component of the metabolome, phytohormones play a particular role in the regulation of cell division, enlargement and differentiation, organ formation, seed dormancy and germination [1, 2, 3]. In most cases phytohormones exist in plants either by endogenous secretion or exogenous treatment to achieve various enhanced agriculture characteristics during some critical growth stages. Besides, plant hormones could stimulate the human body or at least affect their immune cells [4]. Therefore, it is necessary to develop a simple and effective methods to detect phytohormones in plant fertilizer samples.

The separation was carried out on Zorbax SB C18 column (4.6 x 150 mm, 5 µm), using acetonitrile/water containing 0.2% acetic acid, pH=2.65 (60:40, v/v) as the isocratic mobile phase at the flow-rate of 1.0 ml/min and column temperature 25 °C. The indole-3-butyric acid was determined with UV detection at λ = 280 nm. The analyte was eluted within 2.7 min. Validation experiments showed that the optimized method has good linearity within the range 1 – 50 nmol/ml, R^2 = 0.9993 and high recovery (96.4 % - 114.4 %). The detection limit based on a signal-to-noise ratio was 0.15 nmol/ml. The results indicate that the novel method has advantages of convenience, good sensitivity, high efficiency, and can be useful for the determination of indole-3-butyric acid in common plant fertilizer samples (Clonex, Korzonek Z, Korzonek S).

RESULTS

Tab. 1. HPLC CONDITIONS

HPLC Column	Zorbax SB C18 (4.6 × 150 mm, 5 µm)
Mobile phase 0.2% CH_3COOH pH=2.65 : MeCN	40 : 60
Flow rate [ml·min^{-1}]	1
Column temperature [°C]	25
UV detection [nm]	280



Indole-3-butyric acid (IBA)

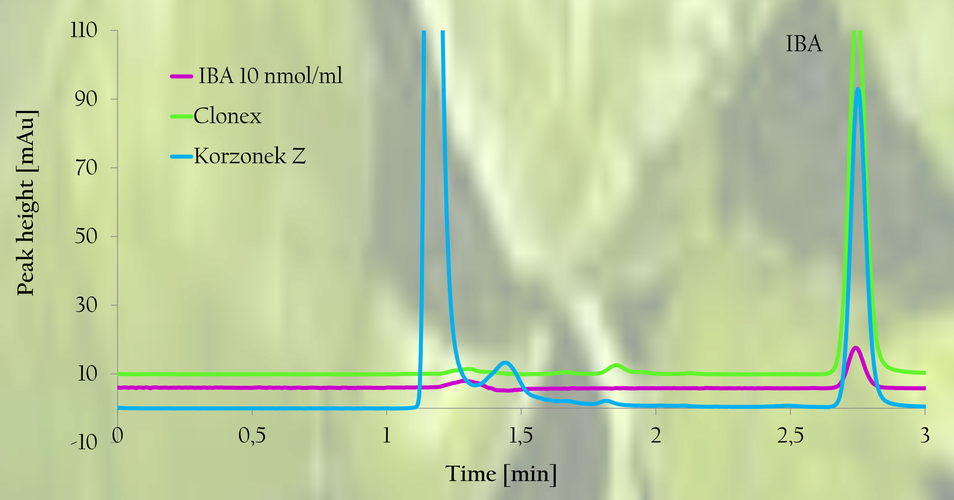


Fig. 1. Representative HPLC chromatogram of standard solution and common plant fertilizer samples. HPLC conditions described in Tab. 1.

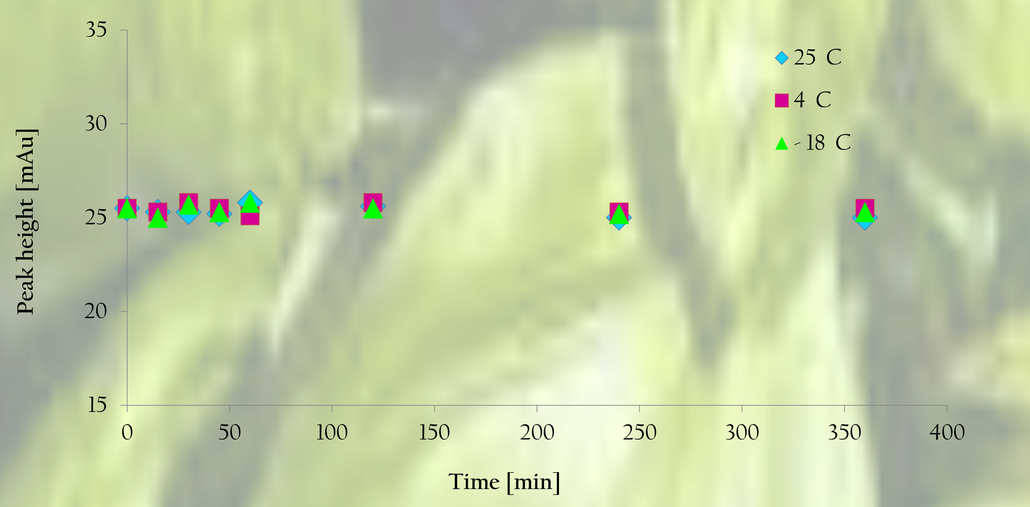


Fig. 2. Studies of IBA stability in -18, 4 and 25 °C. HPLC conditions described in Tab. 1.

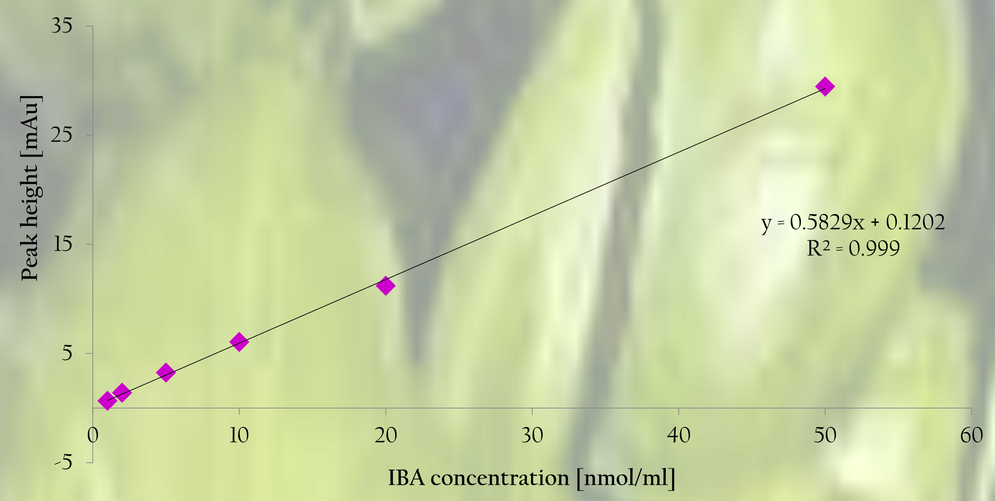


Fig. 3. Calibration curve of IBA in plant fertilizer.

Tab. 2. VALIDATION OF THE METHOD

Linearity	y = 0.5829x + 0.1202 R^2 = 0.9993
Precision [%]	0.44 – 5.83
Recovery [%]	96.4 – 114.4
LOD (signal to noise ratio 3 : 1)	0.15 nmol/ml
LOQ (signal to noise ratio 6 : 1)	0.6 nmol/ml

CONCLUSIONS

1. The methodology allows facile and accurate determination of IBA in common plant fertilizer samples.
2. The analytical figures of linearity, precision and recovery shown during the method validation procedure are well within the criteria for biological sample analysis.
3. In temperature -18, 4 and 25 °C the analyte stays stable for 6 h.
4. The assay should help in control of phytohormones concentration in plants and fertilizers.

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