

國立屏東科技大學熱帶農業暨國際合作系  
Department of Tropical Agriculture and International Cooperation  
National Pingtung University of Science and Technology

博士學位論文  
Ph.D. Dissertation

草藥 (龍牙草、鱧腸、仙草、地耳草、甜珠草及曲序香茅) 之栽培及生物活性研究

Study in Cultivation and Bioactivities of Herbs *Agrimonia pilosa*,  
*Eclipta alba*, *Mesona procumbens*, *Hypericum japonicum*, *Scoparia  
dulcis*, and *Cymbopogon flexuosus*

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中華民國 108 年 6 月 15 日  
June 15, 2019

## 摘要

學號：P10422023

論文題目：草藥(龍牙草、鱧腸、仙草、地耳草、甜珠草及曲序香茅)之栽培及生物活性研究

總頁數：138 頁

學校名稱：國立屏東科技大學 系(所)別：熱帶農業暨國際合作系

畢業時間及摘要別：107 學年度第 2 學期博士學位論文摘要

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論文摘要內容：

本次研究中選擇六種受關注的傳統中草藥應用於天然產品。該研究選用五項體外實驗進行研究。第一項目的為探討蚯蚓糞(VC)和豬糞肥對龍牙草 (*Agrimonia pilosa* Ledeb.) 生長、產量和化學成分含量的影響。其他四項研究目的為研究龍牙草 (*Agrimonia pilosa* Ledeb.) 提取物的抗氧化活性以及對 HepG2 (人類肝癌細胞) 和 A549 (人類肺癌細胞) 的生長抑制效果。鱧腸 (*Eclipta alba* Hassk.)、仙草 (*Mesona procumbens* Hemsl.)、地耳草 (*Hypericum japonicum* Thunb.)、甜珠草 (*Scoparia dulcis* L.) 和曲序香茅 (*Cymbopogon flexuosus* (Nees ex Steud.) W. Watson) 以乙醇萃取後進行實驗。實驗項目為總酚和類黃酮含量以及包含總抗氧化能力 (ABTS)、清除自由基能力 (DPPH) 以及羥自由基清除能力等抗氧化實驗。在某些實驗中還評估了培養基中的乳酸脫氫酶含量。並利用高效液相色譜 (HPLC) 方法用於分析龍牙草 (*Agrimonia pilosa* Ledeb.)、鱧腸 (*Eclipta alba* Hassk.)、仙草 (*Mesona procumbens* Hemsl.)、地耳草 (*Hypericum japonicum* Thunb.) 及甜珠草 (*Scoparia dulcis* L.) 中的酚類化合物。另外透過質譜儀 (MS) 和核磁共振 (NMR) 建立曲序香茅 (*Cymbopogon flexuosus* (Nees ex Steud.) W. Watson) 的化學結構和地上部

分的外部特徵。並透過西方墨點法分析凋亡因子 bcl-2、bax、bad、caspase-3、caspase-9 和 p53 蛋白。

結果表明、對於第一項研究（1）、施用有機肥料增強了龍牙草（*Agrimonia pilosa* Ledeb.）的生長、產量和化學成分含量。pH 值在 6.9 和 7.4 之間、導電率(EC)從 0.5-0.6mS cm<sup>-1</sup> 為此種草藥生長的最佳條件。蚯蚓糞(VC)和豬糞的最佳比例為 12.5 + 16.875 ton ha<sup>-1</sup> 的比例製成。這一結果表明、使用有機肥料可以提升其他草藥的生長、產量和化學成分含量、並進行進一步的詳細調查。在第二項研究（2）中、在龍牙草（*Agrimonia pilosa* Ledeb.）萃取物中鑑定出由 4-羥基苯甲酸和對香豆酸組成的兩種單獨的酚類化合物。與地上部分萃取物相比、根部萃取物有更高的抗氧化力及 A549 細胞生長抑制能力。在第三項研究（3）中、鱧腸（*Eclipta alba* Hassk.）萃取物經鑑定過後得知含有 4-羥基苯甲酸、咖啡酸、對香豆酸和迷迭香酸等化合物。仙草(*Mesona procumbens* Hemsl.) 萃取物經鑑定過後得知含有 7-羥基香豆素、阿魏酸和芸香苷是新化合物和兩種已知化合物。從鱧腸（*Eclipta alba* Hassk.）萃取物中檢測到較高清除自由基能力(DPPH)和總抗氧化能力(ABTS)及抗癌活性。在第四項研究（4）中、綠原酸和迷迭香酸是地耳草（*Hypericum japonicum* Thunb.）萃取物經鑑定後所得知的化合物。芸香苷和迷迭香酸是從甜珠草(*Scoparia dulcis* L.) 萃取物中新測得的化合物。地耳草（*Hypericum japonicum* Thunb.）萃取物有更高水平的抗氧化和抗癌能力。最後的研究（5）、1,3-O-二-E-咖啡酰甘油（SA3）和 1-O-p-香豆酰-3-O-咖啡酰甘油（SA4）兩者為從曲序香茅(*Cymbopogon flexuosus* (Nees ex Steud.) W. Watson) 萃取物中分離出來化合物。SA3 化合物擁有最高的抗氧化力和抗癌能力。

根據本次實驗結果顯示有些中草藥可能有助於為癌症疾病患者開發新型天然藥物、還可以進一步了解草藥的功能和分子機制。製作龍牙草（*Agrimonia pilosa* Ledeb.）、鱧腸（*Eclipta alba* Hassk.）、地耳草（*Hypericum japonicum* Thunb.）、曲序香茅(*Cymbopogon flexuosus* (Nees ex Steud.) W. Watson) 的機能性食品或營養補充品的膠囊。

**關鍵字：** 抗癌、龍牙草、曲序香茅、鱧腸、地耳草

## Abstract

Student ID: P10422023

Title of Dissertation: Study in Cultivation and Bioactivities of Herbs *Agrimonia pilosa*, *Eclipta alba*, *Mesona procumbens*, *Hypericum japonicum*, *Scoparia dulcis*, and *Cymbopogon flexuosus*

Total Page: 138 pages

Name of Institute: Department of Tropical Agricultural and International Cooperation, National Pingtung University of Science and Technology

Graduate Date: June 15, 2019

Degree Conferred: Doctoral Degree

Name of Student: Le Quang Ung

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The content of abstract in this dissertation:

Six herbs chosen in this work have received considerable attention as natural products for their applications in traditional medicine. The study consists of five studies being conducted under *in vitro* conditions. The first study aims to investigate effects of vermicompost and hog manure on growth, yield, chemical composition of *Agrimonia pilosa* Ledeb. Four other studies aim to investigate antioxidant activities, HepG2 (human liver cancer cell line) and A549 (adenocarcinomic human alveolar basal epithelial cell line) growth inhibitory effect from extracts of *Agrimonia pilosa* Ledeb. *Eclipta alba* Hassk. *Mesona procumbens* Hemsl. *Hypericum japonicum* Thunb. *Scoparia dulcis* L. and *Cymbopogon flexuosus* (Nees ex Steud.) W. Watson. Extracts of ethanol were chosen for the assays. Total phenolic and flavonoid content and the antioxidant activities comprising ABTS<sup>+</sup>, DPPH, hydroxyl radical

scavenging systems were determined. Lactate dehydrogenase release in medium was also evaluated in some experiments. High performance liquid chromatography (HPLC) method was used to analyze phenolic compounds from *Agrimonia pilosa* Ledeb. *Eclipta alba* Hassk. *Mesona procumbens* Hemsl. *Hypericum japonicum* Thunb. *Scoparia dulcis* L. Chemical structural characterization from the aerial parts of *Cymbopogon flexuosus* was established by MS and NMR spectra techniques. The apoptotic factors bcl-2, bax, bad, caspase-3 and caspase-9, p53 were analyzed by western blotting assays.

The results showed that, for the first study (1), application of organic fertilizers enhanced the morphological growth, yield, chemical composition of *Agrimonia pilosa* Ledeb. The pH between 6.9 and 7.4, EC from 0.5-0.6 mS cm<sup>-1</sup> are optimal for this herbal growth. A mixture of the Vermicompost and Hog manure made with the rate of 12.5 + 16.875 ton ha<sup>-1</sup> produced best parameters. This result suggests that using of organic fertilizers could completely enhance the morphological growth, yield, chemical composition of other herbs and further detailed investigations are in progress. In the second study (2), two individual phenolic compounds consisting of 4-hydroxybenzoic acid and p-coumaric acid were firstly identified from *Agrimonia pilosa* Ledeb. The root extract exhibited higher antioxidant and A549 inhibitory capacity compared to the aerial part extract. In the third study (3), the 4-hydroxybenzoic acid, caffeic acid, p-coumaric acid and rosmarinic acid were the identified compounds in *Eclipta alba* Hassk. The 7-hydroxycoumarin, ferulic acid and rutin being new compounds and two known compounds were identified in the *Mesona procumbens* Hemsl. The higher levels of DPPH and ABTS<sup>+</sup> radical scavenging and anticancer activities were detected from *Eclipta alba* Hassk. In the fourth study (4), the chlorogenic acid and rosmarinic acid were the identified compounds in the *Hypericum japonicum* Thunb. The rutin and rosmarinic acid were newly identified compound from the *Scoparia dulcis* L. The higher levels of antioxidant and anticancer

capacity were detected from *Hypericum japonicum* Thunb. The last study (5), 1,3-O-di-E-caffeoylglycerol (SA3) and 1-O-p-coumaroyl-3-O-caffeoylglycerol (SA4) were firstly isolated from *Cymbopogon flexuosus* (Nees ex Steud.) W. Watson. The SA3 compound showed the highest antioxidant and anticancer potent.

The information obtained from this work will likely contribute to the development of novel natural medicine for cancer diseases. It will also provide new insights for the futher understanding of the functions and molecular mechanism of herbs. An insightful investigation to establish and manufacture capsules used as function and supplemental food from *Agrimonia pilosa* Ledeb. *Eclipta alba* Hassk. *Hepericum japonicum* Thunb. *Cymbopogon flexuosus* (Nees ex Steud.) W. Watson was recommended.

**Keywords:** anticancer, *Agrimonia pilosa* Ledeb, *Cymbopogon flexuosus* (Nees ex Steud.) W. Watson, *Eclipta alba* Hassk, *Hypericum japonicum* Thunb

## Acknowledgements

I would like to express sincere thanks to my two advisors, Professor Dr. Ming-Chang Wu and Professor Dr. Horng-Liang Lay, for their invaluable guidance on my research. I greatly appreciate their intellectual instructions, kindness, and supports during my Ph.D. research program.

I would like to thank the advisory committee members for their valuable comments and constructive suggestions for the successful completion of this research work.

I am grateful to Taiwan Scholarship (Ministry of Education) for budget support for my Ph.D. program in Taiwan. I would also like to thank NPUST, DTAIC for offering me this good opportunity to pursue my doctoral study.

I would like to thank the personnel of the Department of Tropical Agriculture and International Cooperation; Office of International Affairs, Chinese Herbal Medicine and FP201 Laboratory for their help during this study program.

Lastly, I would like to thank my parents for their confidence and love; my colleague, my friends for their encouragement and affection and especially my wife who was always there with wisdom, inspiration and cheer.

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