Effect of herbal acupuncture with Ginseng Radix Extract at ST36 and GB39 on the growth of mice

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Abstract

Objectives: To investigate the effect of Ginseng herbal acupuncture at GB39 and ST36 on growth in mice.

Methods: Mice were divided into four groups (n=10); G-ST36 group treated with Ginseng Radix herbal acupuncture at right ST36, G-GB39 group treated with the same at GB39, Control-ST36 group and Control-GB39 group injected with saline at right ST36 and GB39 respectively. The saline injection and herbal acupuncture were performed 3 times a week for 3 weeks. The length(total height, body length and tail length), weight and the protein efficiency ratio were measured as well as IGF-I level in serum

Results: The weights of Ginseng herbal acupuncture groups were not significantly increased compared to the control groups but the protein efficiency ratios were significantly higher in Ginseng herbal acupuncture groups compared to the control groups. The increases of the total length, body length and tail length were significantly higher in Ginseng herbal acupuncture groups than the control groups. The serum IGF-I level of G-GB39 was significantly increased compared to control-GB39, not that of G-ST36 compared to control-ST36.

Conclusions: Ginseng herbal acupuncture at GB39 and ST36 are both effective to promote growth in terms of height and weight. GB39 may be effective for growth by way of increasing IGF-I, and more closely related to growth in height than ST36.

Key words: Growth, ST36(Joksamni), GB39(Hyeonjong), Ginseng Radix herbal acupuncture, Mice

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I. Introduction

Ginseng is one of the herbs often used to promote growth in oriental medical clinics. Ginseng tonifies vital qi as well as strengthens the spleen and stomach¹⁾. Lim²⁾ reported that Ginseng herbal acupuncture at ST25 promoted growth in mice.

In this study, we investigated the effect of Ginseng herbal acupuncture at GB39 and ST36 on growth in mice. GB39 is the marrow influential point which is closely related with marrow and diseases in marrow. According to oriental medical theory, marrow & bone, growth & development are governed by the kidneys. Therefore we hypothesized that GB39 might be effective to promote growth especially in height.

ST36 is the sea point and self-natured point stomach meridian. Stomach and its meridian, in conjunction with spleen, supplies the body with essential substances. consequently nourishing the body and enriching the flesh. ST36, which is a self-natured point(earth point of earth meridian), benefits stomach and spleen, tonifies gi and blood, strengthens the body, regulates nutritive and defensive qi etc. 3 Additionally, Ginseng enters spleen, and tonifies spleen and stomach1) therefore we hypothesized that ST36 may also be effective to promote growth especially in mass and weight. So, we expect that the herb(Cinseng) and the point(ST36) of Ginseng herbal acupuncture at ST36will correspond and make it even more effective.

II. Material and Method

1. Materials

1) Animals

5weekold female ICR mice were purchased from Deahan Biolink (Chungbuk, Korea). The mice were maintained under conventional conditions in constant temperature (22±2°C), humidity (55±5%), and ventilation, on a 12 hrs light/dark cycle. Mice had feed food and water ad libitum.

2) Medicinal substance

Ginseng Radix was obtained from Oriental Hospital of Deajeon university.

3) Herbal-acupuncture solution

The herbal-acupuncture solution was made according to a standard protocol. Briefly, 55g of Ginseng Radix was ground by mixer. The powder was transferred into a flask and 500ml of distilled water was added to it, and shaken for 3h at 37°C in a shaking incubator. The extracted liquid was filtered twice with 3M paper and concentrated by Rotary evaporator. 30 ml of 95% ethanol was added to the concentrate and filtered. 30 ml of 85% ethanol was added to the filtrate and filtered again. Sequentially, 30 ml of 75% ethanoladded and filt liquid was adjusted to pH 7.0 and sterilized.

2. Methods

1) Experimental Procedure

Mice were divided into four groups (n=10 in each group). The mice in G-ST36 group were injected with 1% Ginseng Radix(100µl/mouse) solution at right ST36(Joksamni) which is located on the anterior lateral side of the lower

limb, three body inches below the knee on the muscle belly of anterior tibialis m. Those in G-GB39 group were injected with the same amount of Ginseng Radix solution at right GB39(Hyeonjong) which is located on the lateral side of the lower limb. 3 body inches above the tip of the lateral malleolus, on the anterior border of the fibrala. On the other hand, the control groups were injected with 100 saline ST36(Joksamni) of at GB39(Hyeonjong). According the to proportional measurement the distance between the popliteal crease and the lateral malleolus is considered to be 16 bodyinches. The injections were performed 3 times a week for 3 weeks. On the twenty-third day, the experimental animals were anesthetized with ether and sacrificed.

2) Measurement of the length.

Heights were measured by observations of total height, body length and tail length, twice a week. The total height means the length from the head to the tail. Body height means the length form head to the junction of the body and the tail.

3) Measurement of the weight and the protein efficiency ratio

The body weight was measured twice a week. The protein efficiency ratio is the ratio of weight increase to protein intake.

4) Measurement of IGF-I by Enzyme Immunoassay (EIA)

Blood was taken from individual mouse from the heart after anesthesia by ether inhalation. Serum was obtained by centrifugation for 20 min at 15000rpm, and stored at -20°C until further analysis.

Serum IGF-I levels were measured using an mouse IGF-I EIA kit (DSL Inc, USA) and performed according to the manufacturer's protocol.

5) Statistical analysis

Statistical analysis was performed using Student's t-test and the results were expressed as mean ± S.E.M. P value < 0.05 was considered to be statistically significant.

III. Results

1. Measurement of Body Weight

The body weights of all mice groups increased until the 11th day but there were no

Table 1. Effect on Protein EfficiencyIncrease in Mice

Protein efficiency ratio									
Group	No. of mice	3 day	7day	11day	14day	18day			
Control-GB39	10	0.57±0.05	0.37±0.07	0.22±0.05	-0.08±0.06	-0.08±0.08			
G-GB39	10	0.46±0.05	0.61±0.09*	0.56±0.06*	0	0			
Control-ST36	10	0.24±0.04	0.6±0.05	0.34±0.06	0.17±0.04	0.17±0.05			
G-ST36	10	0.34±0.06	0.87±0.06**	0.16±0.04	0.01±0.01	0.01±0.01			

Control-GB39 saline injection at right GB39

G-GB39: Herbal acupuncture with Ginseng Radix soultion at right GB39

Control-ST36: saline injection at right ST36

G-ST36: Herbal acupuncture with Ginseng Radix soultion at right GB39

Protein Efficiency ratio is body weight increase/protein intake

Values are means ± S.E.M.

^{*}P<0.05, **P<0.01 compared to control groups

statistical differences between the control groups and the herbal acupuncture groups. (Fig. 1)

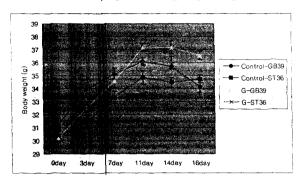


Fig. I. Effect an Body Weight Increase in Mice

Control-GB39: saline injection at right GB39 Control-ST36: saline injection at right ST36

G-GB39: Herbal acupuncture with Ginseng Radix coulting at right GB39

soultion at right GB39

G-ST36: Herbal acupuncture with Ginseng Radix

soultion at right GB39

Values are means ± S.E.M.]

Measurement of the Protein Efficiency ratio

The protein Efficiency ratio of G-GB39 was significantly(p<0.05) higher than that of control-GB39 on day 7 and 11, and that of G-ST36 was significantly(p<0.01) higher than that of control-ST36 on day 7(Table 1).

3. Measurement of the Total Length

The total length of G-GB39 was significantly higher than control-GB39 on day 7, 11 and 14. And the total length of G-ST36 was significantly higher than control-ST36 on day 14 and 18. (Fig. 2)

4. Measurement of the Body Length

The body length of G-GB39 was significantly higher than control-GB39 on day 7 and 14. The body length of G-ST36 was significantly higher than control-ST36 on day 14 and 18. (Fig. 3)

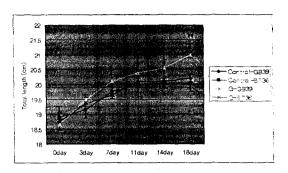


Fig. 2. Effect on Total Length Increase in Mice

Control-GB39: saline injection at right GB39 Control-ST36: saline injection at right ST36

G-GB39: Herbal acupuncture with Ginseng Radix soultion at right GB39

G-ST36: Herbal acupuncture with Ginseng Radix soultion at right GB39

Values are means ± S.E.M.

*P<0.001, **P<0.01 compared to control-GB39 group

#P<0.51 ##P<0.001compared to control-ST36 group

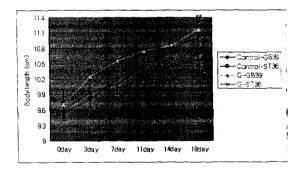


Fig. 3. Effect on Body Length Increase in Mice

Control-GB39: saline injection at right GB39 Control-ST36: saline injection at right ST36

G-GB39: Herbal acupuncture with Ginseng Radix soultion at right GB39

G-ST36: Herbal acupuncture with Ginseng Radix soultion at right GB39-

Values are means ± S.E.M.

*P<0.05 compared to control-GB39 group #P<0.05, ##P<0.01compared to control-ST36 group

5. Measurement of the Tail Length

The tail length of GrGB39 was higher than control GB39 overall but there was no

Table 2. Effect on TailLength ase in Mice Incre

Tail length increase (cm)										
Group	No. of mice	3 day	7day	11day	14day	18day				
Control-GB39	10	0.25±0.01	0.15±0.01	0.1±0.01	0	0				
G-GB39	10	0.3±0.01	0.3±0.01	0.1±0.01	0.5±0.01	0.1±0.01				
Control-ST36	10	0	0.2±0.02	0	0.05±0.01	0				
G-ST36	10	0	0.4±0.01*	0.3±0.01*	0	0.1±0.02				

Control-GB39: saline injection at right GB39

G-GB39: Herbal acupuncture with Ginseng Radix soultion at right GB39

Control-ST36: saline injection at right ST36

G-ST36: Herbal acupuncture with Ginseng Radix soultion at right GB39

Values are means ± S.E.M.

*P<0.05 compared to control groups

statistical significance. The tail length of G-ST36 was significantly higher than control-ST36 on day 7 and 11. (Table 2)

6. Measurement of IGF-I

The serumIGF-I level of G-GB39 group was significantly(p<0.05) higher than that of

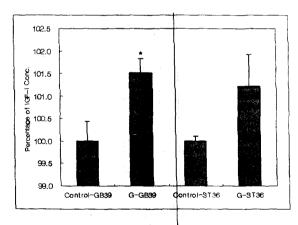


Fig. 4. Effect on IGF+I level in mouse serum

Control-GB39: saline injection at right GB39

G-GB39: Herbal acupuncture with Ginseng Radix coulties at right CP30

soultion at right GB39

Control-ST36: saline injection at right ST36

G-ST36: Herbal acupuncture with Ginseng Radix soultion at right GB39

Serum IGF-I level was measured by Enzyme Immunoassay (EIA).

Values are means ± S.E.M.

*P<0.05 compared to control groups

control-GB39 group. The serum IGF-I level of G-S36 group was higher than that of control-ST36 group but there was no statistical significance. (Fig. 4)

IV. Discussion

In this study, we investigated the effect of acupuncture with Ginseng solution at GB39 and ST36 on growth in mice. Regardingthe increase of mouse body weight, we could not find any significant difference between the experimental groups, but the efficiency ratio of G-GB39 protein G-ST36 were significantly higher than those of control-GB39 and control-ST36 respectively. These results indicate that Ginseng herbal acupuncture at GB39 and ST36 are both effective to promote growth. We expected to find that ST36 would be more effective for mass increase than GB39, but there was no significant difference between GB39 groups and ST36 groups.

In the increase of the total length, body length and tail length, G-GB39 and G-ST36 were significantly higher than control-GB39

and control-ST36 respectively, indicating that both Ginseng herbal acupuncture at GB39 and ST36 are effective to promote growth in height.

Growth hormon(GH) regulates growth directly through the GH receptor and indirectly by stimulating IGF-I expression⁴. IGF-I is considered to be involved in GH effects on chondrocytes, osteroblasts and osteoclasts⁵⁾.

In this study, Ginseng herbal acupuncture at GB39 increased serum IGF- I level significantly compared to control-GB39. The serum IGF- I level in G-ST36 was also higher than control-ST36 but there was no statistical significance. This result indicates that, even though the increase of length of G-GB39 and G-ST36 had no significant difference, GB39 may be related with growth especially in height by way of increasing IGF- I, and may be more effective than ST36 in growth of height or bone length.

V. Conclusion

Ginseng herbal acupuncture at GB39 and ST36 are both effective to promote growth. In addition, It appears that GB39 is effective for

growth by way of increasing IGF-I, and more closely related to growth in length than ST36.

VI. References

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