EFFECTS OF THE TCM FORMULATION "WANG'S KETSUMEISEI EXTRACT" IN THE TREATMENT OF DIABETES MELLITUS

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Summary

A new TCM formulation, Wang's Ketsumeisei Extract (WK), was studied to determine its efficacy in the treatment of diabetes mellitus.

① *Testing in Animals.* Mice with alloxan-induced diabetes were used as a model. The alloxan-treated mice were divided into a reference control group and a WK treatment group, and mice that had not been treated with alloxan were used as a normal control group. The WK group was given WK(1.38/kg) and the reference control group was given an equivalent amount of distilled water *per os*. Blood glucose was measured in each group prior to administration and 30 days after the start of administration, both at fasting and postprandially at 0.5, 1, and 2 hours. ② *Clinical Testing.* 120 patients with Type II diabetes mellitus without complications such as cardiovascular, hepatic, or renal disorders were divided into a WK group and a Liuweiwan (Six-Ingredient-Pill) group. The WK group was given WK (9g/day) and the Liuweiwan group was given an equivalent amount of Liuweiwan without modification of the current anti-diabetic drug regimen or dosage. Prior to administration and 30 days after the start of administration, the blood glucose of the patients was measured both on an empty stomach and at 2 hours postprandially. Fasting serum lipids, urinary glucose, and serum insulin were also measured. In addition, 10 healthy volunteers were administered the same amount of WK. Although WK did not have any effect on the blood glucose levels of the normal mice or healthy subjects, blood glucose-lowering effects were noted in mice with alloxan-induced diabetes. In addition, an improvement in symptoms and blood-glucose-lowering effects at fasting and at 2 hours postprandially were noted in patients with Type II diabetes mellitus when WK was taken alone or in combination with oral blood-glucose-lowering agents. The absence of changes in the patients' blood insulin levels suggested the possibility that endogenous insulin resistance may have been corrected through improvements in constitution. Improved serum lipid levels were also noted in patients with hyperlipidemia. No obvious adverse reactions were observed.

The results of the study thus demonstrated that the TCM formulation, Wang's Ketsumeisei, is effective in diabetic mice and patients with Type II diabetes mellitus.

I. INTRODUCTION

The rapidly changing modern lifestyle has resulted in a sharp increase in the incidence of Type II diabetes mellitus. Type II diabetes mellitus is now a major problem for society.[1),2)]

In recent years, insulin resistance[3)-5)] has been identified as a major factor in the etiology of diabetes mellitus. The concept of insulin resistance is akin to the theory of constitution and "*zheng*" (patterns) in Chinese Medicine. In Ancient China, diabetes

mellitus was called "*xiao-ke*" (wasting-thirst), and it was classified into three stages, "*shang-xiao* (upper wasting)," "*zhong-xiao* (middle wasting)," and "*xia-xiao* (lower wasting)" for treatment. The formulation used in the Han Period, Liuweiwan, and formulations developed from it, including Baweiwan (Eight-Ingredient-Pill) and Niucheshenqiwan (Achyranthes and Plantago Kidney Qi Pill), are still used today.[6)-8)] However, these ancient formulations were developed against a background of food shortages and are only minimally effective against the diabetes of today, characterized by overeating. In this study, we investigated the effectiveness of a new TCM formulation, Wang's Ketsumeisei Extract, in the treatment of diabetes mellitus.

II. MATERIALS AND METHODS

1. Therapeutic Agent

The therapeutic agent was Huabeipai Wangshi Mingjing (Wang's Ketsumeisei, or WK, manufactured by North China Pharmaceutical Group Corporation), an extract produced from pure TCM herbs, including *Juemingzi* (Fetid Cassia), *Shanyao* (Discoreae Rhizoma), *Honghua* (Carthami Flos), *Shanzhizi* (Gardeniae Fructus), and *Gancao* (Glycerrhizae Radix).

2. Experimental Study

Over a period of one week, alloxan (alloxan, 45 mg/kg) was intravenously administered to mice weighing 24-26 g once a day following a 24-hour fasting. At Day 7, the fasting blood glucose levels (5-hour fasting) of the mice were measured. Mice with a blood glucose level greater than 180 mg/dl (10 mmol/L) were used as a diabetes mellitus model. The model was divided into a reference control group and treatment groups. The reference control group received distilled water. The treatment group (WK group) was given 1.38 g/kg/day Wang's Ketsumeisei (equivalent to 10 times the adult human dose (9 g/65 kg)) and a low-dose treatment group (WK1 group) half that amount (0.69 g/kg) *per os* under normal feeding conditions for 30 days. In each group, blood was drawn immediately prior to the start of the experiment and at 30 days after administration, on an empty stomach with alloxan, as well as at 0.5 h, 1 h, and 2 h after oral glucose (2.5 g/kg) administration.

In addition, non-alloxanized mice were divided into a reference control group and a Wang's Ketsumeisei (WK) group. The WK group was given 1.38 g/kg/day Wang's Ketsumeisei and the reference control group an equivalent amount of distilled water *per os* every day. Fasting (5-hour fasting) blood glucose was measured immediately prior to the experiment and at 30 days after the start of administration.

3. Clinical Testing

(1) Diagnostic Criteria for Diabetes Mellitus

The old 1985 WHO diagnostic criteria were used (these criteria are still in use in China). The criteria were: fasting blood glucose ≥140 mg/dl (7.8 mmol/L) and 2-hour postprandial blood glucose ≥ 200 mg/dl (11.1 mmol/L).

(2) Observation Method

120 patients (18-65 years) with Type II diabetes mellitus (without cardiovascular, hepatic, or renal disorders or other complications) were divided into a treatment group

and a Liuweiwan (Six-Ingredient-Pill) group. The treatment group was given WK (9 g/day) and the Liuweiwan group was given an equivalent amount of Liuweiwan, without modification of the current anti-diabetic drug regimen or dosage. Prior to administration and 30 days after the start of administration, the blood glucose of the patients was measured both on an empty stomach and at 2 hours postprandially. Fasting serum insulin and serum lipids were also measured. Hematologic, biochemical and urine glucose testing was performed as well. Abdominal echograms, electrocardiograms and chest X-rays were obtained concurrently. In addition, the degree of improvement in symptoms, including thirst, excessive drinking, excessive urination, overeating, fatigue, shortness of breath, nocturia, and erectile dysfunction, was investigated using a questionnaire. Furthermore, 10 healthy volunteers were given the same amount of WK for 30 days and their fasting blood glucose levels were measured before and after the experiment.

(3) Effect Evaluation Criteria

Markedly effective: Primary symptoms have resolved; fasting blood glucose is ≤ 130 mg/dl or has decreased by ≥30% compared to immediately prior to the start of the experiment.

Effective: Primary symptoms have improved; fasting blood glucose is ≤ 150 mg/dl or has decreased by ≥10% compared to immediately before the start of the experiment.

Ineffective: Primary symptoms have not improved; fasting blood glucose has decreased by <10% compared to immediately before the start of the experiment.

4. Statistical Methods

Results were expressed as means ± SDs. Analytical methods consisted of variance analysis and Student's t-test

III. Results

1. Investigation in Mice

Fasting blood glucose following administration of Wang's Ketsumeisei Extract (WK) to normal mice for 30 days was 151.02 ± 17.26 mg/dl in the reference control group and 145.08 ± 25.56 mg/dl in the WK group. There were no significant differences pre- and post-experiment and no significant intergroup differences (Fig. 1).

In the alloxanized diabetic mice, fasting blood glucose increased to 513.54 ± 31.2 mg/dl in the reference control group but decreased significantly to 441.72 ± 38.22 mg/dl in the WK group ($P<0.05$) following 30 days of administration (Fig. 2).

Fig. 1 Variation in fasting blood glucose levels in normal mice

In the WK group, blood glucose at 1 h and 2 h post-glucose loading was not significantly different from that of the reference control group, but blood glucose at 0.5 h (675.54 ± 59.94 mg/dl) post-glucose loading was significantly lower than that of the reference control group (808.92 ± 46.44 ml/dl) ($P<0.01$) (Fig. 3).

Fig. 2 Variation in fasting blood glucose level in diabetic mice

Fig. 3 Variation in blood glucose level after glucose loading

3. Clinical Testing

(1) Patient Background

Background information on the 120 patients is provided in Table 1.

Table 1 Patient Background

		Liuweiwan Group	WK Group
Age (years)		56.08 ± 6.2	57.14 ± 6.20
Sex	Male (patients)	33	32
	Female (patients)	27	28
Duration of disease (in years)		5.05 ± 4.22	4.93 ± 3.21
Concomitant blood-glucose-lowering medication	SU (patients)	25	25
	BI	15	14
	SU+BI	10	11
	None	10	10

BI: biguanide
SU: sulfonyl urea

(2) Results of clinical study of blood-glucose-lowering effects of WJ

When WK was administered to normal subjects, their fasting blood glucose levels remained practically unchanged from pre- to post-administration at 91.03 ± 11.14 mg/dl and 92.08 ± 12.12 mg/dl respectively.

When the 30-day experiment was over, Type II diabetes mellitus patients in the Liuweiwan group exhibited a tendency to decreased fasting and 2-hour postprandial blood glucose levels, but the differences were not significant. In contrast, fasting blood glucose levels in the WK group (165.62 ± 15.56 mg/dl) decreased significantly ($P<0.01$) relative to pre-administration levels (212.22 ± 17.82 mg/dl) (Fig. 4) and were significantly lower ($P<0.05$) than those of the Liuweiwan group (199.89 ± 17.72 mg/dl). As illustrated in Fig. 5, the post-experiment 2-hour postprandial WK group blood glucose level was 309.8 ± 18.43 mg/dl, which was lower than the pre-administration level (347.58 ± 19.34 mg/dl) ($P<0.05$). A tendency to decreased levels compared to the Liuweiwan group (333.18 ± 28.44 mg/dl) was exhibited, albeit without significant differences. Symptoms including thirst, excessive drinking, and excessive urination improved markedly in the WK group compared to the Liuweiwan group (Table 2).

Fig. 4 Variation in fasting blood glucose level in diabetic patients
//Keys, Fig. 4//
(Bottom, left) Liuweiwan

Fig. 5 Variation in 2-hour postprandial blood glucose level in diabetic patients
//Keys, Fig. 5//
(Bottom, left) Liuweiwan

In the Liuweiwan group, the serum insulin levels before and after the experiment were 16.59 ± 6.10 μU/ml and 18.46 ± 7.33 μU/ml respectively. In the WK group, the pre- and

post-experiment fasting serum insulin levels were 16.82 ± 6.58 μU/ml and 15.05 ± 5.74 μU/ml respectively. No significant inter- or intra-group differences were noted from pre- to post-experiment. Urinary glucose decreased from 65.65% to 57.67% in the Liuweiwan group and markedly from 68.33% to 26.67% in the WK group. The efficacy rate was 26.7% in the Liuweiwan group and 80% in the WK (Table 3). In patients with hyperlipidemic complications, blood lipid improvements were noted in the WK group compared to the Liuweiwan group (Table 4). No significant changes in values from pre- to post-experiment were noted in the groups upon hematologic and biochemical testing (Table 5). Abdominal echograms, electrocardiograms, chest X-rays did not reveal any abnormal changes in either group, and there were no obvious adverse reactions.

Table 2 Rate of Improvement of Major Symptoms

Symptom	No. of Patients	Markedly Effective	Effective	Ineffective	Rate of Improvement (%)
Thirst	34 (32)	4 (0)	23 (4)	7 (28)	79.41 (12.5)
Overeating	33 (34)	1 (3)	24 (4)	8 (27)	75.76 (20.59)
Excessive drinking	30 (38)	0 (2)	21 (5)	9 (31)	70.00 (18.42)
Excessive urination	26 (34)	1 (1)	15 (6)	10 (27)	61.54 (20.59)
Fatigue	36 (39)	5 (4)	21 (2)	13 (33)	72.22 (15.38)
Shortness of breath	19 (21)	1 (0)	9 (3)	9 (18)	52.63 (14.28)
Nocturia	38 (40)	7 (1)	23 (11)	8 (28)	78.95 (30.00)
Erectile dysfunction	8 (9)	2 (0)	4 (1)	2 (8)	75.00 (11.11)

Values for the Liuweiwan group are noted in parentheses.

Table 3 Efficacy

	Liuweiwan Group	WK Group
Markedly effective (patients)	1	13
Effective (patients)	15	35
Ineffective (patients)	44	12
Effectiveness (%)	26.7	80.0

Table 4 Variation in Blood Lipids

		Patients	TC (mg/dl)		TG (mg/dl)	
			0 day	30 days	0 day	30 days
Liuweiwan Group	High lipids	40	308 ± 21	312 ± 23	180 ± 22	184 ± 26
	Normal range	20	186 ± 17	206 ± 21	128 ± 11	119 ± 24
WK Group	High lipids	43	310 ± 20	265 ± 24*	178 ± 15	162 ± 21
	Normal range	17	192 ± 16	210 ± 27	124 ± 21	122 ± 26

Table 5 Hematologic & Biochemical Testing

	WK Group		Liuweiwan Group	
	0 day	30 day	0 day	30 day
HGB (g/L)	136.3 ± 16.4	145.4 ± 11.1	137.1 ± 17.2	144.5 ± 14.7
RBC ($x10^{12}$/L)	4.36 ± 0.52	4.81 ± 0.42	4.42 ± 0.53	4.77 ± 0.45
WBC ($x10^{12}$/L)	5.47 ± 1.53	6.66 ± 1.81	5.54 ± 1.37	6.41 ± 1.12
TP (g/L)	80.67 ± 5.16	74.13 ± 4.66	78.85 ± 3.61	73.54 ± 3.30
ALB (u/L)	51.68 ± 1.93	44.93 ± 1.68	51.08 ± 2.12	44.87 ± 1.65
AST (u/L)	25.46 ± 8.38	22.76 ± 8.54	23.44 ± 6.93	20.61 ± 5.68
BUN (mol/L)	6.44 ± 1.33	5.31 ± 1.04	6.52 ± 1.80	5.48 ± 1.22
CREA (μmol/L)	94.83 ± 13.18	79.5 ± 13.17	93.82 ± 14.84	81.31 ± 11.82

IV Discussion

It is said that over 2% of the world's population has Type II diabetes mellitus, and the incidence of the disease is increasing particularly rapidly in Asia, in conjunction with the increasing economic development; in fact, Type II diabetics were reported to number 51.43 million, or 47% of the world's total, in Asia in 1995.[1),2)] The onset of Type II diabetes mellitus is precipitated by not only inadequate secretion of insulin but also insulin resistance. It is conjectured that Type II diabetes occurs when insulin resistance leads to constant hyperinsulinemia and pancreatic β-cell exhaustion, which result in disordered insulin secretion.

The term "insulin resistance" refers to a condition in which normal amounts of insulin are inadequate to produce an insulin response. In recent years, an increasing number of common diseases, including obesity, Type II diabetes mellitus, hypertension, and lipid metabolism disorders, have been linked to insulin resistance, and a condition resulting in a rapid progression of arteriosclerosis has been termed "insulin resistance syndrome."[3)]. TNF-α and free fatty acids (FFAs) have been identified as contributors to the pathogenesis of insulin resistance. FFAs are produced by fat cells. TNF-α is known to be produced by monocytes and macrophages. In recent years, however, it has also been found that TNF-α is secreted by the muscles and fat cells of obese patients. Both FFAs and TNF-α act on insulin target cells and induce insulin resistance.[4),5)]

In addition to genetic factors, stress, overeating, lack of exercise and other lifestyle habits contribute to the pathogenesis of insulin resistance and inadequate insulin secretion. As these predisposing factors result in a dysmetabolic diabetic constitution, diabetes must be treated through measures to improve the constitution as well as to reduce blood glucose.

In Ancient China, diabetes mellitus was called "*xiao-ke*" (wasting-thirst), and it was classified into three stages, "*shang-xiao* (upper wasting)," "*zhong-xiao* (middle wasting)," and "*xia-xiao* (lower wasting)" for treatment. The formulation used in the Han Period, Liuweiwan, and formulations developed from it, including Baweiwan (Eight-Ingredient-Pill) and Niucheshenqiwan (Achyranthes and Plantago Kidney Qi Pill), are still used today.[6)-8)] It has been reported that an herbal ingredient in Liuweiwan, *Shanzhuyu* (Corni Fructus), possesses anti-diabetic activity.[8)] In Chinese Medicine, the diabetes mellitus constitution is commonly characterized by Qi /Spleen vacuity and blood

stagnation, as well as Liver-Kidney Yin deficiency. The "Spleen" of Chinese Medicine refers to a functional system which includes the pancreas and is responsible for digestion, absorption and metabolism. Spleen Qi has a conveying function (digestion, absorption and metabolism of nutrients). It provides energy to the tissues of the entire body, including the muscles, and also replenishes Liver-Kidney Yin. A Spleen Qi deficiency, in which the functions of the Spleen decline, leads to "Stagnant Blood," or disordered blood circulation. It also causes depletion of the Liver-Kidney Yin. The "Heart-Fire" of stress also depletes Yin, exacerbating the Yin vacuity. In contrast, Wang's Ketsumeisei Extract was developed on the therapeutic principle of "boosting Qi," "nourishing Yin," and "quickening the Blood."

The results of the study revealed that Wang's Ketsumeisei Extract, when used alone, produced a blood-glucose-lowering effect in alloxanized diabetic mice, and, when used alone or in combination with oral blood-glucose-lowering agents, lowered the fasting and postprandial blood glucose levels in Type II diabetes mellitus patients and improved symptoms more effectively than Liuweiwan. The lack of increase in blood insulin levels suggested the possibility that endogenous insulin resistance was corrected through improvements in constitution. Further study into the combined effects of the formulation and insulin is currently underway. A beneficial effect on diabetic complications has been noted, and these will be reported in detail in a forthcoming report. It should also be noted that the Hebei Health Inspection Center has reported that WK has an LD_{50} of >21,500 mg/kg and is negative for teratogenicity in mice

The above results thus demonstrated that WK is safe and effective in a diabetic model and in Type II diabetes mellitus patients.