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A new tactic to treat postprandial hyperlipidemia in diabetic rats with gastroparesis by improving gastrointestinal transit

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Abstract

Improvement of gastrointestinal transit was thought to be a new tactic to treat postprandial hypertriglyceridemia in diabetic individuals with gastroparesis. Diabetic gastroparesis, lipid load testing, and the effect of domperidone or aqueous extract of rhizomes of *Rheum palmatum* L. on postprandial hypertriglyceridemia were evaluated in alloxan-induced diabetic rats. Alloxan diabetic animals had a slow gastrointestinal transit, together with delayed and exaggerated postprandial hypertriglyceridemia, after oral administration of olive oil, which was significantly improved after oral administration of domperidone or *R. palmatum* L. However, atropine could prevent the effects of *R. palmatum* L. The reduced postprandial hypertriglyceridemia was highly correlated with the improvement in gastrointestinal transit. These results suggest that promotion of gastrointestinal transit may be useful for the treatment of postprandial hypertriglyceridemia in diabetic patients with gastroparesis. *R. palmatum* L. may become a new choice for these patients since it has more potential benefits than domperidone.

Keywords: Diabetic gastroparesis; Postprandial hypertriglyceridemia; *Rheum palmatum* L.; Domperidone; Atropine

1. Introduction

Gastrointestinal symptoms are responsible for substantial morbidity among patients with diabetes. Estimates of the incidence of gastrointestinal symptoms in this population range from 10% to 76% (Keshavarzian et al., 1987). Diabetic gastroparesis appears to be relatively common in diabetic patients: 40–50% of patients with diabetes show signs of delayed gastric emptying (Horowitz et al., 1991). The first clinical manifestation of the disorder may be poor glycemic control. Since administration of insulin is usually coordinated with mealtimes, poorly regulated levels of glucose in the blood may reflect the failure of the stomach to empty promptly (Silvers et al., 1998).

Exaggerated and delayed postprandial hypertriglyceridemia is common in diabetic patients (Axelsen et al., 1999; Evans et al., 1999; Golay, 2000; Mero et al., 1998). Postprandial hypertriglyceridemia is an important risk factor for coronary artery disease (Coughlan and Sorrentino, 2000; Gotto, 1998) and atherosclerosis (Yu and Cooper, 2001). Postprandial hypertriglyceridemia is usually treated with lipid-lowering drugs (Cottrell et al., 2003). However, for diabetic patients with gastroparesis, lipid-lowering drugs may not be fully effective because of a slow gastrointestinal transit. Furthermore, we suspect that slight or mild diabetic gastroparesis is associated with postprandial hypertriglyceridemia because slight or moderate slowing of gastrointestinal transit may be not coordinated with mealtimes and may not efficiently clear lipids (Xie et al., 2004). If so, prokinetic drugs which improve gastrointestinal transit might be considered for the treatment of postprandial hypertriglyceridemia.

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Domperidone (Tonini et al., 2004) is a specific peripherally acting dopamine antagonist used in the management of symptoms of gastroparesis, a common and potentially debilitating condition in patients with diabetes mellitus (Brown and Khanderia, 1990). Efficacy, tolerability, and quality-of-life outcomes show that domperidone significantly improves the upper gastrointestinal symptoms of diabetic gastroparesis and is well tolerated by patients with this condition (Silvers et al., 1998). *Rheum palmatum* L. has been long used as a plant drug with cathartic action in Chinese medicine and contains many active compounds with cathartic effect, such as rhein, emodin, aloe-emodin and sennoside A, B, C, D, E, and F. These cathartic compounds mainly improve intestinal transit (Yagi et al., 1997; Zhu et al., 2000), while domperidone mainly promotes gastric emptying. Here, domperidone and *R. palmatum* L. were selected as drugs that improve gastric emptying and intestinal transit, respectively.

In the present study, alloxan-induced diabetic rats were selected as animal model of diabetic gastroparesis (Liu et al., 2002). Experiments were conducted as follows: (1) evaluation of diabetic gastroparesis, (2) lipid load test, (3) effect of domperidone or (4) aqueous extract of rhizomes of *R. palmatum* L. on postprandial hypertriglyceridemia in alloxan-induced diabetic rats.

2. Materials and methods

2.1. Chemicals and reagents

Olive oil was purchased from Beijing Fangcao Medicinal Company (Beijing, China). Glucose and triglyceride kits were purchased from Zhong Sheng High-tech Bioengineering Company (Beijing, China). Domperidone and atropine were purchased from Beijing Medical Co. Ltd. (Beijing, China). Alloxan was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Other reagents used were of analytical grade and were manufactured in China.

2.2. Animals

Adult male Wistar rats (200–220 g) were obtained from the Beijing Vital River Laboratory Animal Center (Beijing, China). The animals were housed under standard conditions of temperature (25 °C), 12-h/12-h light–dark cycles and fed on a standard laboratory pellet chow diet and tap water ad libitum. They were fasted overnight before the experiments. Experimental animals were maintained in accordance with internationally accepted principles for laboratory animal use.

2.3. Collection of plant material

Rhizomes of *R. palmatum* L. were purchased from Beijing Songlan Herbal Drug Company of China and authenticated by Dr. Shouquan Lin, the Institute of Medicinal Plant, Chinese Academy of Medical Sciences. A voucher specimen (No. 020331) was deposited in the herbarium of the Laboratory of Pharmaceutical Sciences, Department of Biological Sciences and Biotechnology, Tsinghua University. This plant was dried in the shade and powdered, and the powder was used for the aqueous extraction.

2.4. Preparation of extract

Dried powder of rhizomes of *R. palmatum* L. was extracted with distilled water for 24 h at 80 °C. The extracted solution was condensed and dried to yield the extract used in the experiments. The yield of the aqueous extract of rhizomes of *R. palmatum* L. was 36.5% (w/w in terms of dried starting material). The aqueous extract of rhizomes of *R. palmatum* L. contained rhein, chrysophanol, emodin, aloe-emodin, physcion and sennoside. Total anthrones were not less than 0.5%.

2.5. Induction of diabetes

Wistar rats were intraperitoneally injected with alloxan at a dose of 150 mg/kg after being fasted for 24 h. A week later, blood glucose was measured in animals fasted overnight. The rats with a fasting blood glucose level exceeding 12 mmol/l were selected and used as diabetic animals. Diabetic rats with gastroparesis were evaluated 2 weeks after alloxan induction of diabetes.

2.6. Assessment of diabetic gastroparesis in alloxan-induced diabetic animals

Alloxan-induced diabetic rats with gastroparesis were divided into five groups: (1) untreated diabetic rats; (2) diabetic rats treated with domperidone; (3) diabetic rats treated with *R. palmatum* L. at an oral dose of 300 mg/kg; (4) diabetic rats treated with *R. palmatum* L. at an oral dose of 150 mg/kg; (5) diabetic rats treated with *R. palmatum* L. plus atropine at oral doses of 300 mg/kg and 0.2 mg/kg, respectively. Drugs were orally administered to rats fasted overnight. The untreated diabetic and normal groups were administered the same volume of saline. Then 0.5 h later, assessment of gastrointestinal function was conducted as follows (each trial is separately conducted).

2.6.1. Gastric emptying

The gastric emptying test was performed according to the method prescribed previously (Xu et al., 1994) with a slight modification. In brief, after an overnight fast, diabetic and normal animals were weighed and 1% (g/100 ml) phenolsulfonphthalein suspension in olive oil was orally administered at a dose of 10 ml/kg. After 25 min, the rats were killed by cervical dislocation. The stomachs were removed and were carefully washed with a solution of 0.5 mol/l

NaOH. The final volume of all washing solutions was adjusted to 60 ml and centrifuged at 3000×*g* for 10 min. Then 0.5 ml of the supernatants was diluted to 4 ml, and optical density was measured at 546 nm, and quantified on the basis of standard curves of phenolsulfonphthalein. Data are expressed as mg (phenolsulfonphthalein)/kg (body weight of animals).

2.6.2. Small intestinal transit

The small intestinal transit in both diabetic and normal animals was measured by a slight modification of the method prescribed previously (El-Salhy, 2001). In brief, after an overnight fast, a 5% charcoal suspension in olive oil was orally administered at a dose of 10 ml/kg to each animal. After 20 min, the animals were killed by cervical dislocation. The small intestine was immediately excised carefully without stretching and the distance traveled by charcoal was measured as well as the total length of the small intestine. Data are expressed as the proportion (%) of the distance traveled by the charcoal along the entire length of the small intestine.

2.6.3. Total gastrointestinal transit

Total gastrointestinal transit was evaluated by the method of Chen (1996) with a slight modification. In brief, after the animals were fasted overnight, a 1% (g/100 ml) charcoal suspension in olive oil was orally administered at a dose of 10 ml/kg. The time that animals first defecated black feces was recorded.

In another experiment, in the non-fasting state, diabetic and normal animals were allowed free access to chow and tap water. Each animal was placed alone in a metabolism cage and all feces in 24 h were carefully collected, dried and weighed. Data are expressed as total feces weights (g)/kg body weight.

2.7. Lipid load test

2.7.1. Oral, intraperitoneal and intraduodenal administration of olive oil in normal and alloxan-induced diabetic rats

Olive oil at a dose of 10 ml/kg was orally, intraperitoneally and intraduodenally administered to alloxan-induced diabetic animals fasted overnight. Normal control rats were treated the same as diabetic animals. Blood samples were collected from the tail vein at 0, 1, 2, 4 and 8 h. Serum was isolated by centrifugation at 1500×*g*, 4 °C for 10 min. Serum triglyceride levels were measured using kits within 2 h of sample collection. The intraduodenal administration of olive oil took less than 5 min.

2.7.2. Influence of domperidone on postprandial hypertriglyceridemia in alloxan-induced diabetic animals

The effect of domperidone, a specific peripherally acting dopamine antagonist, was evaluated its effect on serum triglyceride levels in diabetic rats with gastroparesis. Alloxan-induced diabetic rats with gastroparesis were divided into two groups: untreated and domperidone (10 mg/kg)-treated diabetic rats. Domperidone was orally administered to diabetic rats fasted overnight. The untreated diabetic and normal rats were administered the same volume of saline. Then 0.5 h later, olive oil was orally administered at a dose of 10 ml/kg to each rat. Blood samples for the measurement of serum triglyceride levels were collected from the tail vein just prior to and at 2, 4, 6, 8, 10, 12, 16 and 24 h after olive oil administration. All feces produced in 24 h were carefully collected, dried and weighed after the oral administration of olive oil. Triglycerides in feces were extracted and estimated according to the method of Rizvi et al. (2003).

2.7.3. Influence of *R. palmatum* L. on postprandial hypertriglyceridemia in alloxan-induced diabetic rats

The effect of *R. palmatum* L., a natural plant product with cathartic action, was investigate on the levels of postprandial serum triglycerides in diabetic rats with gastroparesis. Alloxan-induced diabetic rats with gastroparesis were divided into four groups: (1) untreated diabetic rats; (2) diabetic rats treated with *R. palmatum* L. at an oral dose of 300 mg/kg; (3) diabetic rats treated with *R. palmatum* L. at an oral dose of 150 mg/kg; (4) diabetic rats treated with *R. palmatum* L. plus atropine at an oral dose of 150 mg/kg and 0.2 mg/kg, respectively. Drugs were orally administered to rats fasted overnight. For *R. palmatum* L., to avoid excessive excretion of intestinal lipid, an appropriate dose of *R. palmatum* L. had been established in preliminary trials which guaranteed that no loose stools were observed in the treated diabetic animals during the course of experiments. The untreated diabetic and normal rats were administered with the same volume of saline. Then 0.5 h later, olive oil was orally administered at a dose of 10 ml/kg. Blood samples for the measurement of triglycerides were collected from the tail vein just prior to and at 2, 4, 6, 8, 10, 12, 16 and 24 h after olive oil administration. The total feces produced in 24 h were carefully collected, dried and weighed after the oral administration of olive oil. Triglycerides in feces were extracted and estimated according to the method proposed above.

2.8. Biochemical estimations

Blood glucose and triglyceride levels were measured using commercial kits as prescribed previously (Sun et al., 2002).

2.9. Statistical analyses

All values are expressed as means±S.D. Data were statistically analyzed by analysis of variance (ANOVA). *P* values less than 0.05 were considered statistically significant.

3. Results

3.1. Assessment of gastrointestinal transit

3.1.1. Gastric phenolsulfonphthalein emptying

The amount of phenolsulfonphthalein remaining in the stomachs was significantly higher (108.1%, $P<0.01$) in the untreated diabetic rats than in the normal controls. This increase was inhibited (−70.8%, $P<0.01$) in diabetic animals after the oral administration of domperidone (Table 1). The amount of phenolsulfonphthalein remaining in the stomach was also significantly lower (−57.8% and −55.3%, $P<0.01$, respectively) in diabetic rats treated with *R. palmatum* L. at a dose of 300 or 150 mg/kg in a dose-dependent manner than in the untreated diabetic controls. No significant decrease in the amount of gastric phenolsulfonphthalein was observed in diabetic rats treated with *R. palmatum* L. (300 mg/kg) plus atropine (0.2 mg/kg).

3.1.2. Small intestinal transit

The proportion (%) of the distance traveled by the charcoal along the entire length of the small intestine in the untreated diabetic rats was significantly lower than that in normal controls (−19.1%, $P<0.01$), as shown in Table 1. Domperidone had no significant effect on small intestinal transit in the treated diabetic animals. *R. palmatum* L. at doses of 300 and 150 mg/kg significantly promoted (20.3% and 10.3%, $P<0.01$, respectively) small intestinal transit in the treated diabetic animals in a dose-dependent manner. However, no significant effect was observed in diabetic animals treated with *R. palmatum* L. (300 mg/kg) plus atropine (0.2 mg/kg).

3.1.3. Total gastrointestinal transit

As shown in Table 1, the time to the first black feces was significantly longer in the untreated diabetic animals than in the normal control animals (115.9%, $P<0.01$). However, the time was significantly shorter (−16.8%, $P<0.01$) in diabetic rats treated with domperidone than in the untreated diabetic controls. There was also significant a reduction (−43.4%, $P<0.01$ and −30.5%, $P<0.01$, respectively) in the time in diabetic groups treated with *R. palmatum* L. (300 and 150 mg/kg, respectively) in a dose-dependent manner compared with that in the untreated diabetic controls. However, no significant effect was observed in diabetic rats treated with *R. palmatum* L. (300 mg/kg) plus atropine (0.2 mg/kg).

Total feces weight over 24 h was significantly lower in the untreated diabetic animals than that in the normal control animals (−51.6%, $P<0.05$; Table 1). No significant effect on the total feces weight over 24 h was observed in domperidone-treated diabetic animals compared with the untreated diabetic controls. *R. palmatum* L. at doses of 300 and 150 mg/kg significantly increased the weight of feces (36% and 9.3%, $P<0.01$, respectively) in the treated diabetic animals in a dose-dependent manner compared with that of the untreated diabetic controls. However, a significant decrease in the weight of feces was observed in diabetic animals treated with *R. palmatum* L. (300 mg/kg) plus atropine (0.2 mg/kg) compared with that of the untreated diabetic controls.

3.2. Changes in postprandial hypertriglyceridemia in normal and diabetic rats after oral, intraperitoneal and intraduodenal administration of olive oil

A delayed and exaggerated postprandial hypertriglyceridemia was demonstrated in diabetic rats compared with normal controls after oral administration of olive oil (Fig. 1). The area under the curve (AUC) for postprandial serum triglyceride concentrations from 0 to 12 h, i.e., $AUC_{0-12\ h}$ was significantly greater (135.9%, $P<0.01$) in diabetic rats (40.89±4.90 mmol h/l) than in normal controls (17.33±4.80 mmol h/l). There was a significant increase in the levels of serum triglycerides in the diabetic group at 4 h ($P<0.01$), 8 h ($P<0.01$) and 12 h ($P<0.01$) compared with those in the normal control group. Serum triglyceride concentrations peaked at 8 h in diabetic animals and at 2 h in normal controls after oral administration of olive oil.

After the intraperitoneal administration of olive oil, serum triglyceride levels peaked at 8 h in both diabetic and normal rats (Fig. 2). The $AUC_{0-12\ h}$ was significantly greater (91.6%, $P<0.01$) in diabetic rats (34.17±4.58 mmol

Table 1
Parameters of gastrointestinal transit in rats after the oral administration of domperidone or aqueous extract of rhizomes of *R. palmatum* L.

Groups	Doses of drugs (mg/kg)	Phenolsulfonphthalein remaining in stomach (mg/kg)	Small intestinal transit of charcoal (%)	Time to the first defecated black feces (min)	Feces weight in 24 h (g/kg)
(I) Normal rats		9.9±4.8**	74.4±2.1**	452±108**	15.5±1.2**
(II) Untreated diabetic rats		20.6±6.7	60.2±2.0	976±46	7.5±0.4
(III) Diabetic rats treated with domperidone	10	6.0±2.2**	63.2±3.4	812±40**	7.3±1.1
(IV) Diabetic rats treated with *Rheum palmatum*	300	8.7±2.9**	72.4±1.7**	552±48**	10.2±0.4**
(V) Diabetic rats treated with *Rheum palmatum*	150	9.2±2.2**	66.4±2.5**	678±49**	8.2±0.6**
(VI) Diabetic rats treated with *Rheum palmatum* plus atropine	300 and 0.2, respectively	14.3±3.0	57.4±4.8	944±62	3.4±0.9**

Data are expressed as means±S.D. (n=6).
** $P<0.01$ vs. untreated diabetic rats.

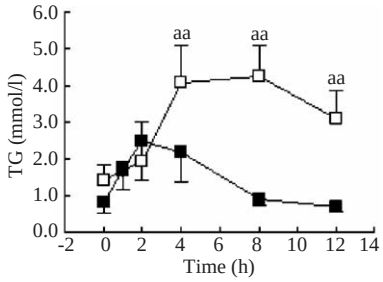

Fig. 1. Changes in serum triglyceride (TG) concentrations with time in diabetic (-□-) and normal rats (-■-) after the oral administration of olive oil. Data are expressed as means±S.D. (n=6). Serum triglyceride levels were significantly higher in diabetic animals at 4, 8 and 12 h than in normal controls (^{aa}P<0.01).

h/l) than in normal controls (17.83±3.31 mmol h/l). The $AUC_{0–12\ h}$ in diabetic rats after the intraperitoneal administration of olive oil was smaller than that in diabetic rats after oral administration (P<0.05). Though serum triglyceride levels in the diabetic group significantly increased at 8 h (P<0.01) and 12 h (P<0.01) compared with those in the normal control, a parallel increase or decrease in the levels of serum triglycerides was observed in the diabetic and normal rats over 12 h. These data indicated that without the involvement in gastrointestinal transit, the delayed and exaggerated postprandial hypertriglyceridemia was ameliorated in diabetic animals with gastroparesis.

After intraduodenal administration of olive oil, serum triglyceride levels peaked at 8 h in both diabetic and normal rats (Fig. 3). The $AUC_{0–12\ h}$ was significantly greater (107.5%, P<0.01) in diabetic rats (32.17±6.31 mmol h/l) than in normal controls (15.5±2.74 mmol h/l). The $AUC_{0–12\ h}$ in diabetic rats after the intraduodenal administration of olive oil was smaller than that in diabetic rats after oral administration (P<0.05). A parallel increase or decrease in serum triglycerides was also observed in diabetic and normal rats over 12 h, though serum triglycerides in diabetic rats increased faster at 8 h (P<0.01) and 12 h (P<0.01) than those in normal controls. These data suggested that the delayed and exaggerated postprandial hypertriglyceridemia was improved in diabetic animals with gastroparesis when gastric emptying was not involved.

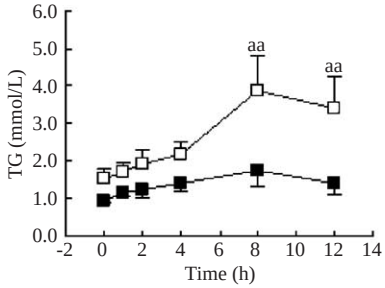

Fig. 2. Changes in serum triglyceride (TG) concentrations with time in diabetic (-□-) and normal rats (-■-) after the intraperitoneal administration of olive oil. Data are expressed as means±S.D. (n=6). Serum triglyceride levels were significantly higher in diabetic animals at 8 and 12 h than in normal controls (^{aa}P<0.01).

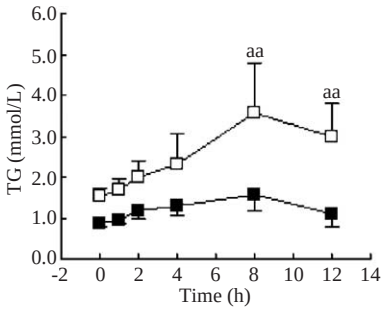

Fig. 3. Changes in serum triglyceride (TG) concentrations with time in diabetic (-□-) and normal rats (-■-) after the intraduodenal administration of olive oil. Data are expressed as means±S.D. (n=6). Serum triglyceride levels were significantly higher in diabetic animals at 8 and 12 h than in normal controls (^{aa}P<0.01).

3.3. Influence of domperidone on postprandial hypertriglyceridemia in diabetic rats

After olive oil administration, an exaggerated and delayed postprandial hypertriglyceridemia developed in the untreated diabetic rats with gastroparesis in comparison to that in normal controls (Fig. 4). The AUC for postprandial serum triglyceride concentrations from 0 to 24 h, i.e., $AUC_{0–24\ h}$ was significantly greater (137.6%, P<0.01) in the untreated diabetic rats (113.24±36.03 mmol h/l) than in the normal controls (47.66±13.56 mmol h/l). Domperidone at the dose of 10 mg/kg significantly inhibited the increase in postprandial hypertriglyceridemia in the treated diabetic rats with gastroparesis within 24 h compared with the untreated diabetic controls with gastroparesis. The $AUC_{0–24\ h}$ was markedly smaller (−43.3%, P<0.05) in the domperidone-treated diabetic rats (64.27±46.98 mmol h/l) than in the untreated diabetic controls (113.34±36.03 mmol h/l). No significant difference in fecal triglycerides was observed between experimental groups during 24 h after olive oil administration (data were not shown).

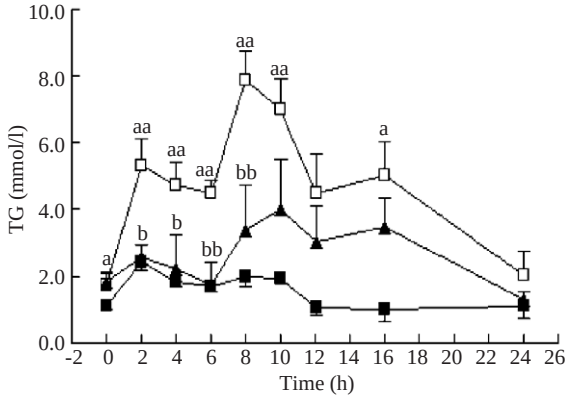

Fig. 4. Changes in postprandial serum triglyceride (TG) concentrations with time in normal (-■-), untreated diabetic (-□-) and domperidone-treated diabetic rats (-▲-) prior to and at 2, 4, 6, 8, 10, 12, 16 and 24 h after olive oil administration. Data are expressed as means±S.D. (n=6). ^{a}P<0.05, ^{aa}P<0.01 untreated diabetic rats vs. normal controls; ^{b}P<0.05, ^{bb}P<0.01 domperidone (10 mg/kg)-treated diabetic rats vs. untreated diabetic controls at different time points.

3.4. Influence of *R. palmatum* L. on postprandial hypertriglyceridemia in rats

The $AUC_{0-24\ h}$ was significantly higher (129.4%, $P<0.01$) in the untreated diabetic rats (98.00±29.50 mmol h/l) than in normal controls (42.72±5.70 mmol h/l) (Fig. 5). *R. palmatum* L. at doses of 300 mg/kg and 150 mg/kg significantly inhibited the increase in postprandial hypertriglyceridemia in the treated diabetic rats with gastroparesis for up to 12 h and 10 h compared with the untreated diabetic controls with gastroparesis, respectively. The data showed that *R. palmatum* L. at doses of 150 and 300 mg/kg significantly inhibited (−51.7%, $P<0.01$; −37.6%, $P<0.05$; respectively) the increase in $AUC_{0-24\ h}$ in the treated diabetic rats (47.36±5.32 and 61.12±22.06 mmol h/l, respectively) in a dose-dependent manner compared with that in the untreated diabetic controls (98.00±29.50 mmol h/l).

Atropine at the dose of 0.2 mg/kg markedly attenuated the effect of 300 mg/kg *R. palmatum* L. on the inhibition of the increase in postprandial serum triglyceride levels in diabetic rats treated with *R. palmatum* L. plus atropine compared with that in diabetic rats treated with *R. palmatum* L. (alone). The peak of serum triglycerides was delayed in diabetic rats treated with *R. palmatum* L. plus atropine compared with that in the untreated diabetic controls with gastroparesis, while the $AUC_{0-24\ h}$ for postprandial triglyceride levels did not change significantly ($P=0.567$) in the diabetic group treated with *R. palmatum* L. plus atropine (85.64±36.24 mmol h/l) compared with that in the untreated diabetic rats with gastroparesis (98.00±29.50 mmol h/l). These data indicated that atropine prevented *R. palmatum* L. from lowering postprandial serum triglyceride levels in diabetic rats with gastroparesis, by slowing intestinal transit. Therefore, intestinal transit may play an important role in regulating postprandial serum triglyceride metabolism in diabetic rats with gastroparesis. There was also no significant difference in fecal triglycerides between the experimental groups during 24 h after olive oil administration (data were not shown).

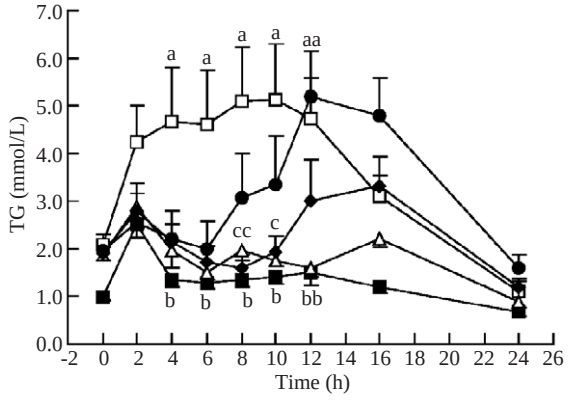

Fig. 5. Changes in postprandial serum triglyceride (TG) concentrations with time in normal rats (-■-), untreated diabetic rats (-□-), diabetic rats treated with *R. palmatum* L. at a dose of 150 mg/kg (-◆-), diabetic rats treated with *R. palmatum* L. at a dose of 300 mg/kg (-△-) and diabetic rats treated with *R. palmatum* L. (300 mg/kg) plus atropine (0.2 mg/kg) (-●-) prior to and at 2, 4, 6, 8, 10, 12, 16 and 24 h after olive oil administration. Data are expressed as means±S.D. ($n=6$). $^{a}P<0.05$, $^{aa}P<0.01$ untreated diabetic rats vs. normal controls; $^{b}P<0.05$, $^{bb}P<0.01$ diabetic rats treated with *R. palmatum* L. at a dose of 300 mg/kg vs. untreated diabetic controls; $^{c}P<0.05$, $^{cc}P<0.01$ diabetic rats treated with *R. palmatum* L. at a dose of 150 mg/kg vs. untreated diabetic rats at different time points.

3.5. Correlation analyses

Correlation analysis was conducted between means of both $AUC_{0-24\ h}$ of serum triglyceride concentrations and parameters of gastrointestinal transit in the experimental groups. The $AUC_{0-24\ h}$ was highly positively correlated with the amount of gastric phenolsulfonphthalein ($r=0.8744$, $P<0.01$) and with the time to the first defecated black feces ($r=0.8820$, $P<0.01$) in the normal control, untreated diabetic control and domperidone-treated diabetic groups. However, no significant correlation was observed between the $AUC_{0-24\ h}$ and small intestinal transit or total feces weight in 24 h in the normal control, untreated diabetic control and domperidone-treated diabetic groups. For the normal control, untreated diabetic control, and diabetic groups treated with *R. palmatum* L., the $AUC_{0-24\ h}$ was highly positively correlated with the amount of gastric phenolsulfonphthalein ($r=0.9143$, $P<0.01$) and with the time to the first defecated black feces ($r=0.9870$, $P<0.01$) and negatively correlated with small intestinal transit ($r=-0.9429$, $P<0.01$) and total feces weight in 24 h ($r=-0.7706$, $P<0.01$). These data suggest that the reduced postprandial hypertriglyceridemia may be correlated with the promotion of gastrointestinal transit in diabetic animals after oral administration of gastrointestinal prokinetic drugs.

4. Discussion

In the present study, alloxan-induced diabetic rats had slight or mild gastroparesis with a slow gastric emptying and intestinal transit compared with those of normal controls, which indicated that alloxan-induced diabetic rats could be used as an animal model of diabetic gastroparesis. These dysfunctions of gastrointestinal transit in diabetic animals could be significantly improved by administration of domperidone and *R. palmatum* L. Domperidone significantly promoted gastric emptying but it appeared not to improve small intestinal transit although it significantly shortened the time to the first defecated feces in diabetic animals. However, *R. palmatum* L. had significant effects on gastric emptying and small intestinal transit in alloxan-induced diabetic animals with gastroparesis. The effect of *R. palmatum* L. on the improvement of gastric emptying may be a secondary response to the promotion of small intestinal transit since most studies have reported that it mainly improves intestinal transit.

Delayed and exaggerated postprandial hypertriglyceridemia was seen in diabetic animals after the oral administration of olive oil but was attenuated after intraperitoneal or intraduodenal administration. This phenomenon might be partly attributed to the slow gastrointestinal transit since the absorption of olive oil after intraperitoneal or intraduodenal administration was not involved in gastrointestinal transit or gastric emptying, respectively. Domperidone, a gastric prokinetic, significantly lowered postprandial hypertriglyceridemia, which further indicates that a slow gastric emptying was one of the important reasons for postprandial hypertriglyceridemia since domperidone had no lipid-lowering action in addition to its improvement of gastric emptying. A good correlation between postprandial hypertriglyceridemia and gastrointestinal transit was demonstrated in domperidone-treated animals, which further confirmed the idea that improvement of gastrointestinal transit might be useful for the treatment of postprandial hypertriglyceridemia.

Moreover, *R. palmatum* L., a plant drug that improves intestinal transit, also significantly decreased postprandial hypertriglyceridemia by promoting intestinal transit in a dose-dependent manner, which indicates that a slow intestinal transit is also an important reason for postprandial hypertriglyceridemia, although *R. palmatum* L. may have a direct action of lipid-lowering (Jin and Jiao, 1994; Zhang et al., 2002). However, atropine could prevent the action of *R. palmatum* L., and a more delayed postprandial hypertriglyceridemia was demonstrated in the treated diabetic rats than in the untreated diabetic controls, which suggests that the improvement of postprandial hypertriglyceridemia by *R. palmatum* L. can mainly be attributed to the improvement of intestinal transit, instead of a direct lipid-lowering action. These results indicate that by improving intestinal transit, *R. palmatum* L. can inhibit postprandial hypertriglyceridemia in diabetic animals with gastroparesis, there being a good correlation between postprandial hypertriglyceridemia and gastrointestinal transit with *R. palmatum* L.

Thus a compound or natural plant product that improves gastric emptying or intestinal transit can be used to treat postprandial hypertriglyceridemia in diabetic rats with gastroparesis, which also indicates that diabetic gastroparesis involves not only in the stomach but also the intestine. The results supported our supposition that domperidone or *R. palmatum* L. could improve postprandial hypertriglyceridemia in diabetic rats by improving gastrointestinal transit. The decrease in postprandial hypertriglyceridemia may be not associated with a poor absorption of intestinal lipid because no obvious difference in fecal triglyceride content was observed between the treated groups and the untreated group in previous trials.

The active compounds of *R. palmatum* L. may improve intestinal transit by stimulating prostaglandin E (Yagi et al., 1991) and histamine (Autore et al., 1990) formation, inhibiting Na^+, K^+-ATPase (Zhou and Chen, 1998) and reducing in the frequency of colonic slow waves (Tong et al., 2000). However, an antagonist of the muscarinic cholinoceptor, atropine, attenuated the effect of the improvement of intestinal transit induced by *R. palmatum* L., which indicates that the effect might be partly attributed to an acetylcholine-like action (Shen, 1997). There tended to be an increase in postprandial serum triglyceride levels in the domperidone (or *R. palmatum* L.)-treated group, which may be associated with the administration of a single dose of drugs because at that time domperidone and active compounds in *R. palmatum* L. may have been undergoing elimination. The half-life of domperidone is about 8–10 h (Michiels et al., 1981) and that of rhein, a marker substance in *R. palmatum* L., is 3–4 h (Lee et al., 2003).

In addition, in Chinese medicine, *R. palmatum* L. is commonly used as a medicinal plant with many pharmacological actions and clinical uses besides improvement of intestinal transit. *R. palmatum* L. is used in the treatment of chronic renal failure (Bi et al., 1982), hyperlipidemia (Jin and Jiao, 1994; Zhang et al., 2002) and memory impairment (Tian et al., 1997). Besides diabetic gastroparesis, diabetic patients experience many other complications such as diabetic nephropathy (Zhao et al., 2003), dyslipidemia (Krauss, 2004), and memory loss (Arvanitakis et al., 2004). In addition, natural plant products with gastric prokinetic activities or which improve intestinal transit are more popular among patients with gastroparesis because natural products are free from adverse effects. Therefore, *R. palmatum* L. may have more potential benefits than domperidone in the management of diabetic patients with gastroparesis.

In conclusion, improvement of gastrointestinal transit may be a new tactic for the treatment of postprandial hypertriglyceridemia in diabetic patients with gastroparesis, and the use of *R. palmatum* L., a natural product that improves gastrointestinal transit, may be a new choice for these patients. Further studies should investigate (1) whether a synergistic hypolipidemic action occurs if prokinetic drugs are combined with lipid-lowering drugs to treat postprandial hypertriglyceridemia in diabetic individuals with gastroparesis; (2) whether prokinetic drugs have a hypolipidemic effect on postprandial hypertriglyceridemia in normal or diabetic individuals with normal gastrointestinal transit. It may be helpful for us to understand the hypolipidemic effect in patients with gastroparesis and to design new drugs with either prokinetic or lipid-lowering actions on the hyperlipidemia of diabetic individuals with gastroparesis.

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