International Journal of Herbs, Spices and Medicinal Plants
Vol. 2(1), pp. 018-024, June, 2017. © www.premierpublishers.org, ISSN: 2123-7362

IJHSMP

Research article

Pharmacodynamic study of Jerusalem artichoke particles in type I and II diabetic rat models

Bo Zhao[1], Linfeng Li[2], Feng Lv[3], Zhuang Li[4], Lanlan Zhang[5], Yujia Zhai[6], Guowei Zhang[7*]

[1,2,3,4,5,6,7] College of Chinese Medicine, Hebei University, Baoding, 071002, Hebei province, China.

To study the therapeutic effect of Jerusalem artichoke particles in type I and type II diabetic rats. Male Sprague–Dawley (SD) rats were intraperitoneally injected with 30 mg/kg streptozotocin (STZ) for 3 consecutive days to generate a type I diabetic rat model. The rats were orally administered Jerusalem artichoke particles (50, 100, or 150 mg/kg) once a day for 3 consecutive weeks. Fasting blood glucose levels were determined by ELISA. Male SD rats were fed a high-fat and high-sugar diet then received an intraperitoneal injection of 35 mg/kg STZ to generate a type II diabetic rat model. The rats were treated as mentioned above for 4 consecutive weeks. Fasting blood glucose levels were determined using the glucose oxidase method. Jerusalem artichoke particles significantly reduced blood glucose concentrations in type I and type II diabetic rats. Following 50, 100 and 150 mg/kg Jerusalem artichoke particles treatment for specified weeks, blood glucose concentrations were decreased by 9.7%, 21.69% and 15.48% in type I diabetic rats, respectively; and type II diabetic rats were decreased by 12.07%, 28.57% and 21.80%, respectively. Jerusalem artichoke particles have a hypoglycemic effect in type I and type II diabetic rats.

Keywords: Jerusalem artichoke, Inulin, Type I diabetes mellitus, Type II diabetes mellitus, Blood glucose

INTRODUCTION

Diabetes mellitus (DM) is an endocrine disease characterized by persistent hyperglycemia due to the absolute or relative lack of insulin secretion. It is often accompanied by lipid metabolic disorder and hyperlipidemia (Andreassen O A et al., 2002). Thus, reducing blood sugar and lipid levels could be the key to alleviate diabetes symptoms (Giugliano D et al., 2016). According to statistics, approximately 10% of the elderly population suffers from non-insulin-dependent diabetes mellitus. Such patients can only control symptoms by changing their dietary habits, which breaks the current situation of frequent insulin injections (Chen ZC et al., 1995). At the same time as drug treatment, some of the new resources of food and health food auxiliary role will help patients to a certain degree of balanced nutrition, maintain weight, and reducing the various symptoms of diabetes and the resulting discomfort (Cui WX, 2008).

Jerusalem artichoke (*Helianthus tuberous L.*) is a perennial herb belonging to the Asteraceae family (Han L et al., 2014). Previous studies with the Jerusalem artichoke have used Illumina RNA sequencing (Jung W Y et al., 2014) and observed attenuated lipid disturbances and insulin resistance (Wan-Ching Chang et al., 2014) and a hypoglycemic effect (Li JH et al., 2015). It has been previously reported that 78% of the carbohydrates in Jerusalem artichokes is inulin (Wang WL et al., 2007). Inulin is a biological polysaccharide composed of D-fructose by a β (1→2) glycosidic linker, and the end contains a glucose base. The average degree of polymerization is 10 (Fan S et al., 2010). Inulin is a water-soluble dietary fiber that can control blood lipids, reduce cardiovascular disease risk (Byungsung P, 2011), and be used as a lipid substitute in food production (Menegas L Z et al., 2013). Previous research has shown that it can effectively reduce blood sugar, thereby improving diabetes (Wang WL et al., 2007).

***Corresponding author**: Guowei Zhang, Department of Chinese Medicine, Hebei University, China No.342 Yuhua Road, Baoding, China. Tel.: +86 13663125198. Email: xxzgw@126.com

Table 1: Factors and levels of L_9 (3^4) orthogonal design.

Level	Factor		
	Distillation time/min	Distillation times/times	Water quantity/times
1	50	1	15
2	60	2	17
3	70	3	20

This article through the Jerusalem artichoke drying section, reflux extraction, and spray drying process, obtained Jerusalem artichoke particles with inulin as the main ingredient, was proved in the streptozotocin (STZ) model of type I and type II diabetic rats (Ren J et al., 1997). We observed the pharmacodynamic effects of Jerusalem artichoke particles on type I and type II diabetic rats. Ultimately, Jerusalem artichoke particles may be able to enter the market as a food or medicine to benefit patients with diabetes.

MATERIALS AND METHODS

Medicinal plant

The Jerusalem artichoke plant was derived from Hebei Baoding planting base and was identified as authentic by Xianmao Liang (senior experimental, Chinese medicine identification teaching and research section of Hebei University).

The optimization process of Jerusalem artichoke particles

The extraction temperature, time and amount of water were selected as variables. We selected three levels for each factor (Table 1). The inulin content was used as the selection standard using an L_9 (3^4) orthogonal table.
Experiments were performed according to the orthogonal table, complete reflux extraction and water bath concentrated into extract for use.

Determination of polysaccharides in Jerusalem artichokes

Inulin content was determined by the anthrone-sulfuric acid method. The polysaccharide content (mg/g) of each sample was calculated by the following formula:

$$\text{polysaccharide content} = (C * D) / (W * E) \quad (1)$$

in which:
C is the concentration of the test solution polysaccharides (g/L),
D is the dilution factor test solution,
E is the weighing polysaccharides extraction of total polysaccharides samples accounted for the sample proportion factor,
W is the quality of the sample (g).

Jerusalem artichoke particle preparation

Concentrated inulin extract was prepared according to the optimum extraction process. The concentration of purified water extract of inulin was dissolved in a certain amount, the use of SY-6000 mini spray drying apparatus for spray drying. Inulin content was determined with the sulfuric acid-anthrone method.

Experimental animals

Male Sprague–Dawley (SD) rats were purchased from the experimental animal center of Hebei Medical University (Beijing, China). The license number is SCXK (Hebei) 2013-1-003. Animals were used after 7 d acclimation. All animals were maintained under standard environment conditions (23 ± 2°C, 55 ± 5% humidity and 12-h/12-h light/dark cycle). All animals were allowed free access to tap water and standard laboratory rat food. Animal studies were approved by the Institutional Animal Ethical Committee (IAEC) of Hebei University.

Animal experiments investigating type I diabetes

After 7 d acclimation, 50 male rats were intraperitoneally injected with STZ (30 mg/kg) dissolved in 0.1 mol/L citrate buffer (pH=4.5) for 3 days after fasting for 12 h. Seven days later following a 12-h fast, blood samples were obtained from the retro-orbital venous plexus with 0.9-mm–1.1-mm capillary tubes. Blood glucose was measured by rat blood glucose ELISA. Rats with fasting blood glucose levels above 7.8 mmol/L and obvious polydipsia, polyphagia and polyuria were selected for the type I diabetic rat model (Zhang YY et al., 2011).

The rats were randomly divided into 6 groups: blank, model, low Jerusalem artichoke particles dose, intermediate Jerusalem artichoke particles dose, high Jerusalem artichoke particles dose and positive control groups. The blank and model groups were administered saline solution. The low-, intermediate- and high-dose groups were administered 50, 100 and 150 mg/kg Jerusalem artichoke particle, respectively. The positive control group was administered metformin hydrochloride (150 mg/kg). The administration period was 3 weeks, and the gavage concentration was 100 g/ml. The dose was selected based on previous studies (Xiao Xia et al., 2014). One hour after the final administration following a 12-h fast, we obtained blood from the inner canthus vein, isolated

Table 2: Results of inulin content determination

Test number	Factor				Inulin content mg/g
	A	B	C	D	
1	1	1	1	1	28.0078
2	1	2	2	2	46.1700
3	1	3	3	3	47.9401
4	2	1	2	3	47.7143
5	2	2	3	1	49.4662
6	2	3	1	2	51.0062
7	3	1	3	2	64.3259
8	3	2	1	3	46.6229
9	3	3	2	1	47.6179
K_1	40.7060	46.6827	41.8790		
K_2	49.3956	47.4197	47.1674		
K_3	52.8556	48.8547	53.9107		
R_j	12.1496	2.1720	12.1312		

serum and measured fasting blood glucose levels.

Determination of blood glucose in rats with type I diabetes

The rats were fasted for 12 h, and blood was collected from a capillary from rat eye angular vein blood (approximately 1 mL in a 1.5-mL centrifuge tube). The samples were centrifuged at 3000 rpm for 15 min. The serum was isolated, and glucose concentrations were measured by glucose ELISA. The serum was added to the ELISA kit, and glucose concentrations were determined by an enzyme scale.

Animal experiments investigating type II diabetes

After 7 d acclimation, rats were fed a high-fat diet. After 4 weeks of feeding, rats were fasted for 12 h and intraperitoneally injected with 35 mg/kg STZ (dissolved in 0.1 mol/L citric acid buffer, pH 4.5). Fed with high fat diet for 1 week after fasting for 12 h, using a glass capillary tube from the rat medial venous blood of 1 ml rats were fasting blood glucose by glucose oxidase method FBG = 7.8mmol/L as type II diabetic rats model was successfully (Lu GB et al., 2011).

The rats were randomly divided into six groups and treated as described above. The positive control group by double positive. Most positive control rats were treated with glibenclamide (1 mg/kg), while a minority were treated with Jin Li Da particles (3 g/kg, a traditional Chinese Medicine) administered continuously for 4 weeks. The last day, after administration of 1h in fasting for 12h, we obtained blood from the inner canthus vein, isolated serum and measured fasting blood glucose levels. The serum was added to the glucose reagent kit, and concentrations were determined by UV spectrophotometer.

Determination of blood glucose levels in type II diabetic rats

Rats were fasted for 12 h. We then used a capillary tube to obtain blood from the inner canthus vein (approximately 1 ml in a 1.5-mL centrifuge tube). Samples were centrifuged at 3000 rpm for 15 min, serum was isolated, and blood glucose levels were determined using the glucose oxidase method.

Statistical analysis

The results are expressed as means ± S.D. Statistical significance was evaluated by one-way analysis of variance (SPSS11.5). A value of $p<0.05$ was considered statistically significant.

RESULTS

Optimum process of Jerusalem artichoke particles

Inulin content using the anthrone-sulfuric acid method was determined using the following equation:

$$O.D.=12.454c+0.072,\ R^2=99.60\%, \quad (2)$$

Absorbances were then measured in nine groups of samples, inulin content was obtained according to the standard curve, and determine the final extraction process (Table 2). The orthogonal test results revealed that the order of magnitude for each factor was A>C>B, it has little effect on the B factor, and B_2 B_3 had little difference, considering the actual extraction situation of the level of B_2, we selected the optimal level. Thus, the optimal scheme for the determination of Jerusalem artichoke was $A_3B_2C_3$, namely, extracting two times, 70 min each time, and 20 times water. Finally, the inulin content of Jerusalem artichoke particles was determined to be 128 mg/g, and

the granulometry was 12.5 μm.

Establishment of the type I diabetic rat model

A total of 39 rats were successfully established. DM rats were randomly grouped by weight. A total of six groups with six rats in each group were established. The first part of each bedding was changed every day, diet doubled, after the administration the symptoms of diabetes mellitus began to slowly reduce. At the beginning of the third week, each group began to display different degrees of tail rot, foot rot and rotten mouth, with the exception of the blank group.

Body weight measurements: The rats in the blank group displayed stable growth. Rat weights decreased significantly, while the weight of the low Jerusalem artichoke particles dose group after the success of a slight increase in weight growth trend. Following administration, every group displayed an obvious growth trend that was statistically significant (P<0.05). On the 18th day of administration, the experimental and positive control group weights were significantly higher than that of the model group (P<0.05). Before and after modeling, the rats body weight and body weight of rats were decreased, and the weight of the rats after treatment increased to some extent. Thus, treatment may have mitigated diabetes symptoms, but this requires further evidence (Figure 1).

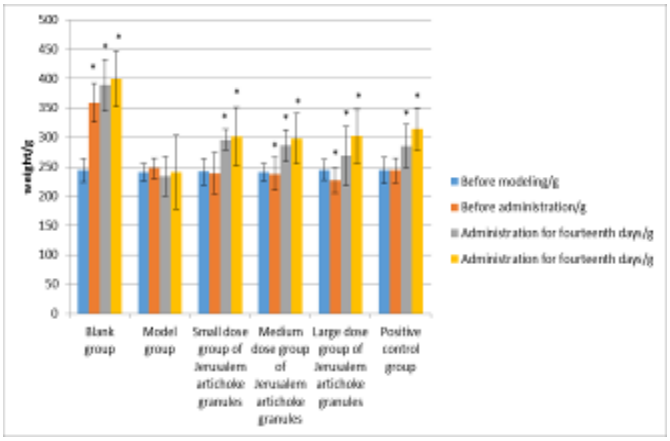

Figure 1: Effects of Jerusalem artichoke particles on body weight in type I diabetic rats

Changes in blood glucose levels in type I diabetic rats

The standard curves of the blood glucose measurements before and after administration were:

$O.D.=0.259c+0.139$, $R^2=99.5\%$ (3)

$O.D.=0.184c+0.074$, $R^2=99.6\%$ (4)

The corresponding blood glucose concentrations were calculated according to the standard curve (Table 3). The results revealed that after 3 weeks, blood glucose levels in the Jerusalem artichoke particles low-, intermediate- and high-dose groups were significantly decreased (P<0.05). The effects in the intermediate- and high-dose groups were similar to that in the metformin hydrochloride group, indicating that Jerusalem artichoke particles had a significant hypoglycemic effect in type I diabetic rats.

Establishment of the type II diabetic rat model

A total of 42 rats were successfully established. DM rats were randomly grouped according to body weight into six groups with seven rats in each group. In the positive control group, five rats were administered glibenclamide, while two rats were administered Jin Li Da particles. Changes in diet and bedding were similar to those of type I diabetic rats.

Body weight measurements: The rats in the blank group displayed stable growth; each rats weight after a slight downward trend, but before and after modeling its change was not statistically significant (P>0.05). The model group displayed a positive growth phenomenon. Following administration in each experimental group, rat body weights significantly increased (P<0.05). On days 14 and 28 after administration, weight changes in the experimental and positive control groups were higher than that of the model group (P<0.05). Before and after modeling, rat body weights were decreased, and the weight of the rats after treatment was increased to some extent. Thus, diabetes symptoms may have been improved, but this observation requires further studies to confirm this point (Figure 2).

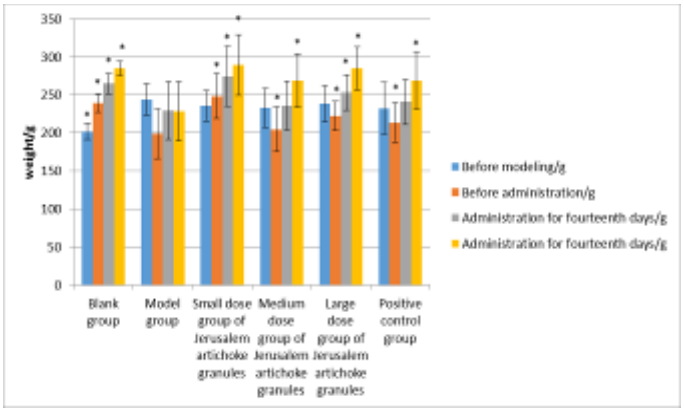

Figure 2: Effects of Jerusalem artichoke particles on body weight in type II diabetic rats

Changes in blood glucose levels in type II diabetic rats

After 4 weeks of administration, fasting glucose levels were measured using the glucose oxidase method and compared with levels before administration (Table 4). The results revealed that 4 weeks after Jerusalem artichoke

Table 3: Effects of Jerusalem artichoke particles on blood glucose levels in type I diabetic rats

Group	Blood glucose/mmol·L^{-1}	
	Before administration	After administration
Blank group	7.0±0.9	5.7±0.3
Model control group	8.0±0.1	5.4±0.5
Small dose group of Jerusalem artichoke particles	8.2±0.5	7.4±0.2*#
Middle dose group of Jerusalem artichoke particles	8.3±0.6	6.5±0.9*#
Large dose group of Jerusalem artichoke particles	8.4±0.4	7.1±0.9*#
Positive control group	8.2±0.7	6.6±0.7#

*P<0.05, compared with before administration; #P<0.05, compared with the model control group after administration.

Table 4: Effects of Jerusalem artichoke particles on blood glucose levels in type II diabetic rats

Group	Blood glucose/mmol·L^{-1}	
	Before administration	After administration
Blank group	4.5±1.4	7.1±1.2#
Model control group	11.4±2.5	14.4±4.3
Small dose group of Jerusalem artichoke particles	11.6±4.4	10.2±5.0#
Middle dose group of Jerusalem artichoke particles	11.9±4.6	8.5±4.6*#
Large dose group of Jerusalem artichoke particles	13.3±6.6	10.4±6.2*#
Positive control group	11.7±1.5	7.6±1.6#

particle administration, blood glucose levels in the Jerusalem artichoke particles high- and intermediate-dose groups were decreased significantly (P<0.05), but the hypoglycemic effect was less than that in the positive control group. Blood glucose levels in the Jerusalem artichoke particles and positive control groups were significantly lower than those in the model group after administration (P<0.05). The above data demonstrated that Jerusalem artichoke particles had hypoglycemic effects on diabetic rats.

DISCUSSION

STZ is a free radical activator that selectively destroys the islet cells and causes glucose metabolism disorders. It also causes hexokinase deficiency, so that glucose cannot be phosphorylated. Thus, the glucose through the cell membrane, resulting in high blood sugar levels and diabetes symptoms (KIHO T, 2002). A high-fat diet- and low-dose STZ-induced type II diabetic rat model mimics the natural history of the disease events (from insulin resistance to β-cell dysfunction) (Zeng Zhang et al., 2011). The rationale for combining a high-fat diet with low-dose STZ was to produce insulin resistance and cause the initial β-cell dysfunction with subsequent frank hyperglycemia in the adult SD rat (Khan H B et al., 2012). In this study, a rat model of type I diabetes mellitus was established successfully by intraperitoneal injection of STZ, while a rat model of type II diabetes mellitus was established by intraperitoneal injection of STZ after rats were fed a high-fat and high-sugar diet.

In this study, we developed a Jerusalem artichoke best preparation technology, developed in this study for two times, each time 70 min, 20 times of water, and retains its full effect. It improved the working process of the Jerusalem artichoke. The program is simple, low cost and highly efficient, and the effectiveness is more significant.
Medicinal plants are becoming increasingly popular. Diabetes has historically been treated with plants or plant-derived formulations in different cultures (Balázs A, 2010). These plants are also consumed as food for the treatment of diabetes (Aslan M et al., 2010). The Jerusalem artichoke has the potential to attenuate lipid disturbances and insulin resistance, but the underlying mechanisms are not well understood. One study proposed that 10% Jerusalem artichoke supplementation may be beneficial for prevention of type II diabetes onset (Chang WC et al., 2014).

The active ingredient of the Jerusalem artichoke is inulin. It is not hydrolyzed into monosaccharides in the upper part of the gut and thus will not raise blood sugar or insulin levels (Wang WL et al., 2007). This study confirmed the pharmacodynamic properties of Jerusalem artichoke particles. The results of the experiment revealed that the high content of inulin from Jerusalem artichoke particles had hypoglycemic effects in a type I diabetic rat model. The hypoglycemic effect in the Jerusalem artichoke particle intermediate-dose group was particularly obvious, and the results were even more dramatic than metformin hydrochloride treatment (Table 3). Both the intermediate- and high-dose groups displayed a significant hypoglycemic effect on diabetic rats (Table 4).

Jerusalem artichokes have high ecological adaptability, wider ecological range and the ability to survive in harsh natural environments and are low cost (Wang SJ et al., 2011). The development and utilization of the Jerusalem artichoke has wide development prospects. The experimental production of Jerusalem artichoke particles has the advantages of low cost, high inulin content, sweet taste and a hypoglycemic effect. Whether it is used as a drug or food, the development prospects are bright, and there are benefits for the majority of patients with diabetes. In this study, we observed a powerful hypoglycemic effect of Jerusalem artichoke particles, probably due to promote to the progress of gluconeogenesis. However, further studies are needed to elucidate the exact mechanism of action.

CONCLUSIONS

The Jerusalem artichoke particles retained the full effect, having powerful hypoglycemic effect, supporting the use of this plant part in folk medicine and promoting the development of new therapeutics based on natural products.

ACKNOWLEDGEMENTS

This study was supported by the innovation and entrepreneurship project of Hebei University (grant numbers: 2016052). This study was funded by the Science and technology research of Hebei colleges and universities Hebei (No. QN2016077) health and family planning commission of Hebei (No. 20160388). The authors wish to thank Guowei Zhang (M.D. and Ph.D., China) for his invaluable assistance. We gratefully acknowledge Guang-fu Yang (director of the physician, China) for supplying the fresh Jerusalem artichoke.

REFERENCES

Andreassen OA, Dedeoglu A, Stanojevic V (2002). Huntington's Disease of the Endocrine Pancreas: Insulin Deficiency and Diabetes Mellitus due to Impaired Insulin Gene Expression. Neurobiology of disease. 11(3):410-24.

Aslan M, Orhan N, Orhan DD (2010). Hypoglycemic activity and antioxidant potential of some medicinal plants traditionally used in Turkey for diabetes. Journal of Ethnopharmacology. 128(2):384.

Balázs A. (2010). Role of phytotherapy in the prevention and treatment of obesity. Orvosi Hetilap. 151(19):763-773.

Byungsung P (2011). Effect of Oral Administration of Jerusalem Artichoke Inulin on Reducing Blood Lipid and Glucose in STZ-Induced Diabetic Rats. Journal of Animal & Veterinary Advances. 10(19):2501-2507.

Chang WC, Jia H, Aw W (2014). Beneficial effects of soluble dietary Jerusalem artichoke (Helianthus tuberosus) in the prevention of the onset of type 2 diabetes and non-alcoholic fatty liver disease in high-fructose diet-fed rats. British Journal of Nutrition. 112(5):709-717.

Chen ZC, Lin YY, Xie BF. (1995). Study on Extraction of Polysaccharides from letinous edodes by enzymatic hydrolysis. Progress in Biotechnology. (01):47-50.

Cui WX. (2008). Health Problems of Adolescents with Type I Diabetes Mellitus and Educational Intervention Strategies. Journal of Applied Clinical Pediatrics. 23(8):628-630.

Fan S, Liu J, Wang Z (2010). Bio-Enzyme-Assisted Extraction Method for Inulin. US20100119651.

Giugliano D, Maiorino MI, Bellastella G, et al. (2016). Glucose, cholesterol, and blood pressure: is lower always better for type 2 diabetes?. Endocrine. 54(1):1-6.

Han L, Zhang J M, Xin-Xiong L U, et al. (2014). Research Progress on Stress Response and Conservation of Jerusalem Artichoke (*Helianthus tuberous* L.) Germplasm. Journal of Plant Genetic Resources. 15(5):999-1005.

Jung WY, Sang SL, Kim CW (2014). RNA-Seq Analysis and De Novo Transcriptome Assembly of Jerusalem Artichoke (Helianthus tuberosus Linne). Plos One. 9(11):e111982.

Khan H B, Vinayagam K S, Sekar A (2012). Antidiabetic and antioxidant effect of Semecarpus anacardium Linn. nut milk extract in a high-fat diet STZ-induced type 2 diabetic rat model. Comparative Clinical Pathology. 21(6):1395-1400.

KIHO T (2002). Hypoglycemic activity of polysaccharide fraction from Rehmannia glutinosa Libosch. f. hueichingensis Hsiao and the effect on carbohydrate metabolism in normal mouse liver. Planta Med. 112(6): 393-400.

Li JH, Wang M, Lin SM, Hu JM, Yang JC (2015). The therapeutic effects of Jerusalem artichoke inulin on rats with streptozotocin-induced type I diabetes. Modern Preventive Medicine. (16):2997-3000.

Lu GB, Ren CJ, Cui WZ (2011). The Preliminary Structure of Polysaccharide MLP II from Mulberry Leaves and its Hypoglycemic Effect on Rat Model of Diabetes. Science of Sericulture. 37(6):1053－1060.

Menegas LZ, Pimentel TC, Garcia S (2013). Dry-fermented chicken sausage produced with inulin and corn oil: Physicochemical, microbiological, and textural characteristics and acceptability during storage. Meat Science. 93(3):501-506.

Ren J, Davidoff AJ (1997). Diabetes rapidly induces contractive dysfunctions in isolated ventricular

myocytes. Am J Physiol. 272(1)：148-158.
Wan-Ching Chang, Huijuan Jia (2014). Beneficial effects of soluble dietary Jerusalem artichoke (Helianthus tuberosus) in the prevention of the onset of type 2 diabetes and non-alcoholic fatty liver disease in high-fructose diet-fed rats. British Journal of Nutrition. 112(5):709-717.
Wang SJ, Meng YY, Sun SQ (2011). Research advances of Jerusalem artichoke. Shandong Science. 24(6):62-67.
Wang WL, Zhang QZ, Sun HC (2007). Study on development and utilization of inulin. The processing of agricultural products. 10:90-91.
Xiao Xia, Wu Xiong, He Chun-lian (2014). Hypoglycemic Effect of Lily Polysaccharides in Type I Diabetic Rats. Food Science. 35(1) : 209.
Zeng Zhang, Hong-Li Xue, Yi Liu, Wen-Jian Wang (2011). Yi-Qi-Zeng-Min-Tang, a Chinese medicine, ameliorates insulin resistance in type 2 diabetic rats. World Journal of Gastroenterology. (08):987-995.
Zhang YY, Yang ZW (2011). Rodent models of diabets mellitus. Acta Laboratorium Animalis Scientia Sinica. (03):269-274.

Accepted 18 June, 2017.

Citation: Zhao B, Linfeng L, Feng L, Zhuang L, Zhang L, Zhai Y, Zhang G (2017). Pharmacodynamic study of Jerusalem artichoke particles in type I and II diabetic rat models. International Journal of Herbs, Spices and Medicinal Plants. 2(1): 018-024.