

การหาปริมาณกรดจิบเบอเรลลิกโดยเทคนิคอะฟิลาเรียเล็กโทรฟอริซิส



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วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต

สาขาวิชาเคมี ภาควิชาเคมี

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ปีการศึกษา 2546

ISBN 974-17-3656-8

ลิขสิทธิ์ของจุฬาลงกรณ์มหาวิทยาลัย

DETERMINATION OF GIBBERELLIC ACID BY
CAPILLARY ELECTROPHORESIS



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A Thesis Submitted in Partial Fulfillment of the requirements
for the Degree of Master of Science in Chemistry

Department of Chemistry

Faculty of Science

Chulalongkorn University

Academic Year 2003

ISBN 974-17-3656-8

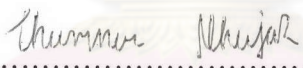
Thesis Title Determination of Gibberellic Acid by Capillary Electrophoresis
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Accepted by the Faculty of Science, Chulalongkorn University in Partial
Fulfillment of the Requirements for the Master's Degree

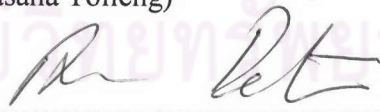

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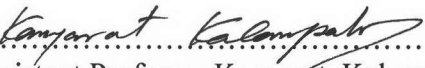
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มนพิชา ศรีสะอาด: การหาปริมาณกรดจิบเบอเรลลิกด้วยเทคนิคอะพอลาร์อิเล็กโทรโฟรีซิส. (DETERMINATION OF GIBBERELIC ACID BY CAPILLARY ELECTROPHORESIS) อาจารย์ที่ปรึกษา: ดร. ธรรมบุญ หนูจักร, อาจารย์ที่ปรึกษาร่วม: อาจารย์วาสนา โดเลี้ยง 76 หน้า. ISBN 974-17-3656-8

ได้พัฒนาเทคนิคไมเซลล์าร์อิเล็กโทรโฟรีซิสโครมาโทกราฟีเพื่อใช้เป็นวิธีหาปริมาณกรดจิบเบอเรลลิกในน้ำหมักและในผลิตภัณฑ์ที่มีจำหน่ายในท้องตลาด โดยใช้โซเดียมเทรโบเรตเป็นบัฟเฟอร์ที่ค่าพีเอช 9.2 และโซเดียมโดเดซิลซัลเฟตเป็นไมเซลล์าร์เฟส การแยกของพิกของกรดจิบเบอเรลลิกจากพิกของสารอื่น ๆ ในน้ำหมักประสบความสำเร็จโดยใช้สถานะของไมเซลล์าร์อิเล็กโทรโฟรีซิสโครมาโทกราฟีดังนี้: ความเข้มข้นของบอเรตบัฟเฟอร์ 25 มิลลิโมลาร์ ความเข้มข้นของโซเดียมโดเดซิลซัลเฟต 100 มิลลิโมลาร์ และใช้ศักย์ไฟฟ้าในการแยก +30 กิโลโวลต์ พบว่าปริมาณของกรดจิบเบอเรลลิกที่วิเคราะห์โดยเทคนิคไมเซลล์าร์อิเล็กโทรโฟรีซิสโครมาโทกราฟีสอดคล้องกับที่วิเคราะห์โดยเทคนิคไฮเพอร์ฟอร์แมนซ์ลิกวิดโครมาโทกราฟี นอกจากนี้ยังประยุกต์เทคนิคไมเซลล์าร์อิเล็กโทรโฟรีซิสโครมาโทกราฟี เพื่อใช้ในการหาปริมาณกรดจิบเบอเรลลิกในผลิตภัณฑ์ที่มีจำหน่ายในท้องตลาด ข้อดีของเทคนิคนี้ คือ ใช้เวลาในการวิเคราะห์เร็ว ให้ความถูกต้องและความเที่ยงสูง และไม่ต้องมีขั้นตอนการเตรียมสารตัวอย่าง ยกเว้นการกรองและการเจือจาง

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ภาควิชา.....เคมี..... ลายมือชื่อผู้สิด.....
สาขาวิชา.....เคมี..... ลายมือชื่ออาจารย์ที่ปรึกษา.....
ปีการศึกษา.....2546..... ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

447 23711 23 : MAJOR CHEMISTRY

KEY WORD: GIBBERELIC ACID/ CAPILLARY ELECTROPHORESIS/
MICELLAR ELECTROKINETIC CHROMATOGRAPHY

MONPICHAR SRISA-ART: DETERMINATION OF GIBBERELIC
ACID BY CAPILLARY ELECTROPHORESIS. THESIS ADVISOR:
THUMNOON NHUJAK, Ph.D., THESIS COADVISOR: VASANA
TOLIENG, 76 pp. ISBN 974-17-3656-8.

Micellar electrokinetic chromatography (MEKC) was developed as a method for quantitative determination of gibberellic acid (GA_3) in fermentation broth, using disodium tetraborate ($Na_2B_4O_7$) as a buffer at pH 9.2 and sodium dodecylsulfate (SDS) as a micellar phase. Baseline resolution of GA_3 from other compounds in the fermentation broth was achieved using the following MEKC conditions: 25 mM $Na_2B_4O_7$, 100 mM SDS and separation voltage of +30 kV. The amount of GA_3 in the fermentation broths determined by MEKC was found to be in good agreement with that by high performance liquid chromatography. The MEKC method was also applied for quantitative determination of GA_3 in commercial products. Advantages of this method include fast analysis, high accuracy and precision and no sample preparation except for filtration and dilution.

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ACKNOWLEDGEMENTS

First of all, I would like to express my deepest gratitude to my advisor, Dr Thumnoon Nhujak, for his professionalism, discerning suggestion, forbearance, encouragement, and critical reading. I also greatest thank to my co-advisor, Vasana Tolieng, for her helpfulness, generous guidance and encouragement throughout this research. Special thanks are extended to the thesis committee for their valuable comments.

This research was financially supported by the Grants for Development of New Faculty Staff to Dr Thumnoon Nhujak, Department of Chemistry and the Graduate School, Chulalongkorn University. Research facilities provided by the Institute of Biotechnology and Genetic Engineering (IBGE) are appreciatively and thankfully acknowledge.

In addition, my scholarship provided by the Development and Promotion of Science and Technology Talent Project (DPST) has been greatly valuable to my study and research.

Futhermore, I specially thank to all members of Separation and Chromatography Research Group for their valuable suggestions. Also, I am thankful to all members of CE group for their helpfulness and encouragement. Moral supports and cheerful willingness of my friends are truly appreciated.

Finally, I am eternally grateful to my beloved parents and the rest of family members for their enormously ever-willing support. Thanks are also extended to everyone who has contributed suggestions and supports throughout this research.

Monpichar Srisa-art

CONTENTS

	PAGE
ABSTRACT (IN THAI).....	iv
ABSTRACT (IN ENGLISH)	v
ACKNOWLEDGEMENTS	vi
CONTENTS	vii
LIST OF TABLES	x
LIST OF FIGURES.....	xi
LIST OF ABBREVIATIONS AND SYMBOLS.....	xiv
CHAPTER I : INTRODUCTION	1
1.1 Introduction to Capillary Electrophoresis	1
1.2 Basic Theories of CE.....	1
1.2.1 Electrophoretic mobility.....	1
1.2.2 Electroosmotic flow	3
1.2.3 Flow profile and migration behaviour of analytes in CE	5
1.2.4 Peak shape, efficiency and resolution in CE	7
1.2.5 Peak dispersion in CE.....	9
1.3 CE Instrumentation.....	13
1.3.1 Basic apparatus of CE	13
1.3.2 Sample introduction	14
1.3.3 Detection	16
1.4 Modes of CE.....	17
1.5 Quantitative Analysis in CE.....	21
1.5.1 Peak area and corrected peak area.....	21
1.5.2 Internal standard	21
1.6 Quantitative Analysis of Gibberellic Acid	22
1.7 Aims and Scope.....	24
CHAPTER II : EXPERIMENTAL	26
2.1 Chemicals and Materials	26
2.2 CE Conditions and Optimisation.....	27
2.2.1 CE conditions and optimisation	27

	PAGE
2.2.2	CZE optimisation 27
2.2.3	MEKC optimisation 28
2.2.4	Stability of GA ₃ in water and diluted BGE 28
2.3	Validation of MEKC Method..... 28
2.3.1	Calibration plot..... 28
2.3.2	Accuracy and precision 29
2.3.3	Limit of detection (LOD) and limit of quantitation (LOQ).... 29
2.4	HPLC Analysis..... 29
2.4.1	HPLC conditions 29
2.4.2	Sample preparation..... 30
2.4.3	Calibration plot..... 30
2.4.4	Recovery of GA ₃ extraction 30
2.4.5	Limit of detection (LOD) and limit of quantitation (LOQ).... 31
2.5	Applications to Real Samples..... 31
2.5.1	Determination of GA ₃ in fermentation broth by MEKC and HPLC 31
2.5.2	Determination of GA ₃ in commercial products by CZE and MEKC..... 31
CHAPTER III : RESULTS AND DISCUSSION 32	
3.1	CE Conditions and Optimisation..... 32
3.1.1	CE conditions 32
3.1.2	CZE optimisation 34
3.1.3	MEKC optimisation 45
3.1.4	Stability of GA ₃ in water and diluted BGE 52
3.2	Validation of MEKC Method..... 54
3.2.1	Calibration plot..... 54
3.2.2	Accuracy and precision 55
3.2.3	Limit of detection (LOD) and limit of quantitation (LOQ).... 58
3.3	HPLC Analysis..... 58
3.3.1	Recovery of GA ₃ extraction 58
3.3.2	Limit of detection (LOD) and limit of quantitation (LOQ).... 61

	PAGE
3.4 Applications to Real Samples.....	62
3.4.1 Determination of GA ₃ in fermentation broth by MEKC and HPLC	62
3.4.2 Determination of GA ₃ in commercial products by CZE and MEKC.....	66
CHAPTER IV : CONCLUSIONS AND FUTURE WORK.....	70
REFERENCES.....	72
BIBLIOGRAPHY	75
VITA	76



ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

LIST OF TABLES

TABLES	PAGE
3.1 Observed and predicted peak variances (σ^2) of GA ₃	44
3.2 The percentage of σ^2_{EMD} (P_{EMD}) and peak asymmetry factor (A_{asym}) of GA ₃	44
3.3 Peak efficiency (N) of GA ₃ and resolution (R_s) of GA ₃ and unknown peak 2	44
3.4 Effect of SDS concentration on peak efficiency (N) of GA ₃ and resolution (R_s) for GA ₃ :unknown 2 and GA ₃ :unknown 4	51
3.5 The determined and actual amounts of GA ₃ spiked in water and the fermentation broths diluted 10 times	56
3.6 Within-day migration time, electrophoretic mobility and corrected peak area of GA ₃ . MEKC conditions as shown in Figure 3.11	57
3.7 Day-to-day migration time, electrophoretic mobility and corrected peak area of GA ₃ ($n = 5$ days). MEKC conditions as shown in Figure 3.11... ..	57
3.8 The determined and actual amounts of GA ₃ spiked in 5 and 10-day fermentation broths obtaining from HPLC	61
3.9 A comparison of GA ₃ in the broths determined by MEKC and HPLC	65
3.10 A comparison of GA ₃ in the tablet of GA ₃ determined by CZE and MEKC.....	69
3.11 A comparison of GA ₃ in the tube of GA ₃ determined by CZE and MEKC.....	69

จุฬาลงกรณ์มหาวิทยาลัย

LIST OF FIGURES

FIGURES	PAGE
1.1 Migration behaviour of each species.....	1
1.2 Electroosmotic flow (EOF)	3
1.3 A model of a double electric layer and electrical potential.....	4
1.4 Flow profiles and peaks in CE and HPLC	5
1.5 Migration behaviour of analytes.....	6
1.6 Gaussian peak.....	7
1.7 Concentration distribution of EMD in the distance domain.....	11
1.8 Schematic diagram illustrating concentration distribution in the presence of EMD. Evaluation (i) tailing and (ii) fronting peaks in the distance domain as a function of time (min)	11
1.9 Basic schematic of CE instrument.....	13
1.10 Sample stacking.....	16
1.11 Separation mechanisms in (a) CZE and (b) MEKC	17
1.12 Schematic illustration of surfactant and micelle	18
1.13 Typical elution order for analytes in MEKC.....	19
1.14 Structure of GA ₃	22
1.15 <i>CU Gibb.</i> (product of IBGE, Chula)	25
3.1 An example of electropherograms of the 10-day fermentation broth diluted 10 times using separation voltage of +25 kV and BGE concentrations of (a) 12.5, (b) 25.0, (c) 40.0 and (d) 50.0 mM. Peak 1 = GA ₃ , 2 = unknown, 3 = AMBA and 4 = unknown. Other CE conditions: uncoated fused silica capillary 50 μm I.D. × 57 cm (50 cm to detector), temperature of 25 °C, 0.5 psi pressure injection for 4 s and UV detection at 214 nm.....	40
3.2 An example of electropherograms of the 10-day fermentation broth diluted 10 times using separation voltage of +30 kV, BGE concentrations of (a) 12.5, (b) 25.0, (c) 40.0 and (d) 50.0 mM. Peak 1 = GA ₃ , 2 = unknown and 3 = AMBA. Other CE conditions as shown in Figure 3.1.....	41

3.3	Effect of BGE ionic strength (I) on electroosmotic mobility (μ_{eo}) at separation voltage of +25 (\blacktriangle) and +30 kV (\blacksquare). Other CE conditions as shown in Figure 3.1.....	42
3.4	Effect of BGE ionic strength (I) on electrophoretic mobility (μ). Symbols: (\blacksquare) μ at +25 kV; (\blacktriangle) μ at +30 kV; (\square) μ^o at +25 kV; (\triangle) μ^o at +30 kV. Other CE conditions as shown in Figure 3.1.....	42
3.5	Effect of BGE ionic strength (I) on total mobility (μ_{net}) at separation voltage of +25 (\blacktriangle) and +30 kV (\blacksquare). Other CE conditions as shown in Figure 3.1	43
3.6	Effect of BGE ionic strength (I) on electric current (I_A) at separation voltage of +25 (\blacktriangle) and +30 kV (\blacksquare). Symbols \triangle and \square are the predicted values of I_A at separation voltage of +25 and +30 kV, respectively. Other CE conditions as shown in Figure 3.1	43
3.7	An example of electropherograms of 10-day fermentation broth diluted 10 times using (a) 20, (b) 40, (c) 50 and (d) 100 mM SDS, 25 mM $\text{Na}_2\text{B}_4\text{O}_7$ and separation voltage of +30 kV. Peak 1 = GA_3 , 2 = unknown, 3 = AMBA and 4 = unknown. Other CE conditions as shown in Figure 3.1.....	49
3.8	Effect of SDS concentration on electroosmotic mobility at separation voltage of +30 kV. Other CE conditions as shown in Figure 3.1.....	50
3.9	Effect of SDS concentration on electrophoretic mobility (μ) of GA_3 (\blacktriangle), unknown 2 (\blacksquare) and unknown 4 (\bullet) at separation voltage of +30 kV. Other CE conditions as shown in Figure 3.1.....	50
3.10	Effect of SDS concentration on total mobility (μ_{net}) of GA_3 (\blacktriangle), unknown 2 (\blacksquare) and unknown 4 (\bullet) at separation voltage of +30 kV. Other CE conditions as shown in Figure 3.1.....	51
3.11	An example of electropherograms of GA_3 dissolved in water at the period of time 0 hour (a) and 48 hours (b) and in 2.5 mM $\text{Na}_2\text{B}_4\text{O}_7$ at the period of time 0 hour (c) and 48 hours (d). CE conditions: uncoated fused silica capillary 50 μm I.D. \times 57 cm (50 cm to detector), 25 mM $\text{Na}_2\text{B}_4\text{O}_7$, 100 mM SDS, separation voltage of +30 kV, temperature of 25 $^\circ\text{C}$, 0.5 psi pressure injection for 4 s and UV detection at 214 nm.....	53
3.12	The amounts of GA_3 in water (\blacksquare) and a 2.5 mM $\text{Na}_2\text{B}_4\text{O}_7$ solution (\blacktriangle) as a function of the period of time	54
3.13	A plot of the A_{corr} ratio of GA_3 and AMBA ($A_{corr, ratio}$) as function of GA_3 concentration	55

3.14	An example of a chromatogram of 600 ppm GA ₃ standard solution containing 100 ppm AP	59
3.15	An example of HPLC chromatograms of GA ₃ extracted from the 10-day fermentation broth without (a) and with (b) spiking 250 ppm GA ₃ standard	60
3.16	Calibration plots between peak area of GA ₃ as a function of GA ₃ concentration in the range of 200 to 1200 ppm.....	61
3.17	An example of HPLC chromatograms of the 5 (a) and 10-day (b) fermentation broths.....	63
3.18	An example of MEKC electropherograms of the 5 (a) and 10-day (b) fermentation broths. CE conditions as shown in Figure 3.11	64
3.19	Relationship of the amounts of GA ₃ in the 3 to 11-day fermentation broths determined by MEKC and HPLC	65
3.20	An example of CZE electropherograms of the GA ₃ tablet (a) and GA ₃ tube (b). CE conditions: uncoated fused silica capillary 50 μm I.D. × 57 cm (50 cm to detector), 40 mM Na ₂ B ₄ O ₇ , separation voltage of +30 kV, temperature of 25 °C, 0.5 psi pressure injection for 4 s and UV detection at 214 nm.....	68
3.21	An example of MEKC electropherograms of the GA ₃ tablet (a) and GA ₃ tube (b). CE conditions as shown in Figure 3.11	68

LIST OF ABBREVIATIONS AND SYMBOLS

AMBA	3-amino-4-methylbenzoic acid
AP	3-acetamidophenol
BGE	background electrolyte
CE	capillary electrophoresis
CEC	capillary electrochromatography
CMC	critical micelle concentration
CZE	capillary zone electrophoresis
EMD	electromigration dispersion
EOF	electroosmotic flow
GA ₃	gibberellic acid
HPLC	high performance liquid chromatography
HPTLC	high performance thin layer chromatography
I.D.	internal diameter
MEKC	micellar electrokinetic chromatography
MS	mass spectrometry
RSD	relative standard deviation
SDS	sodium dodecylsulfate
<i>A</i>	peak area
<i>A</i> _{corr}	corrected peak area
<i>A</i> _{asym}	asymmetry factor
<i>c</i>	concentration of sample
<i>c</i> _{st}	concentration of analyte after stacking
<i>D</i>	diffusion coefficient
<i>D</i> _{th}	thermal dispersion coefficient
<i>E</i>	electric field strength
<i>E</i> _{opt}	optimum electric field
<i>e</i>	electronic charge
<i>F</i>	Faraday's constant
<i>f</i> _T	temperature factor
<i>I</i>	ionic strength
<i>I</i> _A	electric current

k	Boltzmann's constant
k'	retention factor
L	total capillary length
l	the length of capillary to detector
l_{inj}	length of analyte injected
l_{det}	detection width
l_{st}	sample zone length after stacking
N	the number of theoretical plate, or peak efficiency
n_{aq}	the amount of analyte in aqueous phase
n_{mc}	the amount of analyte in micellar phase
P_{EMD}	percentage of EMD
ΔP	pressure difference across the capillary
Q_{inj}	quantity of sample injected
R	gas constant
R_s	resolution
r_c	internal capillary radius
r_h	hydrodynamic radius
S	cross-sectional area of capillary
t_{eo}	migration time of EOF
t_{inj}	injection time
t_m	migration time of an analyte
t_{mc}	migration time of micelle
V	applied voltage
V_{inj}	volume of sample injected
v_{eo}	electroosmotic velocity
v_{ep}	electrophoretic velocity
v_{net}	total electrophoretic velocity
w_b	peak width at base
w_h	peak width at half height
x_{aq}	mole fraction of analyte in aqueous phase
x_{mc}	mole fraction of analyte in micellar phase
z	charge of an ion
α	separation factor

γ_{st}	a factor of sample stacking
η	viscosity
κ_c	electrical conductivity
λ_s	thermal conductivity
μ	electrophoretic mobility
μ^0	electrophoretic mobility at zero ionic strength
μ_{eo}	electroosmotic mobility
μ_{obs}	observed mobility
μ_{mc}	electrophoretic mobility of micelle
μ_{net}	total mobility
σ	standard deviation of peak in distance unit
σ^2	peak variance
σ_{EMD}^2	peak variance due to electromigration dispersion
σ_{det}^2	peak variance due to detection width
σ_{diff}^2	peak variance due to longitudinal diffusion
σ_{inj}^2	peak variance due to injection length
σ_{tc}^2	peak variance due to time constant
σ_{th}^2	peak variance due to thermal dispersion
σ_{tot}^2	total peak variance
τ	standard deviation of peak in time unit
τ_{tc}	time constant
ζ	zeta potential