제출자료 1

Hepatoprotective Effects of *Allium ochotense* Extract on Alcohol-induced Fatty Liver in C57BL/6 Mice

Min Ji Go[1,*], Hyo Lim Lee[1], Han Su Lee[1], Ju Hui Kim[1], In Young Kim[1], Seon Jeong Sim[2], and Ho Jin Heo[1]

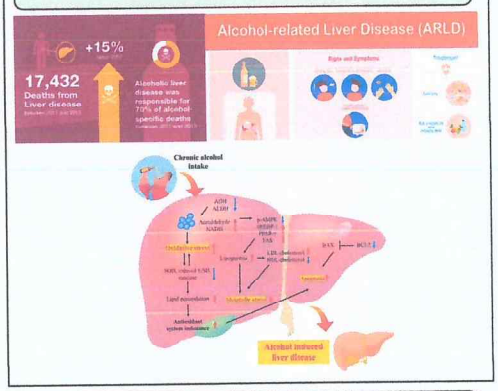
[1]*Division of Applied Life Science (BK21), Institute of Agriculture and Life Science, Gyeongsang National University*
[2]*Forest Research Department, Gyeongsangnam-do Forest Environment Research Institute*

GYEONGSANG NATIONAL UNIVERSITY

Abstract

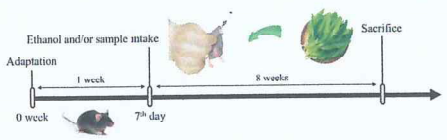
This study was performed to investigate the protective effects of *Allium ochotense* on fatty liver and apoptosis in chronic alcohol-induced hepatotoxicity. The mixture (aqueous and 60% ethanol extract, 2:8) of extract of *Allium ochotense* (AM) regulated the levels of lipid metabolism-related biomarkers such as total cholesterol, triglyceride, LDL and HDL-cholesterol. Also, AM ameliorated the levels of liver toxicity-related biomarkers such as glutamic oxaloacetic transaminase (GOT), glutamic pyruvic transaminase (GPT), and total bilirubin. AM improved antioxidant system by reducing malondialdehyde contents and increasing superoxide dismutase (SOD) level and reduced glutathione content. In addition, AM increased the expression levels of alcohol dehydrogenase and acetaldehyde dehydrogenase. Moreover, AM improved fat accumulation by regulating the expression levels of p-AMP-activated protein kinase (p-AMPK) and peroxisome proliferator-activated receptor-gamma (PPAR-γ). AM showed anti-apoptosis effect by regulating the expression levels of BAX and BCl-2. Therefore, these results suggest that AM might be a potential prophylactic agent for the treatment of alcoholic fatty liver.

Introduction



Materials & Methods

✓ **Animal experimental design**



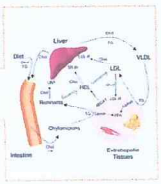
✓ **Blood serum biochemical analysis**

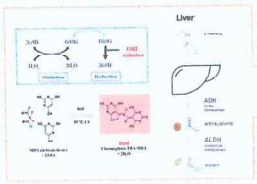
- Fatty liver related factors

: Total cholesterol (TCHO), triglyceride (TG), low-density lipoprotein (LDL), and high-density lipoprotein (HDL)-cholesterol

- Liver toxicity related factors

: GOT, GPT, and total bilirubin





✓ ***ex vivo* test**

- Antioxidant system

: MDA, Reduced GSH, SOD

- Western blot

: ADH, ALDH

: p-AMPK, PPAR-γ

: BAX, BCl-2, BAX/BCl ratio

Results

Table 1. Effect of the mixture of *Allium ochotense* (AM) extract on fatty liver related serum biomarkers.

	TCHO (mg/dl)	TG (mg/dl)	HDLC (mg/dl)	LDLC (mg/dl)
NC	94.50±3.41[c]	115.50±5.20[b]	74.00±4.16[bc]	3.45±1.59[b]
NS	95.75±7.89[bc]	101.75±12.66[b]	76.25±2.06[bc]	4.00±0.59[b]
AL	110.25±1.50[a]	143.50±12.50[a]	70.50±3.70[c]	11.4±3.98[a]
PC	100.50±6.66[bc]	110.75±34.79[b]	80.25±7.80[b]	2.50±2.05[b]
SL	97.25±5.68[bc]	104.25±16.94[b]	82.25±11.90[ab]	0.90±4.67[b]
SH	105.00±9.70[ab]	113.25±2.50[b]	91.00±4.76[a]	0.55±0.91[b]

The results shown are mean ± SD (n = 5). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

Table 2. Effect of the mixture of *Allium ochotense* (AM) extract on liver toxicity related serum biomarkers.

	GOT (U/L)	GPT (U/L)	TBIL (mg/dl)
NC	45.40±4.45[b]	29.40±1.52[ab]	0.50±0.07[b]
NS	44.80±1.30[b]	29.20±4.76[b]	0.52±0.22[b]
AL	54.00±10.12[a]	35.40±4.92[a]	0.70±0.07[a]
PC	46.80±1.79[b]	35.40±2.70[a]	0.44±0.05[b]
SL	47.40±2.30[b]	34.20±4.09[ab]	0.38±0.04[b]
SH	46.00±2.00[b]	29.40±1.82[b]	0.42±0.04[b]

The results shown are mean ± SD (n = 5). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

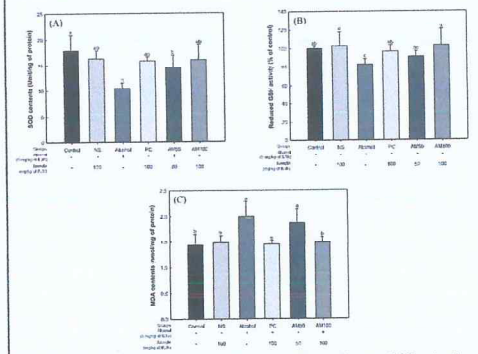


Figure 1. Ameliorating effect of the mixture of *Allium ochotense* (AM) extract on antioxidative system biomarkers. The SOD contents (A), reduced GSH activity (B), and MDA contents (C). The results shown are mean ± SD (n = 5). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

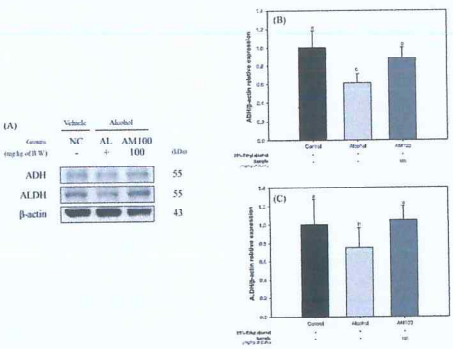


Figure 2. Ameliorating effect of the mixture of *Allium ochotense* (AM) extract on alcohol degradation enzyme in the liver. The Western blot band image (A) and the expression levels of ADH (B) and ALDH (C). The results shown are mean ± SD (n = 5). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

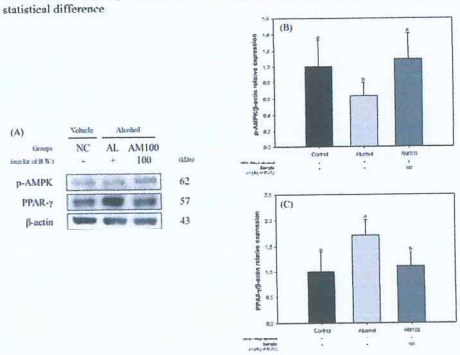


Figure 3. Regulating effect of the mixture of *Allium ochotense* (AM) extract on lipid metabolism in the liver. The Western blot band image (A)and the expression levels of p-AMPK (B) and PPAR-γ (C). The results shown are mean ± SD (n = 5). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

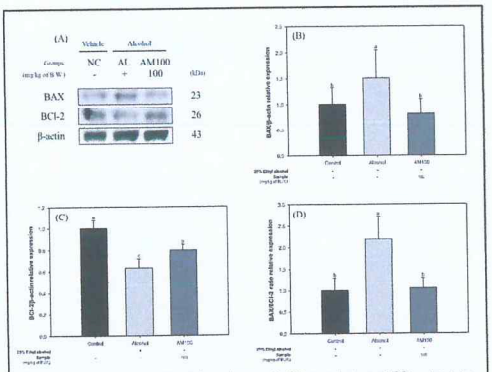
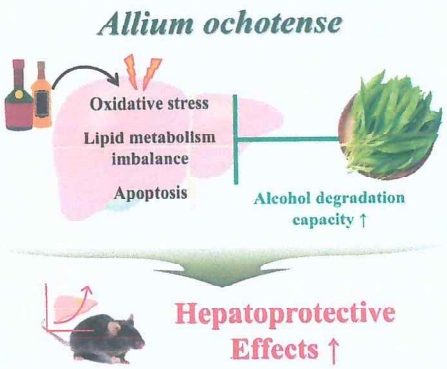


Figure 4. Regulating effect of the mixture of *Allium ochotense* (AM) extract on apoptosis related protein in the liver. The Western blot band image (A) and the expression levels of BAX (B), BCl-2 (C), and BAX/BCl-2 ratio (D). The results shown are mean ± SD (n = 3). Data were statistically considered at p<0.05, and different small letters represent statistical difference.

Conclusion



- *Allium ochotense* (AM) extract improved the levels of lipid metabolism and liver toxicity biomarkers in serum.
- Treatment with AM extract reduced MDA levels by activating the antioxidant system in the liver.
- In addition, AM extract increased the expression levels of alcohol-degrading enzymes such as ADH and ALDH.
- Furthermore, it protected liver cells by regulating the levels of biomarkers related to lipid metabolism and apoptosis in the liver.
- Therefore, this findings suggests that AM extract can be used as a health functional food material to help prevent alcoholic liver disease.

References

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