

**ALLELOPATHIC TANNINS FROM GUYABANO (*Annona muricata* L.) LEAF
EXTRACT AGAINST THE RICE WEED *Echinochloa crus-galli***

ANDREI PHILSEN NUQUE ESPIRITU



An Undergraduate Thesis Submitted to the Faculty of the Department of Chemistry,
College of Arts and Sciences, Central Luzon State University,
Science City of Munoz, Nueva Ecija, Philippines
in Partial Fulfilment of the requirements
for the degree of

BACHELOR OF SCIENCE IN CHEMISTRY

June 2017

CHE-02-17-007

APPROVAL AND ACCEPTANCE

The undergraduate thesis entitled **ALLELOPATHIC TANNINS FROM GUYABANO (*Annona muricata* L.) LEAF EXTRACT AGAINST THE RICE WEED *Echinochloa crus-galli*** prepared and submitted by **ANDREI PHILSEN N. ESPIRITU** in partial fulfillment of the requirements for the degree of **BACHELOR OF SCIENCE IN CHEMISTRY** is hereby

APPROVED:


ROSALIE R. RAFAEL, Ph.D.
Adviser

06/12/17
Date


MARY JANE T. DELA CRUZ, M.Sc.
Critic

06/12/17
Date

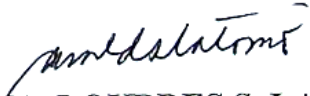

REDINE L. GUTIERREZ, Ph.D.
Department Research Coordinator

06/13/17
Date

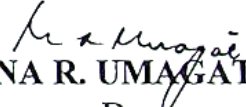

DANILO S. PARAGAS, M.Sc.
Department Chair

6/13/2017
Date

AND ACCEPTED:


ANNA MA. LOURDES S. LATONIO, Ph.D.
College Research Coordinator

06/13/17
Date


MYRNA R. UMAGAT, Ph. D.
Dean

06/13/17
Date

BIBLIOGRAPHICAL SKETCH

PERSONAL INFORMATION

Name : **ANDREI PHILSEN N. ESPIRITU**
Date of Birth : March 02, 1996
Place of Birth : Bantug, Science City of Munoz, Nueva Ecija

EDUCATIONAL BACKGROUND

Elementary : **ACCELERATED CHRISTIAN INTL SCHOOL**
Villa Antonia Homes, Science City of Munoz,
Nueva Ecija
March 2008

High School : **CLSU-SCIENCE HIGH SCHOOL**
CLSU, Science City of Munoz, Nueva Ecija
March 2012

College : **CENTRAL LUZON STATE UNIVERSITY**
Science City of Munoz, Nueva Ecija
Bachelor of Science in Chemistry
June 2017

SCHOLARSHIP AND AWARDS

Dean's Lister : 1st Semester 2012-2013

EXTRACURRICULAR ACTIVITIES

Vice President-External : CLSU Chemical Society (A.Y. 2015-2016)
Member : CLSU Chemical Society (A.Y. 2013-present)
Member : Philippine Association of Chemistry Students, Inc.
(2013-present)

Member : SAVE-NE (2012-2014)

Participant : PACSiklaban
University of the Philippines-Diliman
February 2017

SEMINARS ATTENDED

Participant : Shimadzu Technology for Analytical
Instrumentation
CAS Little Theater, CLSU
October 8, 2015

Participant : PhilRice Nutrition Month Seminar
PhilRice Central Experiment Station
July 30, 2013

Participant : Chemistry is Fun
Chemistry Symposium Seminar
University Auditorium, CLSU
February 19, 2013

Participant : Breathless: Smoking and Its Impact on One's Health
University Auditorium, CLSU
September 14, 2012

ACKNOWLEDGEMENT

I will take this opportunity to thank all the people who helped me conduct and finish this study.

First, to the Almighty God, who gave me strength, wisdom and blessings throughout my life. If not for Him, I will not be here today and finishing my degree. I thank Him for everything He had done and will do throughout my life.

I also give my deepest gratitude to the following people who made this study possible.

To my BONP family, Kuya Eddyson and Ate Kate for assisting us in using the laboratory equipments, to Ate Chai for the words of wisdom and for the food and especially to Ate Sarah-p, thank you very much for guiding us all throughout the study, for sponsoring our little needs like coffee and noodles during the overnights and for being a friend in food and in need. Thank you.

To my adviser, Dr. Rosalie R. Rafael, thank you for the guidance, encouragement and help you had given me. Thank you for pushing me to do my best just to make this study better.

To my critic, Associate Professor Mary Jane T. Dela Cruz, thank you Maam for improving my work through constructive criticism. Thank you for encouraging and believing in me that I can do this study.

To Dr. Redel L.Gutierrez, Department Research Coordinator and the adviser of CLSU Chemical Society, thank you Sir for your patience you gave me. I would also like

to thank you for being a mentor, brother and a friend during my days as a member of the CLSU Chemso.

To Dr. Myrna R. Umagat, Dean of College of Arts and Sciences, and Ma'am Rhanney L. Gonzales, thank you for always pushing me to be better and for believing that I can do it. Thank you for reminding me to be good at times when I get a bit naughty and ignorant of the works I needed to accomplish.

To Prof. Danila S. Paragas, Department Chair, thank you Ma'am for the support and the help in making this study a better one. Thank you Ma'am for the advices.

To the canteen people, Ate Nora and Ate Rose, for the free food you have given me when I am so broke and I have nothing to spare.

To my classmates, thank you for the rollercoaster ride college days and for the never ending support and help. Thank you for making my college days a great one. I love you guys.

To my TEAM SEPANX, Jasmin and Ate Sarah-p, thank you for being there every time. Thank you for making me the greatest 5th wheel of all time! Thank you for the companionship and the love. To more travel and food goals my friends!

To my DBX family, my high school friends, thank you for supporting me, for pushing me to finish my works and for being by my side whenever I need help especially to Hector for helping me find my rice weeds. Yo da best bru.

To my Brods and Sisses in CLSU Chemical Society, thank you for everything. Thank you for treating me as your Kuya for years. I know I haven't taught you good things but I hope the good deeds I have done to you and to other people will stay in your hearts forever.

To my family, Mama, Papa, Ayra, Aeron and Aevann, this is it. The longest wait is over! Thank you for your support and for the patience. Sorry if it really took so long. As I always say, quality education takes time! To my Mama Remy, Ate Joy and Ate Kim, thank you for the support especially financially. To Mommy, Daddy, Cyril, Cedie and CJ, thank you also for your support and love and for also letting me use your laptop in doing my manuscript.

“ANG PAG-AARAL AY PARANG PAG-AASAWA, HINDI MINAMADALI.”

ANDREI PHILSEN N. ESPIRITU

TABLE OF CONTENTS

	Page
TITLE PAGE	i
APPROVAL AND ACCEPTANCE	ii
BIBLIOGRAPHICAL SKETCH	iii
ACKNOWLEDGEMENT	v
LIST OF TABLES	x
LIST OF FIGURES	xi
LIST OF APPENDICES	xii
LIST OF APPENDIX FIGURES	xiii
LIST OF APPENDIX TABLES	xiv
ABSTRACT	1
1 INTRODUCTION	2
1.1 Background of the Study	2
1.2 Significance of the Study	3
1.3 Objectives of the Study	4
1.4 Time and Place of the Study	5
1.5 Scope and Limitations of the Study	5
2 REVIEW OF RELATED LITERATURE	
2.1 Botanical description of <i>Annona muricata</i> L.	6
2.2 Studies on <i>Annona muricata</i> L.	7
2.3 Allelopathy	8
2.4 Tannin	9
2.5 <i>Echinochloa crus-galli</i>	10
3 METHODOLOGY	
3.1 Materials, Chemicals, and Reagents	12
3.2 Sample Collection and Preparation	12
3.3 Preparation of the Plant Extract	12
3.4 Chemical tests for Tannin	13
3.4.1 Test with Ferric chloride solution	13

3.4.2	Test with gelatin solution	13
3.4.3	Thin layer chromatography	13
3.4.3.1	Preparation of the Developing Chamber	13
3.4.3.2	Development of the Chromatogram	13
3.4.3.3	Visualization and Documentation of Chromatogram	14
3.5	Extraction of tannin	14
3.6	Characterization of tannins	14
3.6.1	UV-Vis spectrophotometry of tannins from <i>Annona muricata</i> L.	15
3.6.2	IR spectrophotometry of tannins and related phenolics	15
3.7	Determination of Total Tannins	15
3.8	Bioassay of the extract	15
3.8.1	Test for seed germination	16
3.8.2	Test for seed growth	16
3.9	Statistical Analysis	16
4	RESULTS AND DISCUSSION	
4.1	Crude extract of <i>Annona muricata</i> L.	17
4.2	Test for tannin of crude extract and fractions of <i>Annona muricata</i> L.	17
4.3	Total tannin content of <i>Annona muricata</i> L. Leaves	19
4.4	Weedicide bioassay of tannin from <i>Annona muricata</i> L.	19
4.5	UV-Vis spectrophotometry of tannins from <i>Annona muricata</i> L.	22
4.6	IR spectrophotometry of tannins	23
5	SUMMARY, CONCLUSION AND RECOMMENDATION	
5.1	Summary	25
5.2	Conclusion	26
5.3	Recommendation	27
	LITERATURE CITED	28
	APPENDICES	33

LIST OF TABLES

Table No.	Title	Page
4.1	Effect of tannin from <i>Annona muricata</i> L. on the seedling growth of <i>Echinochloa crus-galli</i> seed at different concentrations	21
4.2	Infrared positions of tannin from <i>Annona muricata</i> L.	24

LIST OF FIGURES

Figure No.	Title	Page
2.1	Images of <i>Annona muricata</i> L.	6
2.2	Chemical structure of tannin	10
2.3	Image of <i>Echinochloa crus-galli</i>	11
4.1	Crude extract of <i>Annona muricata</i> L.	17
4.2	Test for tannins on the leaf extract of <i>Annona muricata</i> L. (a) gelatin test (b) FeCl ₃ test and (c) leaf extract	17
4.3	The thin layer chromatogram of fraction 1 showing (A) gallic acid spot (B) tannin spot	17
4.4	The effect of different concentrations of the tannin fraction of <i>Annona muricata</i> L. on the seed germination of <i>Echinochloa crus-galli</i>	20
4.5	UV-Vis spectrometry of tannin extract of <i>Annona muricata</i> L.	22
4.6	Infrared spectroscopy of tannins from <i>Annona muricata</i> L.	24

LIST OF APPENDICES

Appendix No.	Title	Page
1	Seed Bioassay	34
2	Experimental Results	36

LIST OF APPENDIX FIGURES

Appendix Figure No.	Title	Page
1.1	Percent germination of <i>Echinochloa crus-galli</i> seeds (Trial 1)	34
1.2	Percent germination of <i>Echinochloa crus-galli</i> seeds (Trial 2)	34
1.3	Percent germination of <i>Echinochloa crus-galli</i> seeds (Trial 3)	34
1.4	Seed growth of <i>Echinochloa crus-galli</i> shoots in different concentrations of tannin (Trial 1)	35
1.5	Seed growth of <i>Echinochloa crus-galli</i> shoots in different concentrations of tannin (Trial 2)	35
1.6	Seed growth of <i>Echinochloa crus-galli</i> shoots in different concentrations of tannin (Trial 3)	35
1.7	Calibration curve of the standard gallic acid	35

LIST OF APPENDIX TABLE

Appendix Table No.	Title	Page
2.1	Initial and final length (mm) of the <i>Echinochloa crus-galli</i> seeds treated with different concentration of tannins	37
2.2	Results of Duncan Multiple Range Test on the seedling growth of <i>Echinochloa crus-galli</i> seeds	37
2.3	ANOVA for the effect of tannin fractions on shoot lengths	38

ALLELOPATHIC TANNINS FROM GUYABANO (*Annona muricata* L.) LEAF EXTRACT AGAINST THE RICE WEED *Echinochloa crus-galli*¹

ANDREI PHILSEN N. ESPIRITU

ABSTRACT

Background: Allelopathy is a biological phenomenon where one plant inhibits the growth of another. The application of the allelopathic properties of some plants is a natural and environment-friendly approach which increases crop yields, decreases our dependence on synthetic herbicides, and improves the ecological environment. This study aimed to extract the tannins from *Annona muricata* L. leaves and determine the allelopathic effect on the germination and seedling growth of the rice weed, *Echinochloa crus-galli*. **Methods:** The dried powdered leaves of *Annona muricata* L. were extracted using 80% ethanol. Tannins were extracted with 50% acetone using column chromatography. Phytochemical analyses (FeCl₃ test, gelatin test and TLC) on the crude extract and fractions were done to confirm the presence of tannins. The total tannin content on the leaves of *Annona muricata* L. was also determined. The tannin fraction was characterized by UV-Vis and IR spectrophotometry. The allelopathic effect of the tannin fraction of *Annona muricata* L. on the seed germination and seedling growth of *Echinochloa crus-galli* rice weed was also noted. **Results:** The total tannin content of the fraction was 25.33±00 mg GAE g⁻¹. The tannin present was probably and ellagitannin based from the UV-Vis and FTIR Spectroscopy. The percent germination was 66.67±5.77% and 0 in all treatments. Highest inhibitory effect on the seedling growth was observed in the 8 and 10 mg/mL fractions. However, no significant differences on the tannin fractions were observed but showed significant difference from the control. **Conclusion:** The probable ellagitannin from guyabano leaves inhibits seed germination and seedling growth of the rice weed, *Echinochloa crus-galli* and has the potential as a weedicide.

Keywords: tannin, *Annona muricata* L., allelopathy, *Echinochloa crus-galli* rice weed

¹ An undergraduate thesis presented to the Faculty of the Department of Chemistry, College of Arts and Sciences, Central Luzon State University, Science City of Muñoz, Nueva Ecija in partial fulfillment of the requirements for the degree of Bachelor of Science in Chemistry. Prepared under the supervision of Professor Dr. Rosalie R. Rafael and Associate Prof. Mary Jane T. Dela Cruz. June 2017. CHE-02-17-007

LITERATURE CITED

- Abiodun F, Osakue J, Stephen UA, Sheng-Xiang Q. (2011). Phytochemical and anti-cancer studies on then medicinal plants used in Nigeria. *Baero Journal of Pure and Applied Sciences* 4(1): pp. 36-39
- Ahn JK, Hanh SJ, Kim JT, Khan TD, Chung IM. (2005). Evaluation of allelopathic potential amongrice (*Oryza sativa* L.) germplasm for control of *Echinochloa crus-galli . beauv* in the field. *Crop Protection*Vol 24(5): pp.413-419
- Arowosgabe S, Wintola OA, Afolayan AJ. (2012). Phytochemical constituents and allelopathic effect of *Aloe ferox* Mill. Root extract on tomato. *Journal of Medicinal Plants Research* Vol. 6 (11): pp. 2094-2099.
- Bate-Smith EC, Swein T. Flavonoid compounds. (1962). *Comparative Biochemistry* (H.S. Mason, A.M. Florkin eds.), Academic Press, New York: pp. 755–809.
- Betancur-Galvis J, Saez H, Granados, Salazar A, Ossa JE. (1999). Antitumor and antiviral activity of Colombian medicinal plant extracts: pp. 531–535
- Blum U. (1995). The value of model plantmicrobe-soil systems for understanding processes associated with allelopathic interactions (Inderjit, K. M. M. Dakshini, and F. A. Einhellig, eds.). *Allelopathy: Organisms, processes, and applications*. ACS Symposium Series No. 582. Washington D.C.: American Chemical Society: pp. 127-131
- Bray EA (1993). "Molecular Responses to Water Deficit". *Plant Physiol.* **103** (4): 1035–1040. doi:10.1104: pp.103.4.1035
- Callaway RM. (2002). The detection of neighbors by plants. *Trends Ecological Evolution.* 17: pp. 104–105.
- Chalker-Scott L, Krahmer RL. (1989) Microscopic studies of tannin formation and distribution in plant tissues. *In Chemistry and Significance of Condensed Tannins*. Eds. R W Hemingway and J J Karchesy: pp. 345–368. Plenum Press, New York
- Chung KT, Wei CI, Johnson MG. (1998). Are tannins a double- -edged sword in biology and health? *Trends Food Sci. Nutrition*Vol 9: pp 168–175.
- Chung, I.M., Miller, D.A.(1995), Natural herbicide potential of alfalfa residues on selected weed species. *Agronomy Journal*87: pp. 920–925.
- Dagar JC, Dagar. (1991). Plant folk medicines among the Nicobarese of Katchal Island, India. *Econ. Bot.*, 45: pp. 114–119

- Duke SO, Dayan FE, Romagni JG, Rimando AM. (2000). Natural products as sources of herbicides: current status and future trends. *Weed Research* 40: 99–111
- EBB Greenhouse Staff, University of Connecticut. (2008). *Annona muricata* L. *Ecology & Evolutionary Biology Greenhouse*.
- Enstone DE, Peterson CA, Hallgren SW. (2001). Anatomy of seedling tap roots of loblolly pine (*Pinus taeda* L.). *Trees* Vol 15: pp. 98–111.
- Feucht W, Treutter D. (1999). The role of flavan-3-ols and proanthocyanidins in plant defense. In *Principles and Practices in Plant Ecology – Allochemical Interactions*. (Inderjit, K M M Dakshini and C L Foy eds.): pp. 307–338. CRC Press, Boca Raton.
- Ferreria LE, Castro PMN, Chagas ACS, Franca SC, Beleboni RO. (2012). In vitro anthelmintic activity of aqueous leaf extract of *Annona muricata* L. (Annonaceae) against *Haemonchus contortus* from sheep. *Experimental Parasitology* Vol 134 (3) 2013: pp. 327-332
- Foong C. P. and Hamid R. A. (2012). Evaluation of anti-inflammatory activities of ethanolic extract of *Annona muricata* leaves. *Rev. bras. farmacogn.* 22:6
- Galakshimi S, Vijayalakshimi S, Rajeswari DV. (2012). Phytochemical and Pharmacological Properties of *Annona muricata*: A Review. *International Journal of Pharmacy and Pharmaceutical Sciences* Vol 4 (2) 2012.
- Grard P, Homsombath K, Kessler P, Khuon E, Le Bourgeois T, Prosperi J, Risdale C. 2006. OSWALD V. 1.0. A Multimedia identification system of the major weeds of rice paddy fields of Cambodia and Lao P.D.R. CIRAD.Montpellier, France.
- Grundhöfer P, Niemetz R, Schilling B and Gross GG. (2001). Biosynthesis and subcellular distribution of hydrolyzable tannins. *Phytochemistry* 57: pp. 915–927.
- Hanson JR. (2003). Natural products: the secondary metabolites. Cambridge: *Royal Society of Chemistry*. ISBN0-85404-490-6
- Horvath G, Molnar P, Bencsik, T. 2014. Pharmacognosy 2. TAMPO-4.1.2 A1.
- Hunter P. (2008). Harnessing Nature's wisdom. Turning to Nature for inspiration and avoiding her follies. *EMBO Report*.9 (9): pp. 838–40.
- IAS (1996) International Allelopathy Society (IAS). Constitution and Bylaws. In: Proceedings First World Congress on Allelopathy: A Science for the Future, Ca'diz, Spain.

- Inderjit. (1996). Plant phenolics in allelopathy. *Botanical Review Vol 62*: pp. 186-202.
- Inderjit, Keating K. (1999) Allelopathy: principles, procedures, processes, and promises for biological control. *Advances in Agronomy 67*: pp. 141–232.
- Jung WS, Kim KH, Ahn JK, Hanh SJ, Chung IM. (2004). *Crop Protection Vol 23 (3)* (2004): pp. 211-218
- Khanh T, Chung M, Xuan T, Tawata S (2005). The Exploitation of Crop Allelopathy in Sustainable Agricultural Production. *Agronomy & Crop Science 191*: pp. 172—184.
- Kraus TEC. (2002) Tannins and nutrient dynamics in forest soils: plant-litter-soil interactions. PhD thesis, University of California, Davis, CA. pp. 182
- Kumpulainen JT, Salonen, JT. (1999). Natural Antioxidants and Anticarcinogens in Nutrition, Health and Disease. *The Royal Society of Chemistry, UK*: pp. 178-187.
- Isman LMB.(2004). Insecticidal activity of crude seed extracts of *Annona* spp., *Lansium domesticum* and *Sandoricum koetjape* against Lepidopteran Larvae. *Phytoparasitica*, 32: pp. 30–37
- Lewis NG & Yamamoto E.(1989). Tannins – their place in plant metabolism. In *Chemistry and Significance of Condensed Tannins*. (R W Hemingway and J J Karchesy eds.): pp. 23–46. Plenum Press, New York.
- Li HH, Inoue M, Nishimura H, Mizutani J, Tsuzuki E. (1993). *Journal of Chemical Ecology* (1993) 19: 1775. Doi:10.1007/BF00982307
- Luna JDS, de Carvalho JM, de Lima MRF, Beiber LW, Bento EDS, Franck X, AcetSant'ana AEG. (2004). Acetogenins in *Annona muricata* L. leaves are potent molluscicides. *Natural Product Research Vol 20 (3)* 2006: pp. 253-257
- Macias F. A., Marin D., Oliveros-Bastidas A., Varela R. M., Simonet A. M., Carrera C., et al. (2003). Allelopathy as a new strategy for sustainable ecosystems development. *Biol. Sci. Space* 17, 18–23. 10.2187/bss.17.18
- Magaña LM, Gama R, Mariaca E. (2010). El uso de las plantas medicinales en las comunidades mayachonales de nacajuca, Tabasco, México. *Polibotanica*, 29: pp. 213–262
- Maron J, Vilà M. (2001). When do herbivores affect plant invasion? Evidence from the natural enemies and biotic resistance hypotheses. *Oikos* 95: pp. 361–373.

- Maplestone RA, Stone MJ, Williams DH. (1992). The evolutionary role of secondary metabolites--a review, *Gene* 115 (1-2):pp. 151-7.
- Mbagwu FN. (2006). The allelopathic effects of crude water extracts of *Annona muricata* L. on common weeds. *International Journal of Natural and Applied Sciences*. Vol. 2 (4) 2006: pp. 341-344
- Mersie, W. & Singh, M. (1988): Effect of phenolic acids and ragweed *Parthenium* (*Parthenium hysterophorus*) extract on tomato (*Lycopersicon esculentum*) growth and nutrients and chlorophyll content. *Weed Science*, 36:pp. 278-281
- Mersie, W, Singh, M. (1987): Allelopathic effect of *Parthenium hysterophorus* extract and residue on some agronomic crop and weeds. *Journal of Chemical Ecology*, 13: pp.1739-1747
- Moghandamtousi SZ, Fadaeinasab M, Nikzad S, Mohan G, Mohd Ali H. (2015). *Annona muricata* (Annonaceae): A Review of Its Traditional Uses, Isolated Acetogenins and Biological Activities. *International Journal Mol. Science Vol 16 (7)*: pp. 15625-15628.
- Muller CH (1969). Allelopathy as a factor in ecological process. *Vegetation* 18, 348-357.
- Musco A, Pauccio MR, Sidari M. (2001): The effects of phenolic acids extracted from two different soils in Aspromonte on germination of *Pinus laricio* seeds. *Fresenius Environmental Bulletin*, 10:pp. 659-663
- Olofsdotter, M. (1998): Allelopathic in rice; Proceeding of the workshop on allelopathy in rice. Manila, Phillippines, International Rice Research Institute: pp. 1 - 5
- Okuda T. (1999) Novel aspects of tannins – renewed concepts and structure-activity relationships. *Cur. Org. Chem.* 3:pp. 609-622.
- Ossipov V, Salminen J-P, Ossipova V, Haukioja E and Pihlaja K 2003 Gallic acid and hydrolyzable tannins are formed in birch leaves from an intermediate compound of the shikimate pathway. *Biochem. Sytem. Ecol.* 31:pp. 3-16.
- Peters DJ, Constabel CP. (2002). Molecular analysis of herbivore induced condensed tannin synthesis: cloning and expression of dihydroflavonol reductase from trembling aspen (*Populus tremuloides*). *Plant J.* 32:pp.701-712.
- Peterson CA, Enstone DE, Taylor JH. (1999). Pine root structure and its potential significance for root function. *Plant Soil* 217. :pp. 205-213.
- Putnam A.R. (1988). Allelochemicals from plants as herbicides. *Weed Technology* 2: pp. 510-518.
- Racon L, Sadaka N, Gil G, Le Petit J, Matheron R, Poinsot-Balaguer N, Sigoillot JC, Woltz P. (1988). Histological and chemical 65 changes in tannic compounds of evergreen oak leaf litter. *Can. J. Bot.* 66: pp. 663-667.

- Samuelson G. (1999). *Drugs of Natural Origin: A Textbook of Pharmacognosy*. Taylor & Francis Ltd ISBN 9789186274818.
- Seigler DS. (1996) Chemistry and mechanisms of allelopathic interactions. *Agronomy Journal* 88: pp. 876–885.
- Singh HP, Batish DR, Kohli RK. (2010). Allelopathic Interactions and Allelochemicals: New Possibilities for Sustainable Weed Management. *Critical Reviews and Plant Sciences Journal*, 22 (2010): pp. 239-311
- Tambe VD, Bhambar RS. 2014. Estimation of Total Phenol, Tannin, Alkaloid and Flavanoid in *Hibiscus tillaceus* Linn. Wood extracts. *Journal of Pharmacognosy and Phytochemistry Vol 2* (4).
- Tsai FY, Lin CC, Kao CH. (January 1997). A comparative study of the effects of abscisic acid and methyl jasmonate on seedling growth of rice. *Plant Growth Regulation*. 21 (1): pp. 37–42. doi:10.1023/A:1005761804191
- De Sousa OV, Vieira GDV, De Pinho JDJ, Yamamoto CH, Alves MS. (2010). Antinoceptive and anti-inflammatory Activities of *Annona muricata* L. leaves in Animal models. *International Journal of Molecular Science*, 11(5), 2067-2078; doi:10.3390/ijms11052067
- Sary F, Hans S. (1998). *The National guides to medical herbs and plants*. Tiger Books. Int. Plc. UK
- Vijayameena CG, Subhashini M, Loganayagi A, Ramesh B. (2013). Phytochemical screening and assessment of antibacterial activity for the bioactive compounds in *Annona muricata*. *International Journal Curr. Microbiol. App. Sci.* 2(1): pp. 1-8
- Waller GR, Feng MC, Fujii Y. (1999). Biochemical analysis of allelopathic compounds: plants, microorganisms, and soil secondary metabolites. In: *Principles and Practices in Plant Ecology* (Inderjit, KMM Dakshini & CL Foy eds):pp.75–98. CRC Press, Boca Raton, FL, USA
- Weston LA. (1996). Utilization of allelopathy for weed management in agroecosystems. *Agronomy Journal* 88: pp. 860–866.
- Williams DA, Lemke TL. (2002). *Foye's Principles of Medicinal Chemistry* (5th ed.). Philadelphia: Lippincott Williams Wilkins. p. 25. ISBN 0-683-30737-1.