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Effect of taurine in chronic alcoholic patients

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Abstract

A study was undertaken to investigate the dietary effect of taurine in chronic alcoholic patients. The 30 chronic alcoholic patients with 2 to 5 times over normal activities of aspartate transaminase (AST) or alanine transaminase (ALT) were selected and equally divided into taurine and control groups. In taurine group, each patient took 6 g taurine per day divided into 3 times for three months, and then stopped treatment for 1 month. In control group, patients took placebo without taurine for 4 months. It was found that AST and ALT activities and levels of cholesterol, triglyceride (TG), bilirubin, and thiobarbituric acid relative substances (TBARS) of serum plasma in the taurine group were all decreased, but increased alcohol dehydrogenase (ADH) and aldehyde dehydrogenase (ALDH) activities and serum vitamins concentrations. Except for level of TG, all of them showed significantly different after taking taurine for 2 or 3 months. It indicated that taurine plays an important role in the properties of antioxidation and has some improvements on the liver test of chronic alcoholic patients.

Keywords: Aspartate transaminase; Antioxidation; Alcoholic patient; Liver test; Taurine
1. Introduction

Alcoholic is a disease with powerful negative effects that impact not only the individual afflicted but also society at large. In the United States, alcoholic is the third leading cause of death and often leads to other sequela\(^1\), such as cirrhosis, which are also leading causes of death\(^2\). Hepatic, pancreatic, and cardiovascular systems are the major targets of chronic alcohol abuse and alcoholic, alcoholic hepatitis is the single most common cause of mortality and morbidity from the liver diseases\(^3\). Chronic alcohol abuse is associated with wide-ranging neurological damage, such as Wernicke encephalopathy, dementia, delirium tremens and peripheral polyneuropathy. However, this area of inquiry will need to keep pace with the growing level of knowledge on how to best treat chronic alcoholic. The widespread effects of alcoholic necessitate its study with the goal of increasing treatment efficacy and thus reducing the cost to society.

In our previous papers, we found that dietary taurine could possess a protective liver test and detoxification action against oxidized oil and heavy metal induced injuries\(^4,5\). Taurine is a sulfur-containing amino acid that conjugates with bile acids in the liver\(^6\) and an essential amino acid for cat\(^7\). It is well known that taurine is rich in fish products, especially in mollusks and fish liver\(^8\). Its physiological functions include bile acid conjugation, detoxification, osmoregulation, antioxidation, preventing lipid peroxidation, cell membrane stabilization, neuromodulation and calcium flux regulator\(^9,10\). Taurine is largely obtained from the diet, predominantly through eggs, meat, and seafood\(^11\). Additionally, taurine acts as an antioxidant and plays a role in detoxification, membrane stabilization a comma osmoregulation, neuromodulation, and brain and in retinal development. According to recent reports
by Yang et al.\textsuperscript{12} and Chang et al.\textsuperscript{13}, taurine can accelerate cholesterol degradation to form bile acid thus increasing fecal bile acid excretion, and enhancing peroxisome proliferator-activated receptor-\(\alpha\) (PPAR-\(\alpha\)) expression which leads to an increase in hepatic lipid expenditure in high-fat/cholesterol dietary rodents. Chang et al.\textsuperscript{13} also indicated that supplementing taurine can reduce serum glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT) of high-fat/cholesterol dietary hamsters. At the same time, increased hepatic self-antioxidant capacities and decreased lipid peroxidation were also observed in high-fat/cholesterol dietary hamsters co-treated with taurine, taurine supplementation can lower liver triacylglycerol levels in alcohol fed rats via the downregulation of fatty acid synthase (FAS) and malic enzyme (ME) gene expressions and the upregulation of PPAR-\(\alpha\) gene expression. Additionally, taurine supplementation also decreases TBARS values in sera and livers which result in lower hepatic TNF-\(\alpha\) level and MMP-9 activity. Recently, Fang et al.\textsuperscript{14} indicated that an acceleration of alcohol metabolism via upregulating alcohol dehydrogenase (ADH), catalase (CAT), and aldehyde dehydrogenase (ALDH) activities, less microvesicular steatosis, and necrotic cells in livers were observed when chronic alcohol-fed rats were supplemented with taurine. Hence, it prompted us to investigate the dietary effect of taurine on the chronic alcoholic patients. One of the promising research areas in biomedical science today is the focus on alcoholic and taurine. This article presents a brief overview of taurine, the results of taurine studies related to alcoholic. Therefore, it shows how nursing practice strategies can be derived from biomedical research related to taurine and alcoholic.

\textbf{2. Materials and methods}
2.1. Experiment

The 30 alcoholic patients with 2 to 5 times over normal activities of aspartate transaminase (AST) or alanine transaminase (ALT) were selected and divided into two groups, taurine group and control group. In taurine group, patients consisted of 8 men and 7 women and their average age and body weight were 59±13 years old and 62±11 kg, respectively. In control group, patients included each 9 men and 6 women and their average age and body weight were 60±12 years old and 61±10 kg, respectively. This study was carried out in accordance with the Helsinki Declaration of the World Medical Association and was approved by the University’s Ethics Committee. Written informed consent was obtained from each subject after the study was explained to them. All patients promised the treatment and then the experiment was conducted out. In taurine group, each patient took 6 g taurine per day for three months, and then stopped treatment for 1 month. The daily intake of 6 g taurine was divided into three parts and given to each patient of taurine group after meal. In control group, patients took placebo without taurine. Taurine was purchased from Dokui Chemical Company (Taiwan), its purity was 99.5%. The blood samples of all patients were collected per month and analyzed for blood characteristics, including red blood cells (RBC), white blood cells (WBC), hemoglobin (Hb) and platelet, by using a Cell Hematology Analyzer (DYN 500, Sequoi-Turner, USA). The plasma samples were collected by centrifugation (2,000 \times g, 15 min) from blood and examined for the levels of BUN, creatinine, bilirubin, cholesterol and TG and the activities of AST and ALT in the plasma were assayed by a Vitalab Selectra (E. Merck, Germany) with using an enzymatic kit. Serum concentrations of vitamin B\textsubscript{12}, folate and vitamin B\textsubscript{6}\textsuperscript{15}, were measured according to established published methods.
2.2. Determination of TBARS

Lipid peroxidation level was estimated by measuring thiobarbituric acid reactive substances (TBARS) according to Buege and Aust\textsuperscript{16} and was expressed in terms of malondialdehyde (MDA) content which is the end product of lipid peroxidation. In brief, 125 µl of supernatants were homogenized by sonication with 50 µl of TBS, 125 µl of TCA-BHT in order to precipitate proteins and centrifuged (1,000 × g, 10 min, 4°C). 200 µl of obtained supernatant were mixed with 40 µl of HCl (0.6 M) and 160 µl of TBA dissolved in Tris and the mixture was heated at 80°C for 10 min. The absorbance of the resultant supernatant was read at 530 nm. The amount of TBARS was calculated by using an extinction coefficient of 156×10\textsuperscript{5}M\textsuperscript{-1}cm\textsuperscript{-1}.

2.3. Determination of total ADH activity

Total ADH activity was estimated by the photometric method with p-nitrosodimethylaniline (NDMA) as a substrate\textsuperscript{17}. The reaction mixture (2 ml) contained 1.9 ml of a 26 μM solution of substrate in 0.1 M of sodium phosphate buffer, pH 8.5 and 0.1 ml of a mixture containing 0.25 M n-butanol and 5 mM NAD. The reduction of NDMA was monitored at 440 nm.

2.4. Determination of total ALDH activity

ALDH activity was measured using the fluorogenic method based on the oxidation of 6-methoxy-2-naphtaldehyde to the fluorescent 6-methoxy-2-naphtoate\textsuperscript{18}. The reaction mixture contained 60 µl of substrate, 20 µl of 11.4 mM NAD and 2.8 ml of 50 mM of sodium phosphate buffer, pH 8.5. The mixture contained also 50 µl of a 12 mM solution of 4-methylpyrazole as a specific inhibitor of ADH activity. The
fluorescence was read at excitation wavelength 310 and emission wavelength 360 nm.

2.5. Statistical analysis

Statistical analysis for differences among patients in the experimental groups was performed by the 2-way analysis of variance procedure and Duncan’s new multiple range tests. A $P$ value $< 0.05$ was considered statistically significant.

3. Results

After treating with dietary taurine for 3 months, all indicators of blood characteristics in the alcoholic patients was not affected ($P > 0.05$). The data are similar to those in control group. The data of blood characteristics in normal people are as follows: 4.8-10.8 × $10^3$ cells/µl for WBC, 4.2-6.1 × $10^6$ cells/µl for RBC, 14-18 g/dl for Hb and 130-400 × $10^3$ cells/µl for platelet. All data of blood characteristics in chronic alcoholic patients in taurine and control groups were not different from each other. It indicates that the blood characteristics of alcoholic patients are the same as those normal people and dietary taurine supplement can not change them.

The dietary effect of taurine on AST, ALT, and bilirubin of chronic alcoholic patients is shown in Fig. 1. After treating with dietary taurine for 2 months, AST, ALT, and bilirubin of plasma in the chronic alcoholic patients was significantly decreased ($P < 0.05$). It was also found that the AST, ALT, and bilirubin of chronic alcoholic patients in control group are not changed during experimental period.

The dietary effect of taurine on BUN and creatinine of chronic alcoholic patients is shown in Fig. 2. After treating with dietary taurine for 3 months, There was no
significant difference ($P>0.05$) in the concentration on BUN and creatinine in the plasma among various groups.

The dietary effect of taurine on cholesterol and TG level of chronic alcoholic patients is shown in Fig. 3. After treating with dietary taurine for 3 months, cholesterol and TG level of plasma in the chronic alcoholic patients was significantly decreased ($P < 0.05$).

The dietary effect of taurine on ADH and ALDH activities of chronic alcoholic patients is shown in Fig. 4. After treating with dietary taurine for 3 months, ADH and ALDH activities of plasma in the chronic alcoholic patients was significantly increased ($P < 0.05$). After stopping treatment at the fourth month, the ADH and ALDH activity was also elevated. During the experimental period, the ADH and ALDH activities of chronic alcoholic patients in control group was also not changed.

The dietary effect of taurine on TBARS and GSH level of chronic alcoholic patients is shown in Fig. 5. After treating with dietary taurine for 2 months, TBARS and GSH level of plasma in the chronic alcoholic patients was significantly decreased ($P < 0.05$). It indicates that the peroxidation of blood plasma in alcoholic patients could be availably ameliorated by dietary taurine.

The dietary effect of taurine on vitamin B$_{12}$, folate and vitamin B$_6$ of chronic alcoholic patients is shown in Fig. 6. After treating with dietary taurine for 2 months, vitamin B$_{12}$, folate and vitamin B$_6$ of plasma in the chronic alcoholic patients was significantly increased ($P < 0.05$). Since stopping treatment at the fourth month, vitamin B$_{12}$, folate and vitamin B$_6$ of plasma in the chronic alcoholic was not elevated. However, the variation of vitamin B$_{12}$, folate and vitamin B$_6$ of plasma in the chronic
alcoholic patients without taurine supplement was not found during the experimental
period. It indicates that the vitamins of blood plasma in alcoholic patients could be
availably ameliorated by dietary taurine.

4. Discussion

In this study, the blood characteristics, activities of ALT, AST, ADH, and ALDH
and levels of bilirubin, TG, cholesterol and TBARS in the plasma of chronic alcoholic
patients without supplement of taurine during experimental period of 4 months were
not changed. However the chronic alcoholic patients were treated with dietary taurine,
the clinical symptoms including ALT, AST, ADH, and ALDH activities and bilirubin,
TG, cholesterol and TBARS levels were significantly affected. ALT and AST activities
in plasma serve as biomarkers for liver tests\textsuperscript{19}. The value in normal human is as
follows: 10-40 U/l for ALT and 5-45 U/l for AST. The ALT and AST activities in
chronic alcoholic patients were higher than those of normal human. Among them,
ALT activity was higher than AST activity in the chronic alcoholic patients. It means
that alcoholic might injure liver test. Taurine significantly reduced the enzymatic
activities of ALT and AST in the plasma of chronic alcoholic patients, indicating that
the liver injury by alcoholic could be ameliorated by taurine. Wright et al.\textsuperscript{20} pointed
out that the function of taurine for preserving liver cells was presented by the high
content of taurine in cell membrane.

Alcohol is mainly metabolized by ADH in cytosol, CYP2E1 in endoplasmic
reticulum, and CAT in peroxisomes to form acetaldehyde, and further catabolized to
acetic acid by ALDH\textsuperscript{21}. However, during the alcohol metabolism by CYP2E1 the
reactive oxygen species (ROS) also generates and increases the lipid peroxidation, i.e.
MDA in the liver\textsuperscript{22-24}. However, the level of TBARS in the plasma is an additional indicator of liver injury. The level of TBARS in the plasma of alcoholic patients was significantly reduced when the patients were treated with the supplement of taurine. This result is the same as that reported previously\textsuperscript{4,25}. Therefore, it is reasonable to assume that taurine may act as a good scavenger in reducing lipid peroxidation induced by drugs\textsuperscript{26}, heavy metals\textsuperscript{5} and oxidized oil\textsuperscript{4}.

The levels of TG and cholesterol in the plasma of chronic alcoholic patients were not higher than those of normal human (120-200 mg/dl for cholesterol, 35-170 mg/dl for TG). However, the levels of TG and cholesterol in the plasma of alcoholic patients were significantly reduced when the patients were treated with the supplement of taurine. These results are the same as those reported previously\textsuperscript{27-29}. The reduction of TG and cholesterol levels in the plasma may induce the decrease of lipid peroxidation, resulting in inhibiting production of TBARS. Judging from above data taurine plays an important role in the properties of antioxidation and has some improvements on the liver test of chronic alcoholic patients. Hence, taurine is a valuable dietary nutrient for chronic alcoholic patients.

The level of GSH in the liver of rats was reduced by chronic alcoholic patients, which was similar to that of other report\textsuperscript{30,31}. In our previous study, the level of GSH in the liver of rats was raised significantly when the rats were fed with the supplement of taurine\textsuperscript{4}.

Alcohol has been known for a long time to interfere with the absorption of several nutrients, including vitamins, and to lead to mucosal damage of the upper small intestine, thereby contributing to the qualitative and quantitative malnutrition frequently observed in alcoholics\textsuperscript{32,33}. Therefore, taurine is a safe dietary nutrient and an effective hepatoprotective agent to benefit chronic alcoholic patients.
Acknowledgement

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References


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Legends for Figures:

Fig. 1. Effect of taurine on the AST, ALT, and bilirubin of serum plasma in chronic alcoholic patients after 4 months. Data represent significantly different when the letters are different ($p<0.05$). Taurine group: patients took 6 g taurine per day for three months. Control group: patients took placebo without taurine. a-b: values in the same week with different superscript are significantly different at $P<0.05$ by ANOVA ($n=30$).

Fig. 2. Effect of taurine on the BUN and creatinine of serum plasma in chronic alcoholic patients after 4 months. Taurine group: patients took 6 g taurine per day for three months. Control group: patients took placebo without taurine. a: values in the same week with different superscript are significantly different at $P>0.05$ by ANOVA ($n=30$).

Fig. 3. Effect of taurine on the cholesterol and triglyceride of serum plasma in chronic alcoholic patients after 4 months. Taurine group: patients took 6 g taurine per day for three months. Control group: patients took placebo without taurine. a-b: values in the same week with different superscript are significantly different at $P<0.05$ by ANOVA ($n=30$).

Fig. 4. Effect of taurine on the ADH and ALDH of serum plasma in chronic alcoholic patients after 4 months. Taurine group: patients took 6 g taurine per day for three months. Control group: patients took placebo without taurine. a-b: values in the same week with different superscript are significantly different at $P<0.05$ by ANOVA ($n=30$).

Fig. 5. Effect of taurine on the TBARS and GSH of serum plasma in chronic alcoholic patients after 4 months. Taurine group: patients took 6 g taurine per day for three months. Control group: patients took placebo without taurine. a-b: values in the same week with different superscript are significantly different at $P<0.05$ by ANOVA ($n=30$).

Fig. 6. Effect of taurine on the vitamin $\text{B}_{12}$, vitamin $\text{B}_{6}$, and folate of serum plasma in chronic alcoholic patients after 4 months. Taurine group: patients took 6 g
taurine per day for three months. Control group: patients took placebo without taurine. a-b: values in the same week with different superscript are significantly different at $P<0.05$ by ANOVA (n=30).

Fig. 1. Yeh et al.
Fig. 2. Yeh et al.
Fig. 3. Yeh et al.
Fig. 5. Yeh et al.

Control group
Taurine group

GSH (nmol/ml)

TBARS (MDA nmol/ml)

Experimental time (months)
Fig. 6. Yeh et al.