

The Change of Serum Gastrin Concentration with Oculo-acupuncture in Dogs

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Abstract : This study was performed to clarify the effect of oculo-acupuncture (OA) on change of serum gastrin concentration. Twenty-three clinically healthy mongrel dogs were used in this study. This study was comprised of two experiments. In experiment 1, the effect of OA (stomach/spleen region) with comparison of non-acupoint was examined. On the other hand, the effect of OA (stomach/spleen region) with comparison by body acupuncture (BL-21) was also examined in experiment 2. Serum gastrin levels were measured on before, 0, 30, 60 and 120 minutes after treatment in both non-acupoint and stomach/spleen region groups, and BL-21. The results showed that significant decreases in serum gastrin level was detected on 30 mins ($p < 0.05$) after OA when compared with that of non-acupoint group. Serum gastrin concentrations with OA at stomach/spleen region group were significantly decreased on 60 mins ($p < 0.05$) and 120 mins ($p < 0.01$) compared with those of the AP at BL21 group. In conclusion, it was demonstrated that OA at stomach/spleen region had significantly decreased serum gastrin levels in dogs and OA at stomach/spleen region decreased serum gastrin concentration more than that body acupuncture at BL-21.

Key words : Serum gastrin concentration, oculo-acupuncture, BL-21, stomach/spleen region.

Introduction

It is known that acupuncture (AP) treatments including needle-AP, injection-AP, electro-AP, auriculo-AP and laser-AP have therapeutic effects in human and animal patients with a variety of disease processes (8).

Oculo-acupuncture (OA) is a novel form of needle-AP used to treat diseases by applying AP at acupoints in the periocular tissues. It has been previously described in human clinical practice and employed mainly in China (13). As mentioned in a classical ancient medical book in China, "All meridians are connected with eyes". Therefore, the eyes are closely related to the internal organs and meridians, and AP applied at the acupoints around eye can be used to treat many diseases of the whole body (13). OA had positive therapeutic effects in human diseases including vomiting, acute muscular sprain, apoplexy, hemiplegia and pain (13). Research regarding the therapeutic effect with OA has been done mainly in human clinical practice, however, there are no reports about OA in veterinary clinical practice until now.

Gastrin, secretin and cholecystokinin (CCK) have major roles in stimulating gastric acid secretion, pancreatic fluid,

bicarbonate secretion, gastric muscular contraction and pancreatic enzyme secretion. Gastrin is found in gastrointestinal endocrine cells, released by physiologic stimuli, circulated in blood and evokes a biologic response. The release of gastrin is under direct cholinergic control (5).

Regarding the changes of endocrine hormones by AP, Kim and Choi (2) reported that electro-AP stimulation with high-voltage and high-frequency decreased blood concentration of gastrin in 6 horses. Zhou and Zhuo (15) reported that the concentration of gastrin in serum and antral mucosa of dogs was increased after AP. In addition, Liu *et al.* (3) reported that gastrin concentrations in the G cells after AP at CV12, PC6 and ST36 were decreased in patients with duodenal ulcer, as compared with that before AP.

This study was designed and performed in order to clarify the change of serum gastrin concentration by OA and to compare the difference between OA and body AP in the change of serum gastrin concentration.

Materials and Methods

Experimental animals and grouping

A total of 23 clinically healthy mongrel dogs (1-4 years old and 3-15 kg) were evaluated in the present study. The experimental dogs fed a standard commercial diet (PETIA

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Co., Korea) twice daily (each 400 g) and water *ad libitum*. Evaluations were performed two hours after feeding for all dogs. Eleven mongrel dogs were used for OA study. They were divided into two groups; non-acupoint group (n=5) and Stomach/Spleen region group (n=6), respectively. In addition, 12 mongrel dogs were also used for comparison between OA and body AP. They were divided into two groups; BL21 group (n=6) was stimulated by body AP and stomach/spleen region group (n=6) was stimulated by OA. The present study was performed according to the rules of the ethics Committee for Experimental Animal, Chungnam National University.

OA and body AP

Stomach/spleen region was used in OA of the present study according to the method of human OA (Fig 1). Non-acupoints located about 1 cm above from both orbicularis oculi muscle were used and a needle (Hwato®, Suzhao Medical Appliance Factory, China) was horizontally inserted approximately 13 mm to outside directions in the non-acupoint group. In stomach/spleen region group, the OA needles were obliquely inserted 13 mm from BL1 to ST1 direction in both eyes. Needling was maintained for 20 minutes in all groups. In addition, both BL21 (both tips of the transverse process of the first lumbar vertebrae) was used for body AP and acupoint detector (Ittorator, Ito Electric Co., Japan) was used for correct detection of BL21.

Blood samples

Three milliliters of blood was collected from the cephalic vein of all dogs at before and at 0, 30, 60 and 120 minutes after OA and body AP, respectively. The sera were separated from coagulated blood by centrifugation (150 g for 15 min) and were used to determine the gastrin concentration.

Hormone analysis

Serum gastrin concentration was measured according to the double antibody gastrin radioimmunoassay kit (Diagnostic Product Corporation, USA) using COBRA 2 gamma counter (PACARD Co., USA).

Statistical analysis

Significance of differences among groups was tested with a paired student's *t*-test by Microsoft Excel (Microsoft Corp., USA). The criterion for significance was value of $p < 0.05$.

Results

The changes of serum gastrin concentration by OA

The serum gastrin concentration (pg/ml) was 8.3 ± 2.9 , 8.3 ± 2.9 , 3.3 ± 1.7 , 1.9 ± 2.0 and 1.8 ± 1.7 on before, 0, 30, 60 and 120 minutes in the non-acupoint group, respectively. They showed continuously decreasing pattern after OA by comparison with that of before (Fig 2). On the other hand, the serum gastrin concentration (pg/ml) in Stomach/Spleen region group was 7.4 ± 1.1 , 6.3 ± 2.5 , 1.2 ± 0.7 , 2.5 ± 1.9 and 2.5 ± 2.0 on before, 0, 30, 60 and 120 minutes after OA, respectively. However, they were lower levels than those of non-acupoint group. A significant difference was detected on 30 min ($p < 0.05$) after OA compared with that of non-acupoint group.

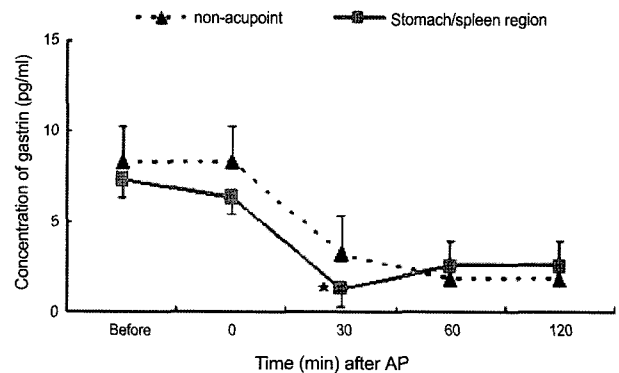


Fig 2. Comparison of the change of serum gastrin concentration between non-acupoint and Stomach/Spleen region groups by OA. *: Significance between non-acupoint and stomach/spleen region group ($p < 0.05$).

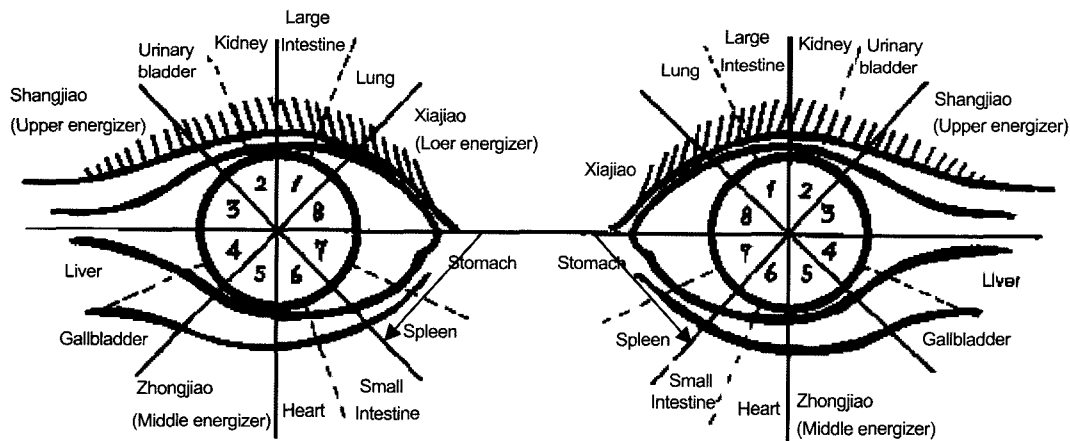


Fig 1. Regions of AP at oculo-acupoints and needle insertion direction (arrow).

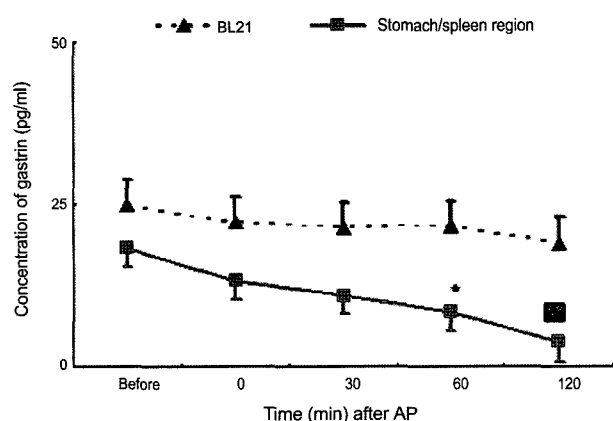


Fig 3. Comparison of the changes of serum gastrin concentration between BL21 and Stomach/Spleen region groups.

*: Significance between BL21 and stomach/spleen region group ($p < 0.05$).

**: Significance between BL21 and stomach/spleen region group ($p < 0.01$).

Comparison of serum gastrin concentration between OA and body AP groups

Serum gastrin concentration (pg/ml) was 24.9 ± 12.1 , 22.2 ± 9.3 , 21.3 ± 8.6 , 21.5 ± 6.5 and 19.0 ± 8.1 on before, 0, 30, 60 and 120 minutes after AP, respectively in BL21 group and they showed gradual decreasing pattern by comparison with that of before (Fig 3). On the other hand, serum gastrin concentrations (pg/ml) were 18.3 ± 13.4 , 13.2 ± 10.7 , 11.0 ± 10.1 , 8.4 ± 7.8 and 4.0 ± 3.7 on before, 0, 30, 60 and 120 minutes in stomach/spleen region group, respectively. They revealed similar decreasing pattern by OA to BL21 group. Significant differences were detected in stomach/spleen region group on 60 min ($p < 0.05$) and 120 min ($p < 0.01$) compared with that of BL21 group.

Discussion

OA therapy is used to treat diseases by applying AP at the periorbital acupoints. After a diagnosis is made by observing the minute changes of blood vessels on conjunctiva of the eye, the acupoints around the eyes are selected according to the principle of differential diagnosis and treatment in traditional Chinese medicine to treat various diseases, such as acute muscular sprain, early stage of apoplexy (stroke) with hemiplegia and pain (13). OA acupoints were divided into 8 regions and defined as 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th (See Fig 1). A horizontal line and a vertical line were drawn across the pupil to divide the eye into 4 quarters and then they were again divided into 8 equal sectors to define the oculo-acupoints (13).

In previous reports about the effect of OA, Shi and Zheng (9) reported that 40 cases of sudden deafness had improvement with ocular needling and Zhao (12), Xiao (10) and Yang (11) reported that OA was used to treat multiple cases with severe vomiting. In China, OA has been mainly used in a variety of

clinical in human patients. In this study, the changes of serum gastrin concentration after OA (stomach/spleen region) were gradually decreased in healthy mongrel dogs and a significant decrease was detected on 30 min after treatment when compared with that of non-acupoints group. These results suggest that OA at stomach/spleen region may decrease serum gastrin concentration in healthy dogs. On the other hand, the non-acupoints group showed a similar decrease in serum gastrin levels when compared to OA at stomach/spleen region group. For this reason it was assumed that non-acupoints about 1 cm above from both orbicularis oculi muscle were closely located with upper OA acupoints and had slightly affected another upper oculo-acupoint such as the large intestine or kidney regions. Additional studies evaluating the effect of AP at other oculo-acupoints should be performed in the future.

Gastrin is found in gastrointestinal endocrine cells, released by physiologic stimuli (e.g. feeding), circulates in blood, binds to cell receptors at distant sites and evokes a biologic response. Cells that synthesize and release the gastrin peptides (G-cells) are numerous in the antrum of the stomach of the dog (5). Feeding is an important regulator of gastrin release and the regulation of circulating gastrin concentration follows a basic feedback inhibition loop (1).

Gastric secretion occurs in three phases, the cephalic, gastric and intestinal phase. The cephalic phase is mediated entirely by the vagus nerve by direct cholinergic stimulation of the parietal cells, cholinergic release of gastrin from the antral G cells, and increase in responsiveness of the parietal cells to gastrin. The most significant biologic action of gastrin is to stimulate gastric acid secretion by gastric oxyntic (parietal) cells (5). When gastrin concentrations rise, the gastric parietal cells are stimulated to secrete acid but when the antral mucosa is bathed in acid, further release of gastrin is inhibited (1).

As for the changes of endocrine hormones by AP treatment, electro-AP stimulation with high-voltage and high-frequency decreased gastrin concentration in blood (2) and repeated massage-like stroking on the ventral side of the abdomen similar to AP stimulation decreased plasma levels of gastrin, insulin and somatostatin (7). Gastrin in the G cells was decreased in patients with duodenal ulcer after AP (3) at CV12, PC06 and ST36, while plasma gastrin concentration was increased by AP at ST36, PC6 and BL21 (14). Most of reports about change of endocrine substances by AP have focused on the use of body acupoints, however, there are no reports about the change of serum gastrin concentration by AP at oculo-acupoints until now.

In the dog, serum gastrin levels increase to a peak of about 160 pg/ml after eating (1). Gastrin concentrations at pre-stimulation in OA and body AP groups were 24.9 ± 12.1 pg/ml and 18.3 ± 13.4 pg/ml, respectively and both groups were in normal range. Serum gastrin concentration followed a similar decreasing pattern in both OA, Stomach/spleen region group and body AP at BL21 in this study. The result of serum gastrin concentration by body AP at BL21 in dogs was similar to the results reported by Kim and Choi (2) that serum gastrin levels were increased by electro-AP (2 V, 5 Hz) stimulation

on the BL21 in horses. These results could not be compared directly with other reports concerning about change of serum gastrin concentration by OA because the change of serum gastrin concentration by OA has not been examined until now. Both OA at Stomach/Spleen region group and body AP at BL21 group were similar to results of repeated massage-like stroking on the ventral side of the abdomen by Sarah *et al.* (7) and those in patients with duodenal ulcer after AP at CV12, PC6 and ST36 by Liu *et al.*(3), as well as those in rats stimulated with electro-AP by Quyang *et al.* (6). In this study the needle was inserted from BL1 in the stomach/spleen region. Accordingly, it was assumed that the Stomach/Spleen region might play a role in change of serum gastrin concentration due to activation of related BL meridians. Therefore, it was assumed that the decrease in serum gastrin concentration might be caused by inhibition of gastric phase rather than gastrin release controlled in the cephalic phase and intestinal phase during feeding.

Significant differences were detected in OA group at 60 min ($p < 0.05$) and 120 min ($p < 0.01$) as compared with that of body AP group in the present study. It was shown that serum gastrin concentration by OA at stomach/spleen region was more affected than that by body AP at BL21. A reason for this difference was not clear in the present study, however, additional studies should be performed to clarify whether canine oculo-acupoints are identical to human eye acupoints in the future.

Considering above findings, it was concluded that OA at stomach/spleen region decreased the serum gastrin concentration and the OA at stomach/spleen region decreased serum gastrin concentration more than that by body AP at BL21.

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개에 있어서 眼針에 따른 혈청 가스트린 농도의 변화

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요 약: 본 연구는 혈청 가스트린 농도 변화에 대한 안침의 효과를 규명하기 위하여 수행되었다. 임상적으로 건강한 23두의 잡종 견을 본 실험에 사용하였다. 본 연구는 두 가지 실험으로 구성되었다. 실험 1에 있어서는 안침의 효과(위 및 비장 영역)를 눈의 비 혈위 영역을 대조로 조사하였다. 한편 실험 2에 있어서는 안침의 효과(위/ 비장 영역)와 胃俞(BL-21)에 대한 體針의 효과를 비교하였다. 혈청 가스트린 농도의 변화는 0, 30, 60 및 120분에 대조 군 및 안침 군에서 각각 조사하였다. 그 결과, 안침 군에 있어서 안침 (위/비장 영역) 적용 후 30분에 대조 군에 비하여 혈청 가스트린 농도의 유의한 감소 소견이 인정되었다($p < 0.05$). 또한, 안침 군의 혈청 가스트린 농도는 BL-21 체침 군에 비하여 60분 및 120분에 각각 유의한 감소 소견을 나타내었다($p < 0.01$). 이상의 결과를 종합해 볼 때, 개에 있어서 안침(위/비장 영역)은 혈청 가스트린 농도를 감소시키며, 또한 BL-21 體針 보다 혈청 가스트린 농도를 더욱 감소시키는 것으로 판단되었다.

주요어: 혈청 가스트린 농도, 안침, 체침, 위/비장 영역