

High Reproducibility by Cell Temperature Control of Fluorescence Detector FP-4020

Introduction

Indole-3-acetic acid known as auxin, a growth inhibitor which play a role in morphosis in plants. Artificially synthesized auxin is widely used as a herbicide and a fruit growth promoter, and is expected to make a significant contribution in increasing research into increasing crop yields and biomass.

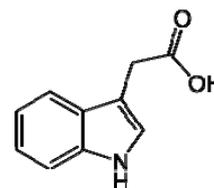
Generally, the intensity of fluorescence in a compound is temperature dependent. Indole-3-acetic acid is a compound whose fluorescence intensity is decreased with a rise of temperature.

In this application note, indole-3-acetic acid was measured using an HPLC with FP-4020 fluorescence detector with a temperature controlled cell, and the effects of fluctuation in room temperature and the benefits of cell temperature control are reported.

Keyword: Indole-3-acetic acid, FP-4020, Cell temperature control, fluorescence detection, HPLC

Experimental Condition

Column: CrestPak C18S
(4.6 mmI.D. x 150 mmL, 5 μ m)
Eluent: 0.3% acetic acid in methanol/water (42/58)
Flow rate: 1.0 mL/min
Column temp.: 40°C
Wave length: Ex 280 nm, Em 360 nm, Gain x100
Cell temp.: 30°C and Off
Injection volume: 5 μ L
Standard: 100 ng/mL Indole-3-acetic acid



Structure of Indole-3-acetic acid

Results

Figure 1 Peak height change of indole-3-acetic acid with rise in room temperature.

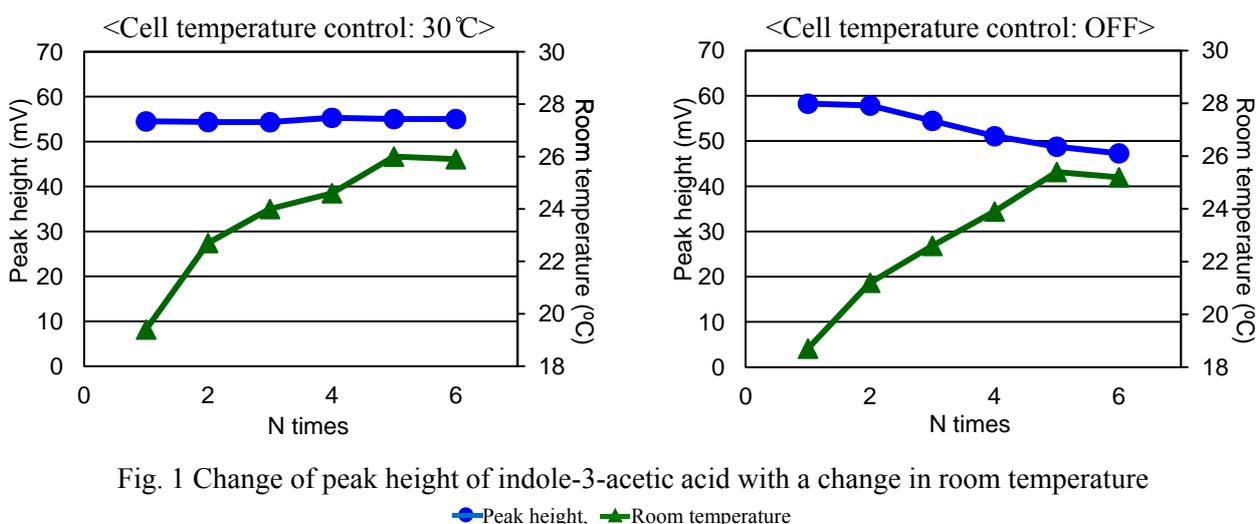


Fig. 1 Change of peak height of indole-3-acetic acid with a change in room temperature

● Peak height, ▲ Room temperature

Figure 2 Chromatograms of indole-3-acetic acid with and without cell temperature control.

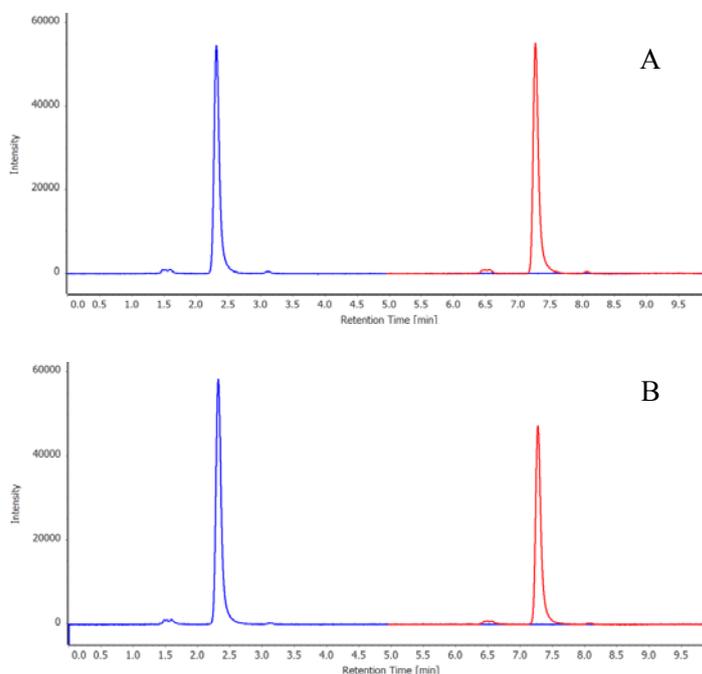


Fig.2 Chromatograms of indole-3-acetic acid with and without cell temperature control

A: Cell temperature control: ON (30°C) (Room temperature Blue: 19.4°C, Red: 26.0°C)
 B: Cell temperature control; OFF (Room temperature Blue: 18.7°C, Red: 25.4°C)

Table 1 Change rate of peak height and the relative standard deviation with and without cell temperature control. As a result of using cell temperature control, the peak intensity is stabilized minimizing the effect of fluctuations in room temperature providing improved reproducibility.

	Temperature change (°C)	Rate of change (%)	%RSD
Cell temperature control: ON (30°C)	6.6	0.95	0.75
Cell temperature control; OFF	6.7	-19.49	8.77

Table 1 The rate of change in peak height and the relative standard deviation
 With and without cell temperature control

*Rate of change: The change rate of peak height.
 The peak height at minimum room temperature is normalized as "1", as the reference value.
 *%RSD: The relative standard deviation of the peak height at six repeat measurement .