

Effect of Byakko-ka-ninjin-to with exercise in early stage symptom of non-insulin dependent diabetic mice

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Abstract

The antidiabetic action of Byakko-ka-ninjin-to (白虎加人参湯 ; BN) with exercise was investigated in KK-Ay mice, an animal model of non-insulin dependent diabetes mellitus (NIDDM). BN (90 mg/kg) decreased the blood glucose level of KK-Ay mice with exercise 1 ($p < 0.01$) and 2 weeks ($p < 0.05$) after the oral administration, while the control group (exercise only) did not change. On short term administration, BN also significantly decreased blood glucose level 5 days after the administration ($p < 0.05$). These findings indicate that BN with exercise is useful for the early stage symptom of NIDDM.

Key words Byakko-ka-ninjin-to (Bai-Hu-Jia-Ren-Shen-Tang, 白虎加人参湯), exercise, NIDDM.

Introduction

Insulin resistance in peripheral tissues is known as one of the major pathogenic factors of non-insulin dependent diabetes mellitus (NIDDM), together with the impairment of glucose-induced insulin secretion from pancreatic beta cells. Although therapeutic agents to stimulate insulin secretion (for example, sulfonylureas) have been used for NIDDM patients, drugs with exercise are not yet directly available clinically, but they are expected to become a new category of drugs which can be used in combination with exercise and medicines because of its use in early stage symptom of NIDDM.

Byakko-ka-ninjin-to (白虎加人参湯 ; BN) has been used in oriental as traditional medicine for diabetes (polyuria and polydipsia).¹⁾ Suzuki *et al.* reported that BN-treated mice reduced the blood glucose of alloxan-induced diabetic mice and KK-Ay mice.²⁾ However, there is no experimental evidence of the antidiabetic effect with exercise. The purpose of this study, therefore, is to examine the antidiabetic

effect with exercise using non-insulin dependent diabetic animal model.

Materials and Methods

The traditional Chinese prescription, Byakko-ka-ninjin-to (BN) was obtained from Tsumura Co., Tokyo Japan. The constituents of BN was indicated including 5 crude drugs, Gypsum Fibrosum (15), Glycyrrhizae Radix (2), Ginseng Radix (1.5), Anemarrhenae Rhizoma (5), Oryzae Semen (8). TJ-34 (crude powder extract) contains spray-dried aqueous extracts of 5 crude drugs as a mixture. These were stored at room temperature until use. The yield of extract was 23.5 %.

Animals : Male KK-Ay mice (Clea, Tokyo, Japan) at the early stage of non-insulin dependent diabetes condition, 6-11 weeks old, were used. Under non-fasting, those with blood glucose levels above 300 mg/dl were considered to be diabetic and used in this study. These mice were housed in an air-conditioned room at $22 \pm 2^\circ\text{C}$ with a 12 hour light and 12 hour dark cycle. The animals were kept in the experimental

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animal room for 7 days with free access to food and water. For the determination of blood glucose levels, blood samples (20 μ g) were withdrawn from the cavernous sinus with a capillary glass tube.

Exercise : For exercise studies, KK-Ay mice were run on a motorized treadmill (Muromachi Kikai Co. Ltd. Osaka, Japan) for 120 min (5 m/min, 20 % grade).³⁾ On 0, 1 and 2 weeks, blood samples were collected before the exercise and at 30, 60 and 120 min later. BN was administered 30 min before the exercise once a day. The studies were started at 13:00-14:00 pm.

Determination of blood glucose : Blood glucose levels in animals were determined by the glucose oxidase method.⁴⁾

Statistical analysis : All the data were expressed as means \pm S.E.M. and Student's *t* test was used for the statistical analysis. The values were considered to be significantly different when the *p* value was less than 0.05.

Results

Effect of BN on exercised mice (2 weeks)

In the basic study, we examined speed-dependence (0, 5, 10 and 15 m/min) on the exercise of normal mice and found that exercise at 5 m/min did not change the blood glucose (data not shown). We also examined dose-dependence (9, 90 and 900 mg/kg) on blood glucose in KK-Ay mice (0, 4, 7, 10 h after the treatment) and found that BN at 90 and 900 mg/kg-treated mice decreased the blood glucose 7 h after the treatment (data not shown). The blood glucose levels of BN (90 mg/kg)-treated mice with exercise are shown in Fig. 1. Administration of BN (90 mg/kg) gave a significant decrement in blood glucose levels when compared with the control (exercise only) values (BN 90 mg/kg, 1 week : $p < 0.01$, 2 week : $p < 0.05$). On the effect of exercise in KK-Ay mice, BN treatment (90 mg/kg) showed a decrease in the blood glucose levels when compared with the 0 week value (Fig. 3). Zero min of Fig.3 is the same as the data of Fig. 1. On the other hand, the control group (exercise only) had showed a significant decrease in blood glucose after 30, 60 and 120 min at 2 weeks when compared with the 0 week value (Fig. 2).

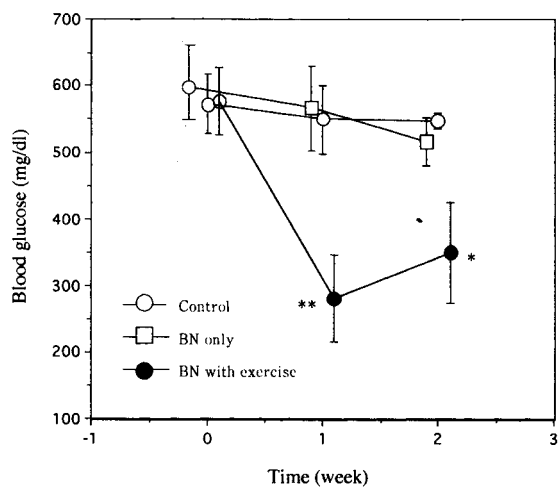


Fig. 1 Effect of BN with exercise on blood glucose in KK-Ay mice (2 weeks)

KK-Ay mice were run on a motorized treadmill for 120 min (5 m/min, 20 % grade). On 0, 1 and 2 weeks, blood samples were collected before the exercise and at 30, 60 and 120 min later. BN was administered 30 min before the exercise once a day (BN only is no exercise). Each point indicates the mean \pm S.E. from 4 mice. Significantly different from control, * $p < 0.05$, ** $p < 0.01$.

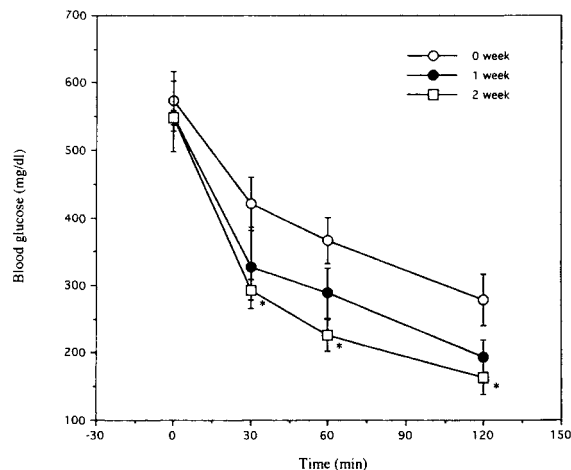


Fig. 2 Effect of exercise on blood glucose in KK-Ay mice (Control)

KK-Ay mice were run on a motorized treadmill for 120 min (5 m/min, 20 % grade). On 0, 1 and 2 weeks, blood samples were collected before the exercise and at 30, 60 and 120 min later. Each point indicates the mean \pm S.E. from 4 mice. Significantly different from corresponding control, * $p < 0.05$.

Effect of BN on exercised mice (1 week)

The blood glucose levels of BN-treated mice with exercise are shown in Fig. 4. After 5 days, administra-

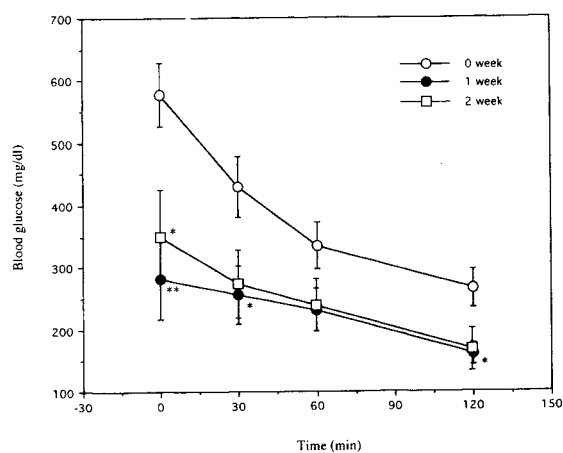


Fig. 3 Effect of BN with exercise on blood glucose in KK-Ay mice (BN)

KK-Ay mice were run on a motorized treadmill for 120 min (5 m/min, 20 % grade). On 0, 1 and 2 weeks, blood samples were collected before the exercise and at 30, 60 and 120 min later. BN was administered 30 min before the exercise once a day. Each point indicates the mean \pm S.E. from 4 mice. Significantly different from control, * p < 0.05, ** p < 0.01.

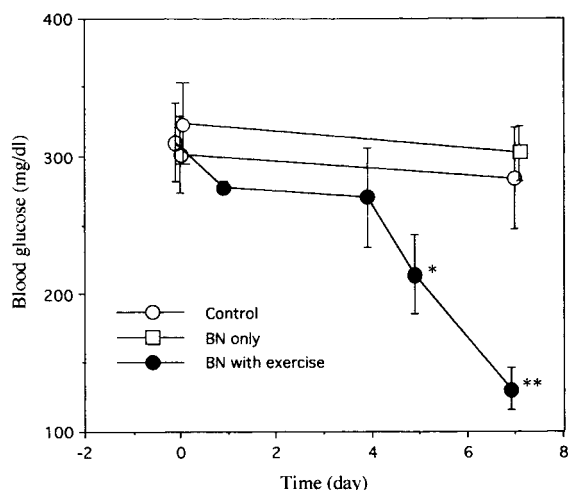


Fig. 4 Effect of BN with exercise on blood glucose in KK-Ay mice (1 week)

KK-Ay mice were run on a motorized treadmill for 120 min (5 m/min, 20 % grade). On 0, 1, 4, 5 and 7 days, blood samples were collected. BN was administered 30 min before the exercise once a day (BN only is no exercise). Each point indicates the mean \pm S.E. from 4 mice. Significantly different from control, * p < 0.05, ** p < 0.01.

tion of BN (90 mg/kg) with exercise gave a significant decrease in blood glucose levels when compared with the control values (exercise only).

Discussion

The present study clearly showed that Byakko-ka-ninjin-to (BN) with exercise produced an antidiabetic activity in KK-Ay mice. BN (90 mg/kg)-treatment decreased blood glucose level of KK-Ay mice with exercise 1 and 2 weeks after the administration while control group (exercise only and BN only) did not change. On 120 min exercise, the control group lowered blood glucose after 2 weeks when compared with 0 weeks (Fig. 2). From this finding, it would appear that the decrease of blood glucose may be due to an increase in insulin sensitivity. Moreover, BN-treated mice showed lowered blood glucose 5 days after the treatment. In general it is shown that exercise training had an increase of insulin sensitivity.^{5,6)} From these findings, it could be suggested that the antidiabetic effect of BN and exercise has a multiplier effect.

Further studies will be needed to elucidate the mechanism of these effects. These results suggest the validity of clinical use of Byakko-ka-ninjin-to with exercise in the early stage symptom of non-insulin dependent diabetes mellitus.

和文抄録

糖尿病発症初期では運動療法と食事療法が行われる。われわれは糖尿病発症初期の運動療法と漢方治療の併用により効果のあるものを検索したところ、白虎加人参湯 (BN) にその作用がみられた。そこで、糖尿病発症初期の動物を用いて運動療法との併用効果について検討した。BN は 2 週間の運動療法との併用で検討したところ、すでに 1 週目で有意な血糖値の低下が認められた。さらに、詳細に検討したところ、運動開始 5 日目より有意な血糖値の低下がみとめられた。以上のことから、BN と運動療法との併用はインスリン非依存型糖尿病の初期治療に有効であることが示唆された。

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