



## Effects of herbal tea (*Platostoma palustre*) on the Hyperlipidemia *in vivo*

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Open Access Research Journal of Life Sciences, 2023, 06(01), 081–089

Publication history: Received on 28 March 2023; revised on 06 August 2023; accepted on 09 August 2023

Article DOI: <https://doi.org/10.53022/oarjls.2023.6.1.0031>

### Abstract

*Platostoma palustre* jelly is a traditional food. *Platostoma palustre* has been used as folk medicine and is effective against heat-shock, hypertension and diabetes. Therefore, the aim of *in vivo* study was to determine the effects of herbal tea (*Platostoma palustre*) on blood lipid regulation. The commercial herbal tea (*Platostoma palustre*) was kindly provided by Yueta Agricultural Biotechnology Inc. Adult male 18 Syrian hamsters (outbred stock) [5 weeks old; body weight (BW) between 90-100 g] with specific pathogen-free conditions were used in this study. In this experiment, all Syrian hamsters (n = 18) were divided respectively the normal control group (n = 6), the negative control group (n = 6), and the herbal tea group (n = 6). The high-fat feed (containing 0.2% cholesterol) was used to feed Syrian hamsters for 8 weeks to induce hyperlipidemia in the negative control group and the herbal tea group. In the herbal tea group, the herbal tea (10 mL/kg BW) was administrated to Syrian hamsters by gavage. Blood were collected before hyperlipidemia was induced (D0) and blood was collected after hyperlipidemia was induced (D28 and D56). The BW of Syrian hamsters were weighed weekly. The TG (triglyceride), TCHO (total cholesterol), HDL (high-density lipoprotein), and LDL (low-density lipoprotein) contents in blood were detected and analyzed at each experimental time point. In addition, at the end of the experiment, the liver tissue was dissected out for analysis of CHO (cholesterol) and TG contents. The results were shown that the average BW of the Syrian hamsters in the negative control group was significantly higher than that of the Syrian hamsters in the normal control group during hyperlipidemia induction (W5-W8). The BW of the herbal tea group was slightly higher than that of the normal control group after hyperlipidemia induction. However, there was no significant difference between the negative group and the herbal tea group each week of the experiment. The TG level of Syrian hamsters in the negative control group was significantly higher than that of Syrian hamsters in the normal control group at the 8<sup>th</sup> weeks-experiment. The TG level of Syrian hamsters in the herbal tea group was between the negative control group and the normal control group and there were no significant differences between two groups (the herbal tea group and the negative control group). The TCHO levels in blood of Syrian hamsters in the negative control group and the herbal tea group were both significantly higher than that of the normal control group at the 4<sup>th</sup> week- and 8<sup>th</sup> week-experiment. At the 4<sup>th</sup> and 8<sup>th</sup> weeks-experiment, the TCHO of the herbal tea group was slightly lower than that of the negative control group after hyperlipidemia induction. The TCHO levels in blood of Syrian hamsters in the herbal tea group and the negative control group had no significant difference. At the experiment (W4 and W8), the HDL cholesterol level in blood of Syrian hamsters in the negative control and the herbal tea group were significantly higher

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than that in the normal control group. The HDL cholesterol level in blood of the herbal tea group was slightly higher than that of the negative control group after hyperlipidemia induction (W4 and W8). At the experiment (W4 and W8), the HDL cholesterol level in blood of the negative control group and the herbal tea group were significantly higher than that of the normal control group. There was no significant difference between the herbal tea group and the negative control group. Additionally, the LDL cholesterol level in blood of the herbal tea group was significantly lower than that of the negative control group after hyperlipidemia induction (W4 and W8). At the experiment (W4 and W8), the ratio of HDL cholesterol level /LDL cholesterol level in blood of the negative control group were significantly lower than that of the normal control group. The ratio of HDL cholesterol level /LDL cholesterol level in blood of the herbal tea group was higher than that of the negative control group after hyperlipidemia induction (W4 and W8). However, there was no significant difference between the herbal tea group and the normal control group. At the end of experiment (W8), the TG and CHO levels in liver tissues of the negative control group and the herbal tea group were significantly higher than that of the normal control group. The TG and CHO contents in liver tissues of the herbal tea group was lower than that of the negative control group at the end of experiment (W8). However, there was no significant difference between the herbal tea group and the negative control group. Taken all *in vivo* results together, the hyperlipidemia was successfully induced in the experimental Syrian hamsters. After administrating with the herbal tea, the blood and liver lipid levels of the Syrian hamsters tended to improve. Therefore, based on the results of this experiment, it is speculated that drinking the herbal tea for 2 months has considerable potential for blood lipid regulation, which can be used as the basis for the development of related products of the herbal tea in the future.

**Keywords:** Blood lipid; Herbal tea; *In vivo*; *Platostoma palustre*; Regulation

## 1 Introduction

According to WHO global estimates, about 2 billion adults aged 18 years and older were overweight in 2016. Of these over 650 million adults were obese. Overall, about 13% of the world's adult population were obese in 2016. The worldwide prevalence of obesity nearly tripled between 1975 and 2016. An estimated 38.2 million children under the age of 5 years were overweight or obese in 2019. Obesity is linked to more deaths worldwide than underweight. It is the excessive accumulation of body fat and it is a consequence of persistent energy intake that exceeds energy expenditure [1-3]. Obesity is defined as abnormal or excessive fat accumulation that may impair health. The fundamental cause of obesity is an energy imbalance between calories consumed and calories expended. Globally, there has been an increased intake of energy-dense foods that are high in fat and sugars and an increase in physical inactivity due to the increasingly sedentary nature of many forms of work, changing modes of transportation, and increasing urbanization. Changes in dietary and physical activity patterns are often the result of environmental and societal changes associated with development and lack of supportive policies in sectors such as health, agriculture, transport, urban planning, environment, food processing, distribution, marketing, and education [4-5].

Hyperlipidemia is a disease with high fat content in the blood. It is mainly related to cholesterol and triglycerides exceeding the normal value. It is also the cause of other major diseases such as heart disease, hypertension, stroke, diabetes, arteriosclerosis, and even kidney disease. Most people with hyperlipidemia have no obvious symptoms at the beginning, and a small number of patients with hyperlipidemia inherited from family. Therefore, if we want to detect it early, we must rely on regular health checks to detect the concentration of cholesterol and triglycerides in the blood. In the recent years, obesity is defined as abnormal or excessive fat accumulation that may impair health. More and more researches on the prevention and treatment of obesity are prevalent. Understanding of the fat cellular molecule mechanisms of obesity was one of the major focuses in obesity field. Thus, it is very important to research and develop high-value crops to reduce blood lipid levels [1-6].

The related food with *Platostoma palustre* as tea, herbal jelly, and sweet soup with herbal jelly are popular during the summer. Additionally, the heated herbal jelly with *Platostoma palustre* is admired by many Taiwanese in winter. *Platostoma palustre* has been used as folk medicine. *Platostoma palustre* has been verified that possessed many functional compounds [7-9]. Therefore, these functional compounds of *Platostoma palustre* have indicated that many biological effects is effective against and attenuating the metabolic syndrome, heat-shock, hypertension, diabetes, liver disease, muscle and/or joint pains, hyperglycemia, inflammation, oxidant activity, free radical scavenging effects, acute and chronic hepatitis, and cancer growth [10-25]. Therefore, the objective of this study was to evaluate the *in vivo* effects of the commercial herbal tea (*Platostoma palustre*) on the regulation of blood lipids.

## 2 Material and methods

### 2.1 Chemicals and Reagents

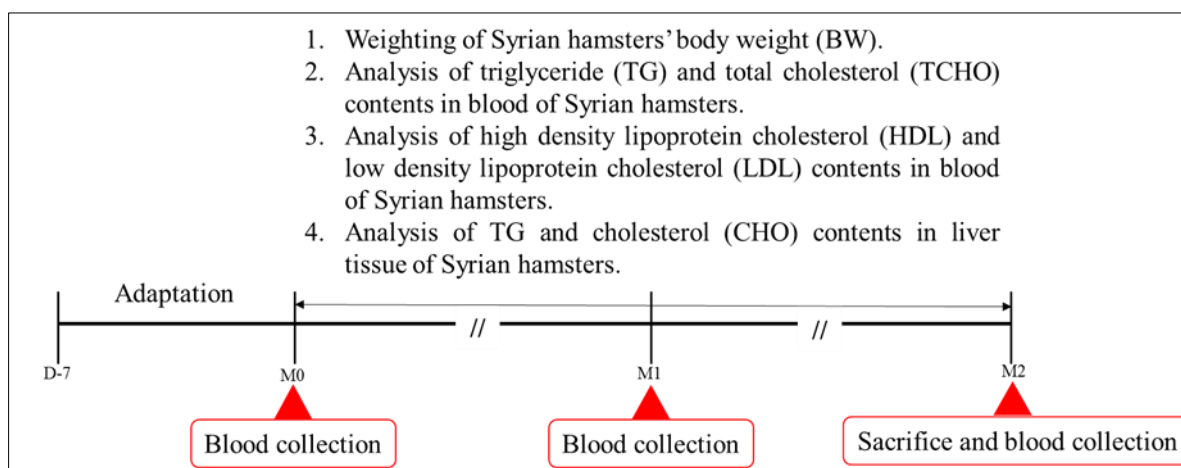
Phosphate-buffered saline (PBS; Sigma-Aldrich, Cat. No. P3813), saline (Taiwan Biotech Co., LTD, Cat. No. 100-120-1101), and Zoletil 50 (Virbac, Carros, France) were applied in this experiment.

### 2.2 Source of Herbal Tea

The herbal tea (*Platostoma palustre*) were kindly provided by Yueta Agricultural Biotechnology Inc. (Guanxi, Hsinchu, Taiwan). Yueta® herbal tea has been passed the SGS pesticide test, and is cooked through high-temperature cooking. The operation process of machinery and equipment is consistent, and it is sterilized by high-temperature sterilizing kettle, without adding preservatives.

### 2.3 Experimental Animals and Experimental Design

Adult male 18 Syrian hamsters (outbred stock) [5 weeks old; body weight (BW) between 90-100 g] with specific pathogen-free conditions were used for this study, were purchased from BioLASCO Taiwan Co., Ltd. (Yilan, Taiwan). Before the experiment, all mice were housed in the animal room for 7 days. The environment was maintained room temperature (24-27°C) and 60%-70% humidity with a photoperiod of 12-hr light/12-hr dark cycle. The study will begin after a week acclimation. The Institutional Animal Care and Use Committee (IACUC) of Agricultural Technology Research Institute inspected all animal experiments and this study comply with the guidelines of protocol IACUC 110111C1 approved by the IACUC ethics committee. The male 18 Syrian hamster were divided respectively the normal control group (n = 6), the negative control group (n = 6), and the herbal tea group (n = 6). In the negative control group and the herbal tea group, Syrian hamsters were fed with the high fat feed [Diet Induced Obesity (DIO) Series diets D12451, Research Diets Inc., NJ, USA] (containing 0.2% cholesterol) to induce hyperlipidemia. However, the Syrian hamsters were fed with the standard laboratory diet (No. 5053, LabDiet®; PMI Nutrition International, St. Louis, MO, USA) ad libitum in the normal group during the experimental period. In the herbal tea group, the herbal tea (10 mL/kg BW) was administrated to Syrian hamsters by gavage. The weighting of Syrian hamsters' BW, the analysis of triglyceride (TG) and total cholesterol (TCHO) contents in blood of Syrian hamsters, the analysis of high density lipoprotein (HDL) cholesterol and low density lipoprotein (LDL) cholesterol contents in blood of Syrian hamsters, and the analysis of TG and cholesterol (CHO) contents in liver tissue of Syrian hamsters were detected during the experiment (Fig. 1).



**Figure 1** Experimental designs and the weighting of Syrian hamsters' body weight (BW), the analysis of triglyceride (TG) and total cholesterol (TCHO) contents in blood of Syrian hamsters, the analysis of high density lipoprotein (HDL) cholesterol and low density lipoprotein (LDL) cholesterol contents in blood of Syrian hamsters, and the analysis of TG and cholesterol (CHO) contents in liver tissue of Syrian hamsters were detected during the experiment

### 2.4 The Weighting of Syrian Hamsters' BW

To monitor the BW of Syrian hamsters every week until the end of the experiment.

## 2.5 The Analysis of Contents of TG, TCHO, HDL Cholesterol, and LDL Cholesterol in Blood of Syrian Hamsters

Syrian hamsters were collected blood and then collected sera via 4°C, 1,500 rpm for 10 minutes. The sera were analyzed the levels of TG, TCHO, HDL cholesterol, and LDL cholesterol by using Fuji DRI-CHEM NX500i (Fuji, Japan) according to the manufacturer's protocols.

## 2.6 The Analysis of TG and CHO Contents in Liver Tissues of Syrian Hamsters

Syrian hamsters were collected liver tissues and then storage at -80°C. Later, the liver tissues were analyzed the levels of TG and CHO by using cholesterol assay kit (Cat. No.: ab65390, abcam) and triglyceride colorimetric assay kit (Cat. No.: 10010303, Cayman Chemical) according to the manufacturer's protocols.

## 2.7 Statistical Analysis

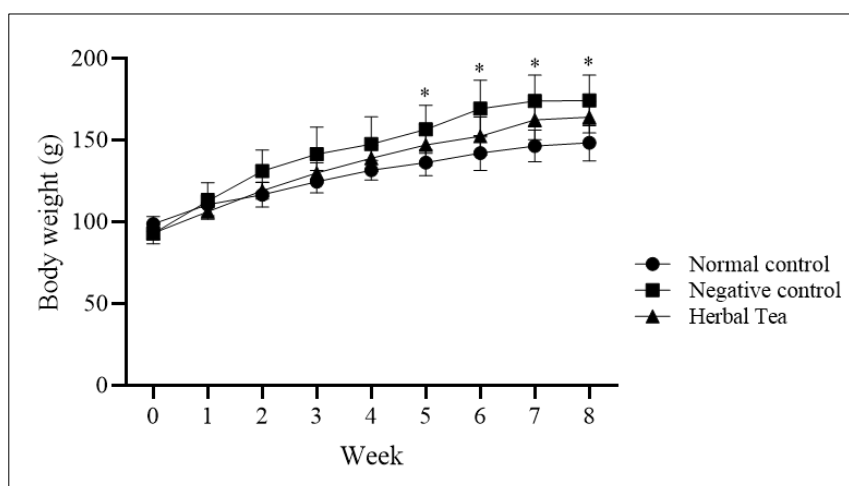
The data were expressed as mean  $\pm$  SD (standard deviation). All comparisons were made by one-way ANOVA and all significant differences are reported at  $*/\#/\&p < 0.05$ ,  $**/\##p < 0.01$ , and  $***/\###p < 0.001$ .

## 3 Results

In this experiment, the high-fat feed (containing 0.2% cholesterol) was used to feed Syrian hamsters for 8 weeks to induce hyperlipidemia in the negative control group and herbal tea group mice. After the Syrian hamsters were induced with hyperlipidemia, the Syrian hamsters in the negative control group were given drinking water by gavage every day, and the Syrian hamsters in the herbal tea group were given the herbal tea (administration dose 10 mL/kg BW) by gavage every day. In the experiment, blood were collected before hyperlipidemia was induced (D0) and blood was collected after hyperlipidemia was induced (D28 and D56). The BW of Syrian hamsters were weighed weekly. The TG, TCHO, HDL cholesterol, and LDL cholesterol contents in blood were detected and analyzed at each experimental time point. In addition, at the end of the experiment, the liver tissues were dissected out for analysis of CHO and TG contents.

### 3.1 BW Change of Syrian Hamsters in Each Group during the Experiment

The results were shown that the average BW of the Syrian hamsters in the negative control group (Negative control) was significantly higher than that of the Syrian hamsters in the normal control group (Normal control) during hyperlipidemia induction (W5-W8). The BW of the herbal tea group was slightly higher than that of the normal control group after hyperlipidemia induction. However, there was no significant difference between the negative group and the herbal tea group each week of the experiment (Fig. 2).

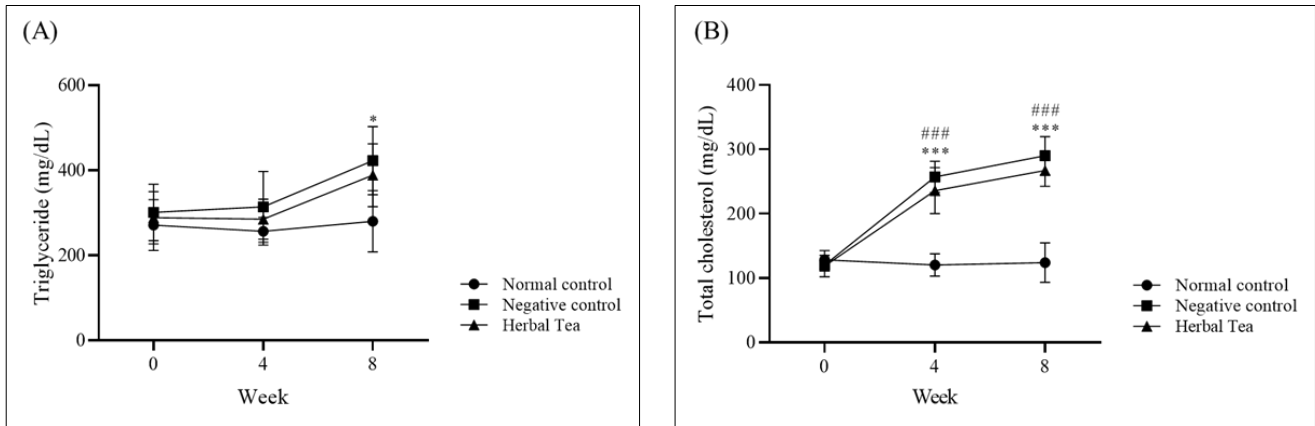


**Figure 2** BW change of Syrian hamsters in each group during the experiment. Data are presented as Mean  $\pm$  SD. \* indicates the normal control group vs. the negative control group.  $*p < 0.05$ . BW: body weight

### 3.2 Detection of the Levels of TG and TCHO in Blood of Syrian Hamsters

To compare the TG level and TCHO level at each experimental point in the normal control group, the negative control group, and the herbal tea group. The results were shown that (1) the level of TG: the TG level of Syrian hamsters in the negative control group was significantly higher than that of Syrian hamsters in the normal control group at the 8<sup>th</sup>

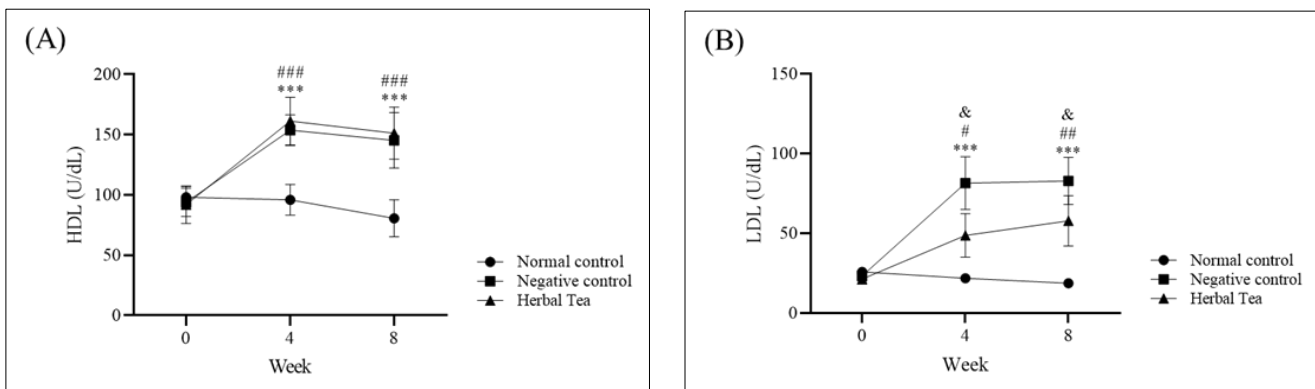
weeks-experiment. The TG level of Syrian hamsters in the herbal tea group was between the negative control group and the normal control group and there were no significant differences between two groups (the herbal tea group and the negative control group) (Fig. 3A). (2) the level of TCHO: the TCHO levels in blood of Syrian hamsters in the negative control group and the herbal tea group were both significantly higher than that of the normal control group at the 4<sup>th</sup> week- and 8<sup>th</sup> week-experiment. At the 4<sup>th</sup> and 8<sup>th</sup> weeks-experiment, the TCHO of the herbal tea group was slightly lower than that of the negative control group after hyperlipidemia induction. The TCHO levels in blood of Syrian hamsters in the herbal tea group and the negative control group had no significant difference (Fig. 3B).

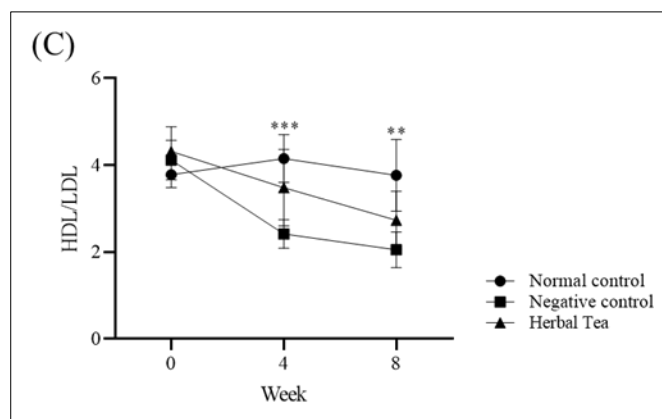


**Figure 3** Detection of the levels of TG and TCHO in blood in Syrian hamsters. (A) The level of TG at each experimental point. (B) The level of TCHO at each experimental point. Data were presented as mean  $\pm$  SD. \* indicates the normal control group vs. the negative control group. # indicates the normal control group vs. the herbal tea group. \* $p < 0.05$ ; \*\*\*/### $p < 0.001$ . TG: triglyceride; TCHO: total cholesterol

### 3.3 Analysis of HDL and LDL Cholesterol Contents in Blood of Syrian Hamsters

The results of HDL and LDL cholesterol contents at each experimental point were compared among the normal control group, the negative control group and the herbal tea group. It can be seen from the results that at the experiment (W0), there was no significant difference in HDL cholesterol level at each experimental point in each group. At the experiment (W4 and W8), the HDL cholesterol level in blood of Syrian hamsters in the negative control and the herbal tea group were significantly higher than that in the normal control group. The HDL cholesterol level in blood of the herbal tea group was slightly higher than that of the negative control group after hyperlipidemia induction (W4 and W8). At the experiment (W4 and W8), the HDL cholesterol level in blood of the negative control group and the herbal tea group were significantly higher than that of the normal control group. There was no significant difference between the herbal tea group and the negative control group (Fig. 4A). Additionally, the LDL cholesterol level in blood of the herbal tea group was significantly lower than that of the negative control group after hyperlipidemia induction (W4 and W8) (Fig. 4B). At the experiment (W4 and W8), the ratio of HDL cholesterol level /LDL cholesterol level in blood of the negative control group were significantly lower than that of the normal control group. The ratio of HDL cholesterol level /LDL cholesterol level in blood of the herbal tea group was higher than that of the negative control group after hyperlipidemia induction (W4 and W8). However, there was no significant difference between the herbal tea group and the normal control group (Fig. 4C).

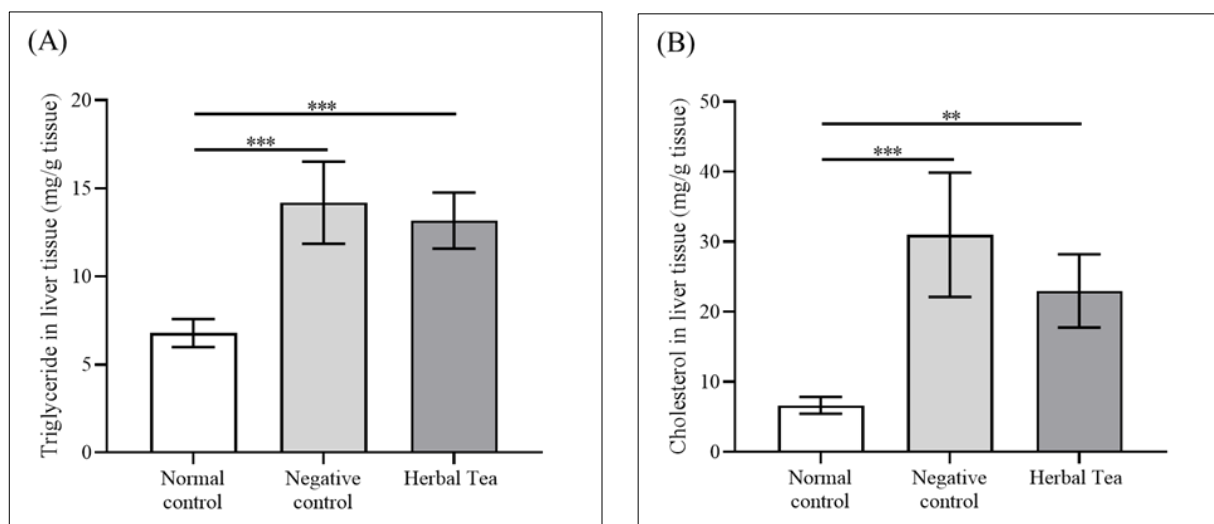




**Figure 4** Analysis of HDL cholesterol, LDL cholesterol, and the ratio of HDL/LDL cholesterol in blood of Syrian hamsters. (A) Changes in blood HDL cholesterol at each time point, (B) Changes in blood LDL cholesterol at each time point, (C) Changes in the ratio of blood HDL cholesterol / LDL cholesterol at each time point (HDL/LDL). Data are presented as Mean  $\pm$  SD. \* indicates normal control group vs. negative control group, # indicates normal control group vs. herbal tea group, & indicates negative control group vs. herbal tea group. #/&p < 0.05; \*\*/##p < 0.01; \*\*\*/###p < 0.001. HDL: high density lipoprotein; LDL: low density lipoprotein

### 3.4 Analysis of TG and CHO contents in liver tissues in Syrian hamsters

At the end of experiment (W8), the TG and CHO levels in liver tissues of the negative control group and the herbal tea group were significantly higher than that of the normal control group. The TG and CHO contents in liver tissues of the herbal tea group was lower than that of the negative control group at the end of experiment (W8). However, there was no significant difference between the herbal tea group and the negative control group (Fig. 5).



**Figure 5** Analysis of TG and CHO contents in liver tissues of Syrian hamsters. (A) TG content in liver tissue of each group, (B) CHO content in liver tissue of each group. Data are presented as Mean  $\pm$  SD. \*\*p < 0.01; \*\*\*p < 0.001. TG: triglyceride; CHO: cholesterol

## 4 Discussion

Blood lipids refer to the content of adipose elements such as free fatty acids, phospholipids, sterols, and triglycerides in plasma. The healthy value of blood lipid content in plasma is generally below 130 mg/dL. Higher than 160 mg/dL is called excessive blood fat in medicine, that is also called hyperlipidemia. Hyperlipidemia, hypertension, and hyperglycemia are together called the "three highs", and are known as the "invisible killer" of health. They are one of the main diseases that require health management of chronic diseases [26-28].

With the improvement of living levels, there are more and more opportunities for exquisite diet. Our blood lipid value is also getting higher and higher, and there is a trend of high blood lipid value for younger people. However, many people think that this is not a disease, so they usually ignore it. If hyperlipidemia is not controlled in time, it will cause complications, and these complications are usually very serious [29-30].

Hyperlipidemia may cause many complicated disease as coronary heart disease (CHD), cerebral infarction, diabetes, fatty liver etc. The CHD is coronary atherosclerotic heart disease, and once excessive blood lipids block the coronary arteries, causing arteriosclerosis, myocardial ischemia and hypoxia. Hyperlipidemia is one of the most dangerous factors causing CHD. It was verified that an average 1% decrease of total cholesterol in serum will reduce 2% incidence of CHD. Therefore, regulating blood lipids is the most basic treatment for preventing and treating CHD. In addition, the excessive cholesterol in the blood can cause atherosclerotic plaques, resulting in narrowing and blockage of the arterial lumen. When this situation occurs in the cerebral blood vessels, it will cause cerebral infarction. A number of studies have proved that the incidence and disability rates of cerebral apoplexy are significantly related to the effect of long-term lipid-lowering treatment. Among the factors that cause stroke, such as drinking, obesity, high blood pressure, smoking, diabetes and so on. Hyperlipidemia is one of the most important influencing factors [26-32].

Hypertension, hyperlipidemia, and hyperglycemia are collectively referred to as the "three highs", which pose a serious threat to the life and health of diabetic patients. Hyperlipidemia will aggravate the patient's condition. Therefore, clinically, while treating hyperglycemia, a certain amount of blood lipid regulation will also be carried out. Moreover, hyperlipidemia is an important cause of late complications of diabetes, such as CHD, fundus necrosis, kidney disease, neuropathy, etc. Active treatment of hyperlipidemia can effectively prevent the occurrence of these complications. Moreover, the appearance of fatty liver is mainly caused by the accumulation of fat in the liver, and patients often have hyperlipidemia. Hyperlipidemia, long-term heavy drinking, obesity, diabetes, abdominal fat accumulation, and patients with viral hepatitis are all high-risk groups for fatty liver. Therefore, regulating blood lipids in patients with hyperlipidemia is an important measure to prevent fatty liver.

In this study, the results were shown that the BW of the herbal tea group was slightly higher than that of the normal control group after hyperlipidemia induction. However, there was no significant difference between the negative group and the herbal tea group each week of the experiment. The TG level of Syrian hamsters in the herbal tea group was between the negative control group and the normal control group and there were no significant differences between two groups (the herbal tea group and the negative control group). The TCHO levels in blood of Syrian hamsters in the negative control group and the herbal tea group were both significantly higher than that of the normal control group at the 4<sup>th</sup> week- and 8<sup>th</sup> week-experiment. At the 4<sup>th</sup> and 8<sup>th</sup> weeks-experiment, the TCHO of the herbal tea group was slightly lower than that of the negative control group after hyperlipidemia induction. The TCHO levels in blood of Syrian hamsters in the herbal tea group and the negative control group had no significant difference. At the experiment (W4 and W8), the HDL cholesterol level in blood of Syrian hamsters in the negative control and the herbal tea group were significantly higher than that in the normal control group. The HDL cholesterol level in blood of the herbal tea group was slightly higher than that of the negative control group after hyperlipidemia induction (W4 and W8). At the experiment (W4 and W8), the HDL cholesterol level in blood of the negative control group and the herbal tea group were significantly higher than that of the normal control group. There was no significant difference between the herbal tea group and the negative control group. Additionally, the LDL cholesterol level in blood of the herbal tea group was significantly lower than that of the negative control group after hyperlipidemia induction (W4 and W8). At the experiment (W4 and W8), the ratio of HDL cholesterol level /LDL cholesterol level in blood of the negative control group were significantly lower than that of the normal control group. The ratio of HDL cholesterol level /LDL cholesterol level in blood of the herbal tea group was higher than that of the negative control group after hyperlipidemia induction (W4 and W8). However, there was no significant difference between the herbal tea group and the normal control group. At the end of experiment (W8), the TG and CHO levels in liver tissues of the negative control group and the herbal tea group were significantly higher than that of the normal control group. The TG and CHO contents in liver tissues of the herbal tea group was lower than that of the negative control group at the end of experiment (W8). However, there was no significant difference between the herbal tea group and the negative control group. Taken all results together, after administrating with the herbal tea, the blood and liver lipid levels of the Syrian hamsters tended to improve.

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## 5 Conclusion

The commercial herbal tea (*Platostoma palustre*) was kindly provided by Yueta Agricultural Biotechnology Inc. Taken all *in vivo* results together, the hyperlipidemia was successfully induced in the experimental Syrian hamsters. After administrating with the herbal tea, the lipid levels in blood and liver tissues of the Syrian hamsters tended to improve. Therefore, based on the results of this experiment, it is speculated that drinking the herbal tea for 2 months has

considerable potential for blood lipid regulation, which can be used as the basis for the development of related products of the herbal tea in the future.

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## Compliance with ethical standards

### Acknowledgments

All authors thank Ministry of Agriculture [grant number 111AS-11.3.2-ST-a2], Yueta Agricultural Biotechnology Inc. [contract number AAI-09-E-P11105], and National Science and Technology Council (grant number MOST 109-2314-B-866 -001 -MY3 and NSTC 111-2622-B-866-003) for supporting this study.

### Disclosure of conflict of interest

The authors declare no conflict of interest.

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