

LIPID AND BLOOD PRESSURE LOWERING POTENTIAL OF *MIKANIA MICRANTHA* THROUGH ENZYMATIC INHIBITION

Amirah Haziyah Ishak¹, Nurul Husna Shafie^{1,2*}, Norhaizan Mohd Esa¹, and Hasnah Bahari³

¹Department of Nutrition and Dietetics, Faculty of Medicine and Health Sciences, UPM Serdang, Selangor, Malaysia

²Laboratory of UPM-MAKNA Cancer Research, Institute of Bioscience, UPM Serdang, Selangor, Malaysia

³Department of Human Anatomy, Faculty of Medicine and Health Sciences, UPM Serdang, Selangor, Malaysia

* Corresponding author's email: nhusnashafie@upm.edu.my

INTRODUCTION

Mikania micrantha Kunth (Asteraceae) or locally known as "Selaput tunggul" is a perennial creeping vine that widely used by local practitioners for treatment or prevention of various diseases. In Malaysia, *M. micrantha* is consumed as a juice (by boiling in hot water) as an alternative to reduce cholesterol, high blood pressure, and glucose.

Hyperlipidemia is defined as increased blood cholesterol, triglycerides or both, while hypertension is defined as persistent elevation of systolic (> 140 mmHg) and diastolic (> 80 mmHg) blood pressures [1]. A combination of different strategies is used to treat and manage hyperlipidemia and hypertension. One of them is through inhibition of the key enzymes responsible for hyperlipidemia and hypertension.

Aim: To examine the potential of various extracts of the leaves and stems of *M. micrantha* to inhibit enzymes relevant to hyperlipidemia *i.e.*, pancreatic lipase (PL), lipoprotein lipase (LPL), 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMGR) and hypertension *i.e.*, angiotensin-I converting enzyme (ACE).

METHODOLOGY

Sample Preparation

The leaves and stems of *M. micrantha* were separated, washed, dried, and ground to produce fine powder.

Sample Extraction

The leaves and stems of *M. micrantha* were extracted with hot water, cold water, 70% ethanol and ethyl acetate.

Determination of Enzymatic Inhibition

The inhibition activities of *M. micrantha* extracts were determined spectrophotometrically using PL, LPL, HMGR, and ACE inhibition assays.

Table 1 – Enzymatic inhibition assays

Inhibition assays	Enzymatic inhibition assays	Measure rate of NADPH consumed at 340 nm [4]
Pancreatic lipase (PL)	Measure the hydrolysis of <i>p</i> -nitrophenyl butyrate (<i>p</i> -NPB) to <i>p</i> -nitrophenol at 405 nm [2,3]	
Lipoprotein lipase (LPL)		
HMG-CoA reductase (HMGR)		
Angiotensin-I converting enzyme (ACE)	Measure release of hippuric acid (HA) from synthetic substrate Hippuryl-His-Leu (HHL) at 410 nm [5]	

RESULTS

Pancreatic Lipase Inhibition

Table 2 – IC₅₀ values for pancreatic lipase (PL) inhibitory activity of *M. micrantha* extracts

Extraction solvents	IC ₅₀ (µg/mL)	
	Leaves	Stems
Hot water	4.56 ± 0.07 ^{ab}	42.37 ± 4.63 ^d
Cold water	28.97 ± 4.22 ^{cd}	16.93 ± 1.99 ^{bc}
70% ethanol	8.02 ± 1.56 ^{ab}	4.49 ± 2.50^{ab}
Ethyl acetate	18.00 ± 3.78 ^{bc}	16.73 ± 0.70 ^{bc}
Orlistat	0.31 ± 0.01 ^a	

Results are expressed as the means ± SEM (n=3). Means with different letters are significant at p < 0.05. Orlistat is a positive control. The concentration of extracts and orlistat used were 0 – 100 µg/mL. IC₅₀ is the concentration of extracts (µg/mL) required to inhibit PL and LPL activity by 50%. A low IC₅₀ indicates the highest PL and LPL inhibitory activity.

Lipoprotein Lipase Inhibition

Table 3 – IC₅₀ values for lipoprotein lipase (LPL) inhibitory activity of *M. micrantha* extracts

Extraction solvents	IC ₅₀ (µg/mL)	
	Leaves	Stems
Hot water	4.59 ± 0.87 ^a	8.04 ± 2.75 ^a
Cold water	2.34 ± 1.88 ^a	2.70 ± 1.79 ^a
70% ethanol	1.42 ± 0.48^a	4.26 ± 1.23 ^a
Ethyl acetate	7.35 ± 2.68 ^a	5.69 ± 2.46 ^a
Orlistat	1.98 ± 1.22 ^a	

HMG-CoA Reductase Inhibition

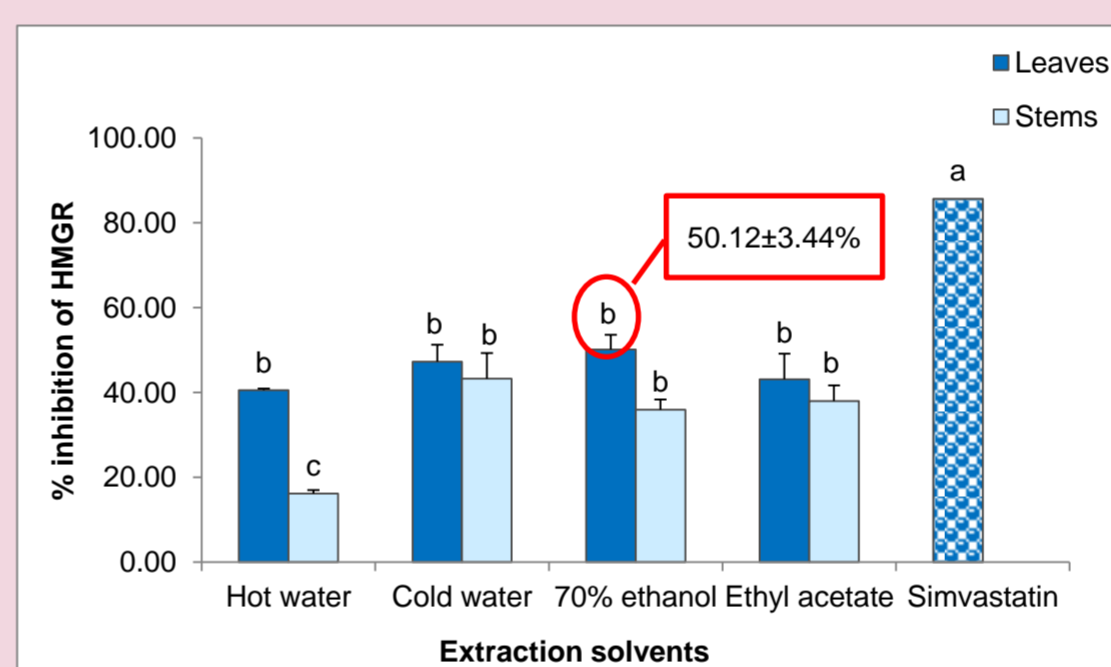


Fig. 1 - HMGR inhibitory activity of *M. micrantha* extracts. Data are shown as the means ± SEM (n=3). Bars with different letters are significant at p < 0.05. Concentration of extracts used was 1000 µg/mL. Simvastatin is a positive control at 100 µM (419 µg/mL).

ACE Inhibition

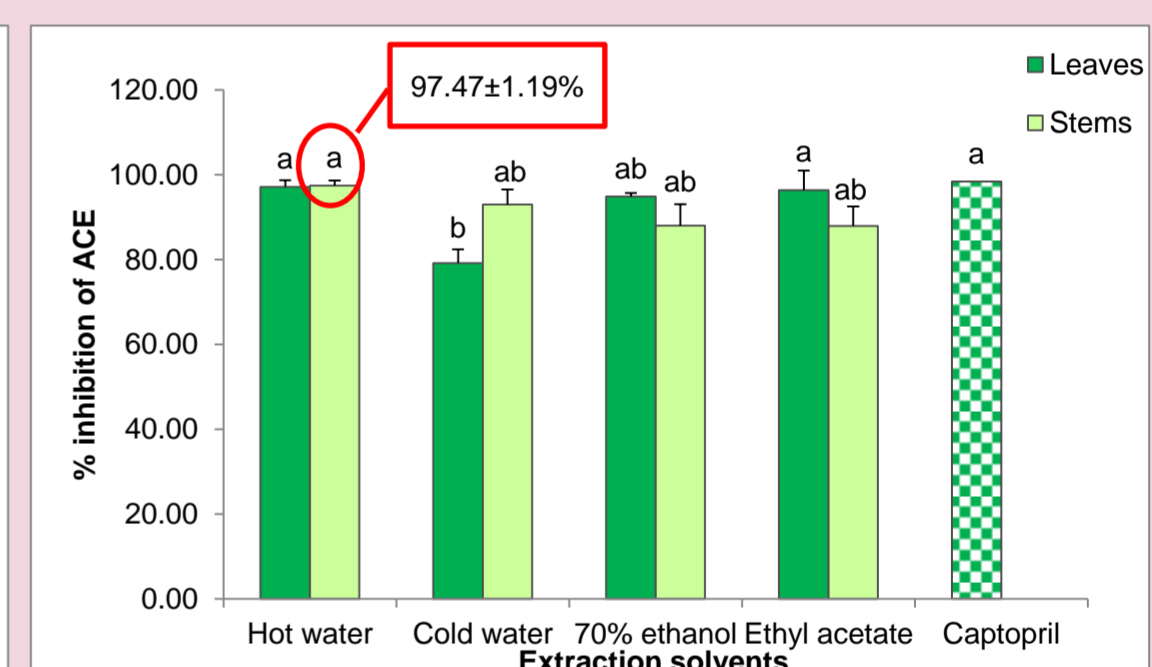


Fig. 2 - ACE inhibitory activity of *M. micrantha* extracts. Data are shown as the means ± SEM (n=3). Bars with different letters are significant at p < 0.05. Concentration of extracts and captopril used was 1000 µg/mL.

DISCUSSION

- The ethanol stems, hot water leaves and ethanol leaves extract exhibited the highest PL inhibitory activity.
- The ethanol leaves extract demonstrated the highest LPL and HMGR activities.
- Hot water stems extract showed the highest ACE inhibitory activity but least inhibitory activity against PL, LPL, and HMG-CoA reductase.
- Presence of alkaloids, terpenoids, tannins, cardiac glycosides, and luteolin in 70% ethanol extract of *M. micrantha* could be the potential PL, LPL and HMGR inhibitors [6-7].

CONCLUSION

All extracts exhibited remarkable inhibitory activities against pancreatic lipase, lipoprotein lipase, HMG-CoA reductase, and angiotensin-I converting enzyme *in vitro*.

This study revealed the potential of *M. micrantha* extracts as anti-hyperlipidemic and anti-hypertensive agents.

REFERENCES

- Otsuka, T., Takada, H., Nishiyama, Y., Kodani, E., Saiki, Y., Kato, K., & Kawada, T. (2016). Dyslipidemia and the risk of developing hypertension in a working-age male population. *Journal of the American Heart Association*, 5(3), e003053.
- Kim, Y. S., Lee, Y., Kim, J., Sohn, E., Kim, C. S., Lee, Y. M., ... & Kim, J. S. (2012). Inhibitory activities of *Cudrania tricuspidata* leaves on pancreatic lipase *in vitro* and lipolysis *in vivo*. *Evidence-based Complementary and Alternative Medicine*, 2012.
- Saraphanchotiwitthaya, A., & Sripalakit, P. (2014). Effect of *Morinda citrifolia* Linn. leaf extracts on *in vitro* lipase activity. *Chiang Mai Journal of Science*, 41(5-1), 1182-1193.
- Iqbal, D., Khan, M. S., Khan, A., Khan, M., Ahmad, S., Srivastava, A. K., & Bagga, P. (2014). *In vitro* screening for β-hydroxy-β-methylglutaryl-coa reductase inhibitory and antioxidant activity of sequentially extracted fractions of *Ficus palmata* Forsk. *BioMed Research International*, 2014.
- Ghanbari, R., Zarei, M., Ebrahimpour, A., Abdul-Hamid, A., Ismail, A., & Saari, N. (2015). Angiotensin-I converting enzyme (ACE) inhibitory and anti-oxidant activities of sea cucumber (*Actinopyga lecanora*) hydrolysates. *International Journal of Molecular Sciences*, 16(12), 28870-28885.
- Khatun, R., Roy, S., & Rahman, M. A. A. (2017). *In vitro* comparative evaluation of anti-inflammatory and thrombolytic activity of three Mikania species available in Bangladesh. *Journal of Pharmacognosy and Phytochemistry*, 6(5), 1007-1011.
- Wang, R. L., Peng, S. L., Zeng, R. S., Ding, L. W., & Xu, Z. F. (2009). Cloning, expression and wounding induction of β-caryophyllene synthase gene from *Mikania micrantha* HBK and allelopathic potential of β-caryophyllene. *Allelopathy Journal*, 24, 35-44.