RESEARCH REVIEW JOURNALS
Volume-03
Issue-11
November -2018

ISSN: 2455-3085 (Online)
RESEARCH REVIEW International Journal of Multidisciplinary
www.rrjournals.com [UGC Listed Journal]

Antimicrobial (Bacteria, Fungi) Activity of Andrographis Paniculata and Detection of Andrographolide through TLC & HPLC Technique

[1]Miss. J.R.Kavitha & [2]A. Pamila Joyce

[1]Lecturer, Department of biotechnology, Mother Teresa Women's University, Kodaikanal (India)
[2]M.Phil Scholar, Department of Biotechnology, Mother Teresa Women's University, Kodaikanal (India)

ARTICLE DETAILS

Article History
Published Online: 10 November 2018

Keywords
TLC-Thin layer chromatography, HPLC-High Performance Liquid Chromatography, HPTLC-High Performance Thin Layer Chromatography

***Corresponding Author**
Email: kavi.216[at]rediffmail.com

ABSTRACT

Andrographis paniculata is annual herbaceous plant belonging to acanthaceae family it widely distributed on Asian countries. It's height about 30-110cm, its flowers having a rose purple spots on their petals. It is having a dark green stem about 0.3-1.0 m in height and 2-6 nm in diameter.Antiheptotoxicactivity, Antimalarial activity, Antithrombogenic activity, Anti-inflammatory activity, Antisnakevenom activity, respiratory system benefits, and antifertility activity. These are the main pharmacological potential of Andrographis paniculata. In addition the allopathic medicines are not curing all the human diseases for Eg; Jaundice, cancer, paralysis, diabetics, and skin diseases etc., which are not curable by allopathic medicines. Hence the perception of medicine turned around towards the herbal plants. There are five cultures were tested in this project, among that three of them is bacteria, and the remaining two of that are fungi bacteria i) vibrio cholera ii) klebsiella pneumonia.iii) salmonella typhi. Fungi Candida albicans, cryptococci. All of these cultures are purchases from Tamilnadu Agricultural University (TNAU) Coimbatore. From this current investigation it was observed that the methanolic, petroleum ether and dichloromethane extract of Andrographis paniculata inhibited the growth of human pathogens 90%, 0% and 40% respectively. The above findings recommend the further investigations of Andrographis paniculata to evaluate their potential for use as antimicrobials to plant pathogens. The developed HPLC method can be utilized for the quantitative determination of andrographolide in Andrographis paniculata herb samples, extracts and dosage forms.HPTLC method also can utilized for quantitative determination of Andrographis paniculata herb samples, extracts and dosage forms. The complexity of the chemical composition of herbal extracts, quality of the herbal extracts can only be assured by the use of validated analytical methods for identification and quantification of the active ingredients. The HPLC and HPTLC methods for the quantitative estimation of andrographolide were validated with regard to their specificity, precision, accuracy and linearity. At the same retention time the leaf extract of Andrographis paniculata having a high amount of Andrographolide compare than their standard.

1. Introduction

Andrographis paniculata is annual herbaceous plant belonging to acanthaceae family it widely distributed on Asian countries. It's height about 30-110cm, its flowers having a rose purple spots on their petals. It is having a dark green stem about 0.3-1.0 m in height and 2-6 nm in diameter.

Antiheptotoxic activity, Antimalarial activity, Antithrombogenic activity, Anti-inflammatory activity, Antisnakevenom activity, respiratory system benefits, antifertility activity. These are the main pharmacological potential of *Andrographis paniculata*. These are the main pharmacological potential of ***Andrographis paniculata.*** There is a continuous and urgent need to discover new antimicrobial compounds with diverse chemical structures and novel mechanisms of action because there has been an alarming increase in the incidence of neem and reemerging infectious disease. Another big concern is the development of resistance to the antibiotics in current clinical use, In addition the allopathic medicines are not curing all the human diseases for Eg; Jaundice, cancer, paralysis, diabetics, and skin diseases etc., which are not curable by allopathic medicines. Hence the perception of medicine turned around towards the herbal plants.

Anti-microbial activity and detection of Andrographolide through HPLC and TLC techniques.

a) Enumerate the antibacterial activity of *Andrographis paniculata*
b) Enumerates the antifungal activity of *Andrographis paniculata*
c) Detection of andrographolide through HPLC &TLC techniques*.*

2. Materials and Methods

There are five cultures were tested in this project, among that three of them is bacteria, and the remaining two of that are fungi bacteria i) vibrio cholera ii) klebsiella pneumonia.iii) salmonella typhi. Fungi Candida albicans, cryptococci. All of these cultures are purchases from Tamilnadu agricultural university (TNAU) Coimbatore.

Plant Samples

The leaf samples were purchased from ABS botanical garden, karipatti, Salem. The leaf of the *Andrographis*

paniculata is collected for this work, these leaves were allowed to shade dry at room temperature (28 ^{0} + 2^{0} c) for 4-7 days. The dried samples were ground by using a mixer and the fine powder were kept in airtight containers and stored that in room temperature in a dark place.

Antimicrobial Screening:

The petroleum Ether, Methanol, Dichloro methane extracts of the *Andrographis paniculata* were screened for antimicrobial activity by Disc Diffusion Method.

3. Results and Discussion

The efficacy of three different extracts of *Andrographis paniculata*t revealed that methanol extracts were more potent than petroleum ether followed by dichloromethane extract of *Andrographis paniculata*t. All human pathogens were highly susceptible to methanol extracts with variable concentrations viz., 10ml, 20ml and 25ml/well.Methanolic extracts showed maximum activity against three bacterial and two fungal strains. The methanolic extracts show the antimicrobial activity against all tested organisms, because methanol is a high polar in nature so all the active molecules of *Andrographis paniculata* is moved from the leaf extract easily.

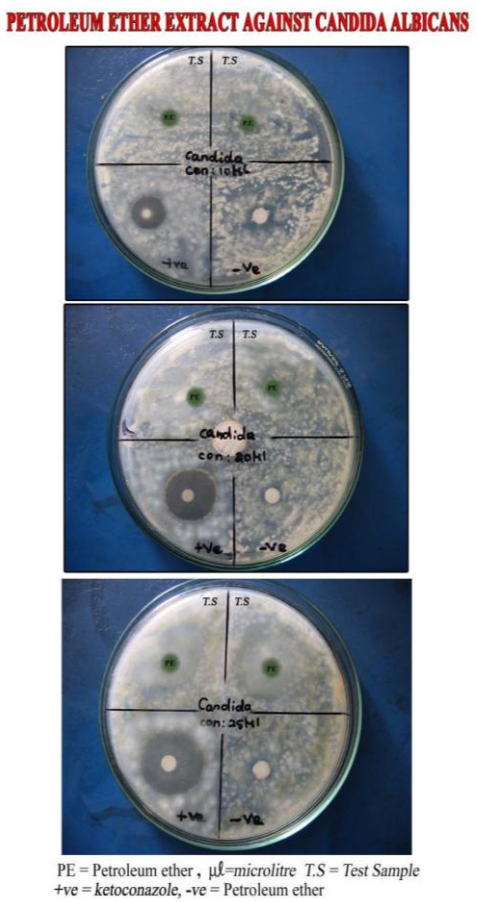
The petroleum ether did not show the antimicrobial activity so the active molecules do not move from the leaf extract. Antimicrobial activity against limited number of tested organism was reported with Dichloro methane extract of andrographis paniculata. From this current investigation it was observed that the methanolic, petroleum ether and dichloromethane extract of *Andrographis paniculata*t inhibited the growth of human pathogens 90%, 0% and 40% respectively. The above findings recommend the further investigations of *Andrographis paniculata* to evaluate their potential for use as antimicrobials to plant pathogens.

Discussion for TLC &HPLC:

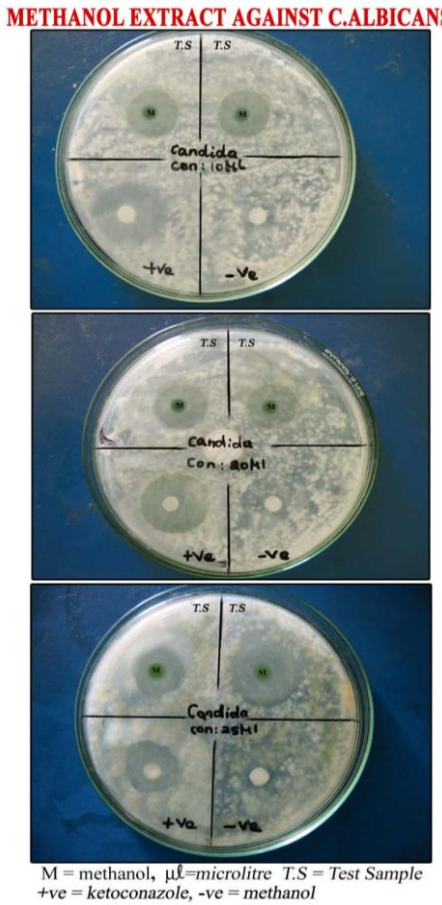
1. The complexity of the chemical composition of herbal extracts, quality of the herbal extracts can only be assured by the use of validated analytical methods for identification and quantification of the active ingredients. The HPLC and HPTLC methods for the quantitative estimation of andrographolide were validated with regard to their specificity, precision, accuracy and linearity.

Table 1. Effect of the three extracts of Andrographis paniculata on Candida albicans by disc diffusion method.

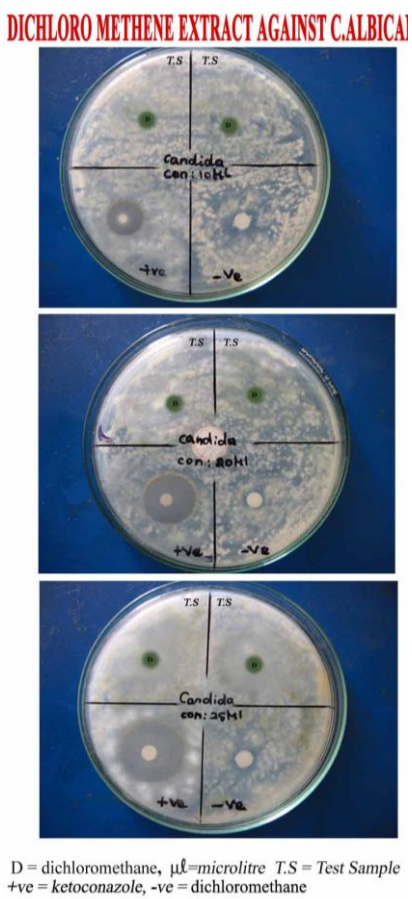
Name of the extract	Zone of Inhibition (Dia			
	Concentration	Test Sample	+ ve (Chloropenicol)	-ve (solvent)
Petroleum ether extract	10ml	-	-	-
	20ml	-	-	-
	25ml	-	-	-
Methanol extract	10ml	1.5 Dia	2.5 Dia	-
	20ml	2.4 Dia	2.9 Dia	-
	25ml	3.0 Dia	3.4 Dia	-
Dichloro methane extract	10ml	-	-	-
	20ml	-	-	-
	25ml	-	-	-



PE = Petroleum ether, µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = Petroleum ether



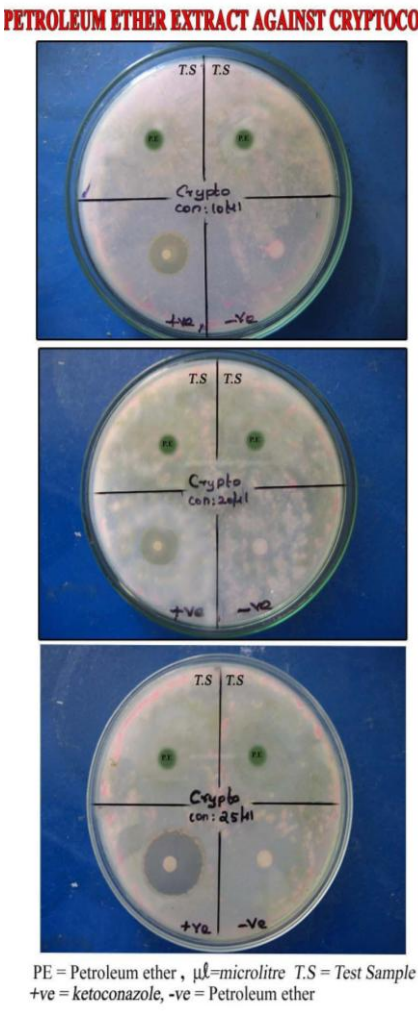
M = methanol, µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = methanol



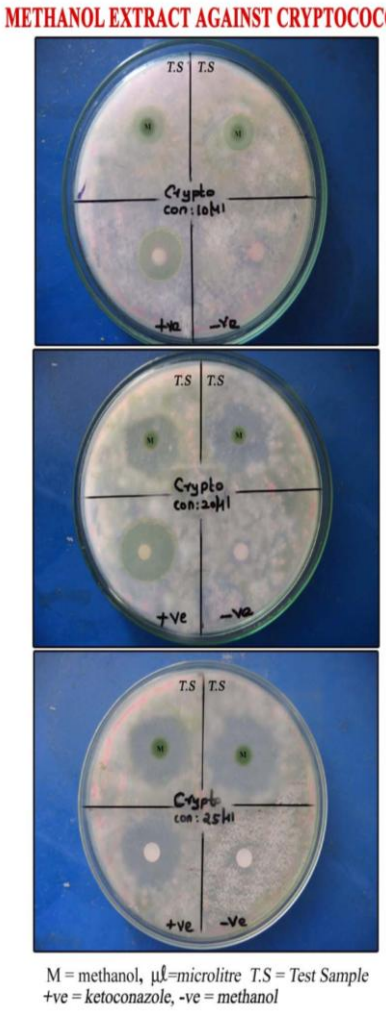
D = dichloromethane, µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = dichloromethane

Table 2. Effect of the three extracts of Andrographis paniculata on Cryptococci by dis diffusion method

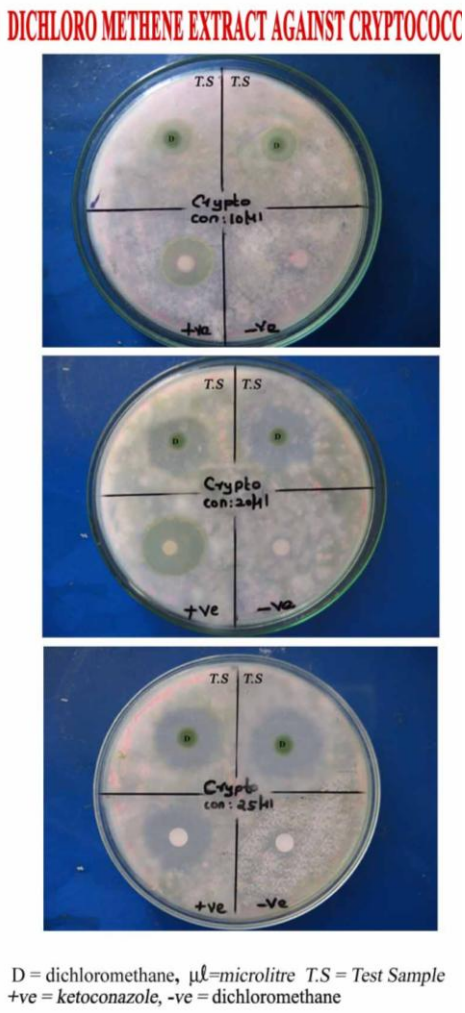
Name of the extract	Zone of Inhibition (Dia			
	Concentration	Test Sample	+ ve (Chlorempenicol)	-ve (solvent)
Petroleum ether extract	10ml	-	-	-
	20ml	-	-	-
	25ml	-	-	-
Methanol extract	10ml	1.7 Dia	2.0 Dia	-
	20ml	2.0 Dia	2.5 Dia	-
	25ml	2.6 Dia	3.0 Dia	-
Dichloro methane extract	10ml	1.4 Dia	1.0 Dia	-
	20ml	1.6 Dia	1.2 Dia	-
	25ml	1.9 Dia	2.0 Dia	



PE = Petroleum ether , µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = Petroleum ether

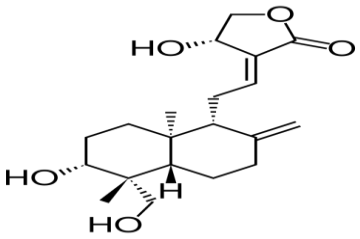


M = methanol, µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = methanol

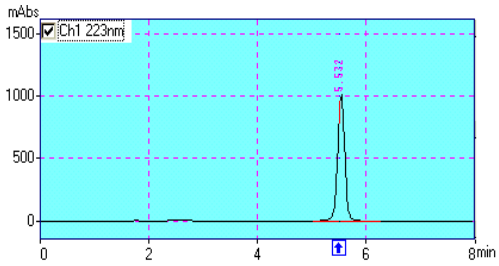


D = dichloromethane, µl=microlitre T.S = Test Sample
+ve = ketoconazole, -ve = dichloromethane

Detection of Andrographolide



Calculate the percentage from Peak areas in HPLC



HPLC and TLC

Compare than the other standard the 1g of test sample having a high amount of andrographolide it can identified at the same retention time.

4. Conclusion

From this current investigation it was observed that the methanolic, petroleum ether and dichloromethane extract of *Andrographis paniculata* inhibited the growth of human pathogens 90%, 0% and 40% respectively. The above findings recommend the further investigations of *Andrographis paniculata* to evaluate their potential for use as antimicrobials to plant pathogens.

The developed HPLC method can be utilized for the quantitative determination of andrographolide in *Andrographis paniculata* herb samples, extracts and dosage forms.HPTLC method also can utilized for quantitative determination of *Andrographis paniculata* herb samples, extracts and dosage forms. The complexity of the chemical composition of herbal extracts, quality of the herbal extracts can only be assured by the use of validated analytical methods for identification and quantification of the active ingredients. The HPLC and HPTLC methods for the quantitative estimation of andrographolide were validated with regard to their specificity, precision, accuracy and linearity. At the same retension time the leaf extract of Andrographis paniculata having a high amount of Andrographolide compare than their standard.

References

1. Amaryan, G., Astvatsatryan, V., Gabrielyan, E., et al., 2003, Double-blinding, placebo-controlled, randomized, pilot clinical trial of Immuno Guard-a standardized fixed combination of Andrographis paniculata Nees, with Eleutherococcus senticosus Maxim, Schizandra chinensis Bail. And Glycyrrhiza glabra L. extract in patients with Familial Mediterranean Fever. Phytomedicine, 10(4): 271-285
2. Amornsiriratanakul, P., 2000, Microencapsulation of Andrographis paniculata with water-soluble biopolymers. M.Sc. Thesis. Chulalongkorn University
3. Amroyan, E., Gabrielain, E., Panossian, A. et al., 1999, Inhibitory effect of andrographolide from Andrographis paniculata on PAF-induced platet aggregation. Phytomedicine, 6(1): 27-31
4. Ana, Ac. and Avadhoot, Y., 1991, Hepatotective effect of Andrographis paniculata against carbontetrachloride-induced liver damage. Arch Pharm Res, 14(1): 93-95
5. Anonymous, 1978, Beijing Medical College, Physiology Department: Experimental observation on the termination of pregnancy by the Andrographis herb (Andrographis paniculata N.). Acta Physiologica Sinica, 30(10: 75-80
6. Anonymous, 1990, Medicinal Plants Research Institute, Department of Medical Sciences. Handbook of Medicinal Plants for Primary Health Care. Text and Journal Corporation, Bangkok. pp 38-39
7. Anonymous, 2000, Bureau of Agricultural Commodities Promotion and Management. Department of Agricultural Extension. Handbook of Medicinal Plants and Spices. Volume 2. Drugs from Medicinal Plants. 1st ed. Bangkok
8. Atsawasuwan, P., 1996, Comparative Clinical and Microbiological Effects of Topical Subgingival Administration of Andrographis paniculata Gel and Metronidazole Gel as Adjunct in the treatment of Adult Periodontitis. M.Sc. Thesis, Graduate School, Mahidol University. Bangkok, Thailand
9. Atsawasuwan, P., Sirirat, M., Amomchat, C., Yudhasarasitihi, S. et al., 1998, Subgingival administration of Andrographis paniculata gel and metronidazole gel as an adjunct in the treatment of adult periodontitis: clinical and microbiological effect effects. Mahidol, J 5(2): 97-101
10. Batkhun, J., Hattori, K., Takano, F., et al., 2002, Suppression of NO production in activate macrophages in vito and ex vivo by neoandrographolide isolated from Andrographis paniculata. Biol Pharm Bull, 25(9): 1169-1174
11. Boonchaipanichwatana, P., 1996, The comparative clinical and microbiological effects of Andrographis paniculata gel and minocycline ointment as adjunct in the treatment of early onset periodontitis. M.Sc. Thesis, Graduate School, Mahidol University. Bangkok, Thailand
12. Burgos, R. A., Caballero, E. E., Sanche, N. S. et al., 1997, Testicular toxicity assessment of Andrographis paniculata dried extract in rats. J. Ethnopharmacol., 58: 219-224
13. Caceres, D. D. et al., 1997, Prevemtion of common cold with Andrographis paniculata dried extract. A pilot doble-blind study. Phytomedicine, 4: 101-104
14. Caceres, D. D. et al., 1997, Prevention of common colds with Andrographis paniculata dried extract. A oilot double-blind study. Phytomedicine, 4: 101-104
15. Caceres, D. D. et al., 1999, Use of visual analogue scale measurement (VAS) to assess the effectiveness of standardized Andrographis paniculata extract SHA-10 in reducing the symptoms of common cold. A randomized double blind-placebo study. Phytomedicine, 64: 217-223
16. Chang, R. S., Ding, L., Chen, G. Q. et al., 1991, Dehydroandrographolide succinic acid monoester as an inhibitor against the human immunodeficiency virus. Prc Soc Exp Biol Med, 197(1): 56-66
17. Chanwitheesuk, A., Rakriyatham, N., Teerawutgukrag, A. ang Wonchanapiboon, T., 2002 Screening of antioxidants from Thai vegatables and herbs. Chaing Mai J Sci, 29(1): 1-5
18. Chiou, W. F., Chen, C. F. and Lin, J. J., 2000, Mechanism of suppression of inducible nitric oxide synthase (iNOS) expression in RAW 264.7 cells by andrographolide. Br J Pharmacol, 129(8): 31553-31560
19. Chiou, W. F., Lin, J. J. and Chen, C. F., 1998, Andrographolide suppressures the expression of inducible nitric oxide synthase in macrophage and restores the vasoconstriction in rat aorta treated with lipopolysaccharide. Br J Pharmacol, 125(2): 327-334
20. Choudhury, B. R. and Podder, M. K., 1984, Andrographolide and kalmegh(Andrographis paniculata) extract: in vivo and in vito effect on hepatic lipid peroxidation. Methods Find Exp Chin Pharmacol, 6(9): 481-485
21. Guo, Z. L., Zhao, H. Y. and Fu, L., 1996, Protective effects of API0134 on myocardial ischemia and reperfusion injury. J Tongji Med, 16(4): 193-197
22. Gupta, S., Choudhry, M. A., Yadava, J. N. S. et al., 1990, Antidiarrhoeal activity of diterpenes of Andrographis paniculata (Kal-Megh) against Escherichia coli enterotoxin in in vivo models. Int. J. Crude Drug Res., 28(4): 273-283
23. Gupta, S., Choudhry, M. A., Yadava, J. N. S. et al., 1990, Antidiarrhoeal activity of diterpenes of Andrographis paniculata (Kal-Megh) against Escherichia coli enterotoxin in In vivo models. Int. J. Crude Drug Res., 28(4): 273-283
24. Hancke, J., Burgos, R., Caceres, D., et al., 1995, A double-blind study with a new monodrug Kan Jang: Decrease of symptoms and improvement in the recovery from common cold. Phytother. Res., 9: 559-562

25. Handa, S. S. and Sharma, A., 1990, Hepatoprotective activity of andrographolide from Andrographis paniculata against carbontetrachloride. Ind J Med Res, 92: 276-283
26. Handa, S. S. and Sharma, A., 1990, Hepatoprotective activity of andrographolide against galactosamine & paracetamol intoxication in rats. Ind J Med Res, 92: 284-292
27. Husen, R., Pihie, A. H. and Nallappan, M., 2004, Screening for antihyperglycaemic activity in several local of Malaysia. J Ehtnopharmacol, 95(2-3): 205-208
28. Iruretagoyena, M. I., Tobar, J. A., Gonzalez, P. A. et al., 2005, Andrographolide interferes with T cell activation and reduces experimental autoimmune encephalomyelitis in the mouse. J Pharmacol Exp Ther., 312(1): 366-372
29. Kamdem, R. E., Sang, S., and Ho, C. T., 2002, Mechanism of superoxide scavenging activity of neoandrographolide – a natural product from Andrographis paniculata Nees. J Agric Food Chem, 50(16): 4662-4665
30. Kapil, A., Koul, I. B., Banerjee, S. K. and Gupta, B. D., 1993, antihepatotoxic effects of major diterpenoid constituents of Andrographis paniculata. Biochem Pharmacol, 46(6): 182-185
31. Ketpanyapong, W., 1999, The effects of Andrographis paniculata and Psidium guajava Leaves on Growth and Feed Efficiency of Diarrheic Piglets. M.Sc. Thesis. Graduate School, Prince of Songkla University, Thailand
32. Komwatchara, T., 1996, The development of Andrographis paniculata Extract Gel for Microbial Inhibition in adult Periodontitis. M.Sc. Thesis, Graduate School, Mahidol University. Bangkok, Thailand
33. Kumar, R. A., Sridevi, K., Kumar, N. V. et al., Anticancer and immumostimulatory compounds from Andrographis paniculata. J Ethnopharmacol., 92(2): 291-295
34. Leelarasamee, A., Trakulsomboon, S. and Sittissomwong, N., Undertectable antibacterial activity of Andrographis paniculata (Burm.) Wall. Ex Ness. J Med Assoc Thai, 73(6): 299-304
35. Lomsong, J., Benjavongkulchai, E. and Kuvatanasuchati, J., Inhibitory effect of some herbal extracts on adherence of Streptococcus mutans. J Ethnopharmacol., 92(2): 281-289
36. Muangman, V., Viseshindh, V., Ratana-Olarn, K. and Buadilok, S., 1995, The usage of Andrographis paniculata following extracorporeal shock wave lithotripsy (ESWL). J Med Assoc Thai, 78(6): 310-313
37. Onlaor, S., 2000, Microencapsulation of Andrographis paniculata Nees Extract for sustained release by solvent evaporation technique. M.Sc. thesis. Chulalongkorn University
38. Panossian, A., Davtyan, T., Gukassyan, N. et al., 2002, Andrographolide and kan Jang-fixed combination of extract SHA-10 and extract SHE-3- on proliferation of human lymphocytes, production of cytokines and immune activation markers in the whole blood cells culture. Phytomedicine, 9(7): 598-605
39. Panossian, A., Kochkian, A., Gabrielain, E. et al., Effect of Andrographis paniculata extract on progesterone in blood plasma of pregma,t rats. Phytomedicine, 3(3): 157-162
40. Pearnpijit, P., 1993, Antimicrobial activities of Boesenbergia pandurata, Clinacanthus nutans, Andrographis paniculata, Garcinia mangostana, Garcinia hanburyi, Kaempferia rotunda and Synthetic polyamine compounds. M.Sc. Thesis, Graduate School, Prince of Songkla University, Thailand
41. Puri, A. et al., 1993, Immomostimulant agents from Andrographis paniculata. J Nat. Prod., 56: 995-999
42. Rajagopal, S., Kumar, R. A., Deevi, D. S. et al., 2003, Andrographolide, a potential cancer therapeutic agent isolated from Andrographis paniculata. J Exp Ther Oncol, 3(3): 147-158
43. Rassameemasmaung, S., 1996, Subgingival administration of Andrographis paniculate Gel as an Adjunct in the Treatment of Adult Periodontitis. M.Sc. Thesis, Graduate School, Mahidol University. Bangkok, Thailand
44. Rassameemasmaung, S., Sirirat, M., Komwatchara, T., Roianapanthu, P. et al., 1998, Subgingival administration of Andrographis paniculata gel as an adjunct in the treatment of adult periodontitis. Mahidol, J 5(1): 9-15
45. Sangalunkarn, V. et al., 1990, Effects of andrographolide, neoandrographolide and 14-deoxy-11,12-didehydroandrographolide on Isolated Rat Smooth Muscle Contraction. Th J Pharm Sci, 15(1): 5-16
46. Shen, Y. C., Chen, C. F. and Chioys, W. F., 2000, Suppression of rat neutrophil reactive oxygen species production and adhesion by the diterpenoid lactone andrographolide. Planta Med, 66(4): 314-317
47. Shen, Y. C., Chen, C. F. and Chioys, W. F., 2002, Andrographolide prevents oxygen radical production by human neutrophil: possible mechanism(s) involved in its anti-inflammatory effect. Br J Pharmacol, 135(2): 399-406
48. Shukla, B., Visen, P. K., Patnaik, G. K. and Dhawan, B. N., 1992, Choleretic effect of andrographolide in rats and guinea pigs. Planta Med, 58(2): 146-149
49. Sindermsuk, J., 1993, The Antibacterial Activities of the Pure Extract Compounds from Andrographis paniculata on Predominate Pathogenic Enteric Bacilli in Thailand. Bull Dept Med Serv, 18: 394-400
50. Singh, R. P., Banerjee, A. and Rao, A. R., 2001, Modulatory influence of Andrographis paniculata on mouse hepatic and extrahephatic carcinogen metabolizing enzymes and antioxidant status. Phytother Res 15: 382-390
51. Sittisomwongse, N. et al., 1989, Acute and sub-chronic toxicity studies of Andrographis paniculata Nees. Th. J. Pharm. Sci., 14(2): 109-118
52. Suttajit, M., Immak, K., Sukonpradit, R. and Suttajit, S., 1998, Antioxidative activity in medicinal herbs for HIV-infected patients. Proceeding of the 9th Annual Technical Meeting of the Department of Medical Sciences. pp 53
53. Suttajit, M., Immark, K., Sukonpradit, R. and Suttajit, S., 1998, Antioxidative activityin medicinal herbs for HIV-infected patients. Proceeding of the 9th Annual Technical Meeting of the Department of Medical Sciences. pp 53
54. Thamlikitkul, V. et al., 1991, Effect of Andrographis paniculata Nees for pharyngotonsilltis in adults. J. Med. Assoc. Thai, 74(10): 433-442
55. Trivedi, N. and Rawal, U. M., 1998, Effect of aqueous extract of Andrographis paniculata on severe liver tumor. Ind J Pharmacol, 30: 318-322
56. Trivedi, N. and Rawal, U. M., 2000, Hepatoprotective and toxicological evaluation of Andrographis paniculata on severe liver damage. Ind J Pharmacol, 32: 288-293
57. Trivedi, N. and Rawal, U. M., 2001, Hepatoprotective and antioxidant property of Andrographis paniculata(Nees) in BHC induced liver damagein mice. Ind J Exp Biol, 39(1): 41-46
58. Udomsksom, W., 1993, Effect of andrographolide and Andrographis paniculata on ethanol hepatotoxicity in rats. MS Thesis, Chulalongkorn University, Bangkok. pp 155
59. Vedavathy, S. and Rao, K. N., 1991, Antipyretic activity of six indigenous medicinal plants of Tirumala Hills, Andhra Pradesh, India. J Ethnopharmacol., 33: 193-196
60. Visen, P. K., Shukla, B., Patnaik, G. K. and Dhawan, B. N., 1993, Andrographolide protects rat hepatocytes against paracetamol-induced damage. J Ethnopharmacol, 40(2): 131-136
61. Wang, D. W. and Zhao, H. Y., 1993, Experimental studies on prevention of atherosclerotic arterial arterial atenosis and restenosis after angioplasty with Andrographis paniculata Nees and fish oil. J Tongji Med Univ, 13(4): 193-198
62. Wang, D. W. and Zhao, H. Y., 1994, P revention of atherosclerotic arterial stenosis after angioplasty with

Andrographis paniculata Nees and fish oil. Experimental studies of effects and mecharnism. Chin Med J (Eng), 107(6): 464-450

63. WHO, 2002, Herba Andrographidis. In: WHO monographs of selected medicineal plants. Vol. 2. pp 12-24
64. Yu, B. C., Hung, C. R., Chen, W. C. et al., 2003, Antihyperglycaemic effect of andrographolide in streptozotocin-induced diabetic rats. Planta Med, 69(12): 1075-1079
65. Zhang, C. Y. and Tan, B. K., 1996, Hypotensive activity of aqueous extract of Andrographis paniculata in rats. Clin Exp Pharmacol Physiol, 23(8): 657-658
66. Zhang, C. Y. and Tan, B. K., 1997, Mechanismm of cardiovascular activity of Andrographis paniculata in the anaesthetized rat. J Ehanopharmacol, 56(2): 97-101
67. Zhang, X. F. and Tan, B. K., 2000, Anti-diabetic property of ethanolic extract of Andrographis paniculata in streptozotocin-diabetic rats. Acta Pharmacol Sin, 21(12): 1157-1164

