Gum Arabic Enhances Intestinal Calcium Absorption in Rats

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We investigated whether the efficiency of intestinal calcium (Ca) absorption was improved by concomitant ingestion of gum arabic (GA) in rats. We used the Ussing chamber method to clarify the effect of GA on upper and lower small intestinal absorption of Ca. Increased *in vitro* Ca permeation was observed in rats who ingested water with 7.5% GA for 10 days. These results suggested that administration of GA with Ca could increase the efficiency of oral Ca absorption.

Key words —— gum arabic, calcium, absorption, rat, intestine

INTRODUCTION

Calcium (Ca) is the fifth most abundant element in the body, with the major portion in the bones. It has important physiological roles. It is essential for the functional integrity of nerves and muscles, where it has a major influence on excitability and the release of neurotransmitters. It is necessary for muscle contraction, cardiac function, maintenance of membrane integrity, and blood coagulation. To carry out these various roles, Ca must be available to the appropriate tissues in the proper concentrations. Shortage of Ca induces some bone diseases such as osteoporosis and osteomalacia.[1–3] It is therefore important to have an appropriate Ca intake so as to maintain Ca homeostasis in the body.

The process of Ca transport across cell membranes involves numerous transporters.[4–6] The efficiency of intestinal Ca absorption is determined by its availability in the diet, its solubility in the gut, and the net capacity to absorb it across the intestinal wall. In this study, we used gum arabic (GA) as an additive agent to modify the efficiency of Ca absorption. GA is a branched polymer of galactose, rhamnose, arabinose and glucuronic acid.[7,8] It has been reported that the addition of GA to sodium-L glucose oral rehydration solution enhanced the effectiveness of water and electrolyte absorption in normal rats due to morphologic changes in the intestinal villi.[9,10] Therefore, we assumed that the absorption of ions such as Ca, magnesium and zinc would also be affected by the use of GA. The objective of the present study was to elucidate whether GA improves the efficiency of Ca absorption.

To assess the effects of GA on intestinal Ca absorption, we used the *in vitro* Ussing chamber method with rat intestine. The efficiency of intestinal Ca absorption was significantly improved by pretreatment with 7.5% (w/v) GA in drinking water for 10 days. This study shows that GA is a useful additive for promotion of intestinal Ca absorption.

MATERIALS AND METHODS

Chemicals —— Sudanese GA (resin of *Acacia senegal*) supplied by Auron Investments Ltd. (Dubai, U.A.E.) was purified by Sankyo Food Industry Co., Ltd. (Saitama, Japan). The GA was dissolved in water and filtrated, and the filtrate was spray-dried at about 120°C to obtain the GA used in this experiment. Ca L-lactate was purchased from Sigma (St. Louis, MO, U.S.A.).

Animals —— Six- to eight-week-old male Wistar rats weighing 200–250 g were purchased from Japan SLC Co. (Shizuoka, Japan). The animals were housed in a temperature- and humidity-controlled room and were allowed free access to standard laboratory chow and water. The experiments were approved by the Committee for the Care and Use of Laboratory Animals at Kinki University School of Pharmaceutical Science.

Determination of Intestinal Permeability Using the Ussing Chamber Method —— We determined intestinal permeability using the Ussing chamber method in rats pretreated with free-feeding drinking water containing 1%, 7.5% or 15% GA for 10 days. The rats were anesthetized with 50 mg/kg intraperitoneal sodium pentobarbital, and a segment of upper and lower small intestine was removed. The tissue was rapidly stripped of serosal muscle layers, and

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mounted vertically in an Ussing chamber that provided an exposed area of about 2.0 cm^2. The volume of bathing solution on each side was 10 ml, and the temperature was maintained at 37°C. The transport medium consisted of 125 mM NaCl, 4.0 mM KCl, 6.0 mM L-glutamine, 30 mM HEPES (pH 7.4), and was gassed with 95% O_2 + 5% CO_2 before and during the transport experiments. Ca L-lactate solution [1 mg (Ca^{2+})/ml] was added to the mucosal chambers. Samples were collected at 0, 10, 30, 60 and 90 min. The Ca concentrations in the mucosal chambers were determined by Osaka Kessei Research Laboratories (Osaka, Japan).

Measurement of Urinary Excretion of Ca —— Rats were placed in a metabolism cage for 3 days and given free access to food and water. They were fed a Ca free diet (Oriental Yeast Co., Tokyo, Japan) to remove the effect of normal dietary Ca on the urinary excretion of Ca. Each rat was fed 40 ml free-feeding drinking water containing 7.7 mg Ca L-lactate (1 mg Ca^{2+})/ml only, Ca with 7.5% GA, or 7.5% GA only. Urine was collected for 3 days, and the volume and Ca level of each urine specimen were measured by Osaka Kessei Research Laboratories.

RESULTS AND DISCUSSION

We investigated whether GA affected intestinal Ca absorption *in vitro* using the Ussing chamber method (Fig. 1). In the rats who ingested water with 1%, 7.5% or 15% GA for 10 days, the absorption of Ca was higher in both the upper and lower small intestine compared to control rats, and ingestion of 7.5% GA showed a significantly higher absorption than the other concentrations. The permeation clearance in rats pretreated with 7.5% GA solution was 2.5- to 3.5-fold that of control rats. Ca^{2+} is absorbed in the small intestine via active and passive pathways.[4–6] In the duodenum and upper jejunum, Ca^{2+} absorption involves active transport. The effect of GA is not specific to a particular part of the small intestine, because promotion of Ca absorption was observed in both the upper and lower small intestine (Fig. 1). It is possible that similar effects might be observed in the upper and lower small intestine due to saturation of active transport in the upper part at a high GA concentration. It appeared that 7.5% GA gave the best results of the concentrations tested. A 15% GA concentration would be too high for optimum Ca absorption. This biphasic effect of GA concentration on intestinal Ca absorption may be due to interaction between some components of GA and Ca^{2+}.

Next, we investigated Ca absorption in the presence or absence of 7.5% GA. Without pretreatment with GA, there was little difference in Ca absorption with or without 7.5% GA in the upper or lower small intestine (data not shown), suggesting that pretreatment with GA was necessary for expression of the effect on intestinal Ca absorption. It is possible that expansion of the basolateral intercellular spaces between villus absorptive epithelial cells and the lamina propria by GA[9, 10] could be involved in the improved Ca absorption. Further studies are needed to clarify the precise mechanisms of the effect of GA on Ca absorption. It is necessary to clarify the effects of the pretreatment period on intestinal Ca permeability.

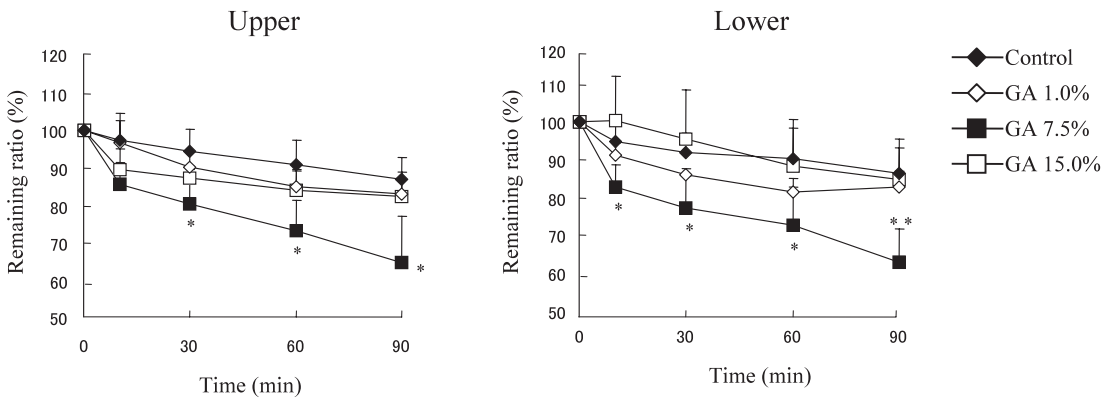

Fig. 1. Absorption of Ca Across Isolated Rat Intestinal Tissues

The effect of GA on the Ca permeation was evaluated using the Ussing chamber method, from the mucosal to the serosal side. Rats were fed water with 1%, 7.5% or 15% GA for 10 days. Each value represents the mean ± S.D. of four to six rats. $^{*}p < 0.05$, $^{**}p < 0.01$, significant difference between the values for control and 7.5% GA.

The blood concentration of Ca does not usually reflect the amount of Ca absorbed because Ca levels are kept in the normal range by homeostatic mechanisms. The oral administration of Ca L-lactate did not change the blood concentrations of Ca with or without GA (data not shown). Therefore, we investigated the levels of urinary Ca excretion as an indicator of the amount of Ca absorbed after ingestion of Ca L-lactate with or without GA. The cumulative amounts of Ca excreted in urine over 3 days in rats who ingested Ca L-lactate with 7.5% GA (0.708 ± 0.083 mg) were significantly higher than in rats who ingested Ca L-lactate alone (0.297 ± 0.072 mg) or 7.5% GA alone (0.131 ± 0.111 mg). This result suggests that the efficiency of Ca absorption was low after administration of oral Ca alone, but absorption was significantly increased after administration of Ca L-lactate with 7.5% GA. Retention of Ca in the small intestine might be also improved by GA. This result indicates that intestinal Ca absorption is promoted by GA *in vivo* as well as *in vitro*. Further *in vivo* experiments are needed to validate this result.

In this study, the efficiency of intestinal Ca absorption in rats was promoted by use of GA. Ca L-lactate was used as the Ca salt because: it has relatively high solubility (9600 mg/100 ml H_2O); it has a minor effect on the taste of food; and it contributes to gelation and form retention by cross linking with pectin or alginic acid. It is unclear whether GA exerts a similar effect on the absorption of other Ca salts, but it is known that Ca L-lactate has superior Ca absorption. We assume that the use of GA is practical because GA has been shown to be very safe for use as a food additive as an emulsifier or stabilizer. The precise mechanism of promotion of Ca absorption still needs clarification.

In conclusion, this study showed that the efficiency of Ca absorption was improved by GA. It is possible that GA could be used to produce more efficient Ca supplements.

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