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RESEARCH ARTICLE

Enhancing effects of chitosan and chitosan hydrochloride on intestinal absorption of berberine in rats

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Abstract

Berberine chloride (BBR) is a plant alkaloid that has been used for centuries for treatment of inflammation, dysentery, and liver diseases. It is poorly absorbed from the gastrointestinal (GI) tract and its various clinical uses are limited because of its poor bioavailability. The object of the present study was to investigate the absorption enhancing effect of chitosan on BBR. Mixtures of BBR and chitosan were prepared and the absorption enhancement was investigated in rats. The results showed a dose-dependent absorption enhancement produced by chitosan. Formulations containing 0.5%, 1.5%, and 3.0% chitosan resulted in improvement of $AUC_{0-36\ h}$ values by 1.9, 2.2, 2.5 times. The absorption enhancing ability of chitosan may be due to its ability to improve the BBR paracellular pathway in the intestinal tract. Chitosan hydrochloride, a salt of chitosan, was also investigated in this study. However, the addition of 2.0% and 3.3% chitosan hydrochloride to BBR solution did not produce any increase in either C_{max} or $AUC_{0-36\ h}$ of BBR. Subsequent solubility studies suggested that the reduced berberine chloride solubility in chitosan hydrochloride may limit the enhancement ability. This study showed that the optimum formulation producing the highest BBR absorption is the BBR solution containing 3.0% chitosan.

Keywords: Berberine chloride, chitosan, chitosan hydrochloride, absorption enhancer, histopathological evaluation

Introduction

Berberine chloride (BBR, Figure 1) is a well-known plant alkaloid isolated from medicinal herbs, such as *Coptis chinensis* and *Berberis aristata*, and it has been used for centuries in traditional Chinese and Ayurvedic medicine to treat inflammation, dysentery, and liver diseases[1,2,3]. Recent research has shown that BBR has many other biological effects, such as hypolipidemic, hypoglycemic, antiarrhythmic, antiproliferative and antineoplastic activities[4–8]. As a traditional botanical drug, BBR has recently aroused great interest because of its variety of bioactivities, low toxicity, and low cost. However, pharmacokinetic studies have demonstrated that BBR has a very low bioavailability and its clinical application is greatly limited due to poor absorption[9]. It is thought that the effective dosage of BBR as a hypolipidemic or hypoglycemic drug is so large that concomitant side effects may occur, especially during long-term administration[10]. Therefore, it is necessary to improve the absorption of BBR and develop more efficient BBR formulations to provide alternative treatments.

BBR is a hydrophilic compound with a temperature-dependent aqueous solubility which increases with an increase in temperature. It is found to be stable at different pH and temperature conditions over 6 months[11]. Tests we carried out earlier show that BBR has a log P value of –1.0 in the octanol-water system and this is comparable with that reported by Battu et al.[11], indicating that BBR is a biopharmaceutical classification system (BCS) Class III drug. Drugs belong to BCS Class III are mostly lipophobic with poor membrane permeability. In order to enhance the bioavailability of drugs such as BBR, the

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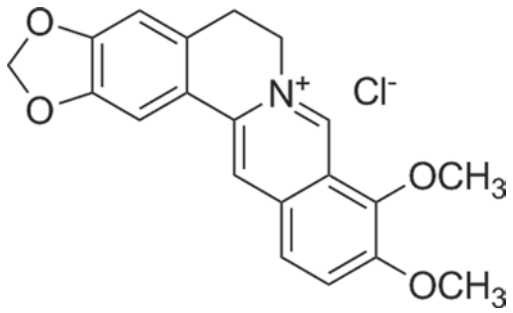

Figure 1. The chemical structure of berberine chloride.

permeability through intestinal membranes must be improved. It is considered that absorption of hydrophilic compounds is mostly limited to the paracellular pathway. The paracellular route is considered as an aqueous pathway along the intercellular space between adjacent cells and is restricted by tight junctions at the most apical part of the cells[12]. The aqueous nature of the paracellular pathway makes it favorable for small hydrophilic solutes. Manipulating the paracellular permeability has been proposed and used to enhance drug absorption[13,14]. Compounds capable of influencing the paracellular pathway are believed to be able to affect the paracellular permeability of drugs.

Chitosan is a commercially available polymer obtained by alkaline deacetylation of the natural polysaccharide chitin. It has been reported to be capable of enhancing drug absorption via the paracellular route[12]. Chitosan products have different degrees of deacetylation and molecular weights, and those properties are related to its absorption-enhancing ability. Several studies have been conducted to investigate the mechanism whereby chitosan enhances the permeability of epithelia. Chitosan, as a cationic macromolecule, can interact with the anionic components of the glycoproteins on the epithelial cell surface. This interaction may alter the relative concentration of ions within the hydrated interior channels of tight junctions, and result in a relaxation and opening of the tight junctions. Furthermore, it has been found that chitosan affects the permeability of tight junctions by inducing changes in the cytoskeletal F-actin distribution which is associated with the proteins in the tight junctions[15]. Chitosan has attracted great attention as a permeation enhancer not only due to the enhancing properties but also due to the high molecular weight, and as it is not absorbed from the GI tract it produces no systemic side effects. In addition, due to its biodegradability, biocompatibility, and FDA approval, chitosan is of great interest as an excipient for use in drug formulations[16–18].

In the present study, chitosan was studied as an absorption enhancer to improve the GI absorption of BBR in rats. Chitosan hydrochloride, a salt of chitosan, was also investigated in this study. Histopathological evaluation after using chitosan and chitosan hydrochloride was performed in order to investigate the local toxicity of chitosan and chitosan hydrochloride in the intestinal tract.

Materials and methods

Materials

BBR was purchased from Northeast Pharmaceutical Group Co., Ltd. (Shenyang, PR China). The internal standard, tetrahydropalmatine, was purchased from the National Institute for the Control of Pharmaceutical and Biological Products (Beijing, PR China). Chitosan (deacetylation degree > 83%) and chitosan hydrochloride (deacetylation degree > 83%) were purchased from Dalian Xindie (Xiangrui) Chitin Co., Ltd. (Dalian, PR China) and Golden-Shell Biomedical Co., Ltd. (Taizhou, PR China), respectively. LC grade acetonitrile, methanol and formic acid were purchased from Dikma (New York, USA). Purified water, obtained using a Barnstead EASYpure® II RF/UV ultra pure water system (Dubuque, Iowa, USA) and passed through a 0.22 μm filter, was used throughout the study. Male Wistar rats used in the study were provided by the Laboratory Animal Center of Shenyang Pharmaceutical University.

Preparation of BBR-chitosan mixture formulation

Chitosan solutions with concentrations of 0.5%, 1.5%, and 3.0% were prepared by dissolving chitosan powder in 1.0% acetate acid solution and agitating for 2 h. A weighed quantity of BBR was added to the chitosan solutions and mixed well. The ratio (w/w) of chitosan to BBR was 0.77, 2.31, and 4.62, respectively. And the pH of the formulation solution was 3.8, 4.2, and 4.2, respectively. As chitosan hydrochloride is soluble in water, chitosan hydrochloride solutions with concentrations of 2.0% and 3.3% were prepared by direct dissolution in distilled water. The subsequent steps were the same as that of chitosan solutions. The control formulation in rat administration was prepared by adding quantitative BBR in 0.5% sodium carboxymethyl cellulose solution and mixed well. The ratio (w/w) of chitosan hydrochloride to BBR was 3.33 and 6.67. The pH of the formulation solution was both 3.8.

Animal experiments

Male Wistar rats were kept in air-conditioned animal quarters at a temperature of 22 ± 2°C and a relative humidity of 50 ± 10%. Access to water and laboratory food was ad libitum. The rats were acclimatized to the environment for 7 days and then fasted, with free access to water, for 12 h prior to the experiment. The BBR oral formulation was administered by oral gavage at a dose of 100 mg/kg. Blood samples (0.3 ml) were obtained from the retroorbital sinus before dosing and subsequently at 0.25, 0.5, 0.75, 1, 2, 3, 4, 6, 8, 12, 24, and 36 h following administration. The obtained blood was transferred to heparinized Eppendorf tubes and centrifuged at 4000 rpm for 10 min. The plasma was then transferred to clean tubes and frozen at –20°C until analysis. Five animals were used per sampling time point.

The entire experimental protocol was carried out in accordance with the University Ethics Committee

guidelines for the use of experimental animals and conformed to the Guide for Care and Use of Laboratory Animals.

UPLC-MS/MS analysis of plasma samples

Several methods have been reported for BBR determination in biological samples using LC-MS methods[19,20]. In this study, a UPLC-MS/MS method was employed for the identification and quantification of BBR in rat plasma. Liquid-liquid extraction was used for the plasma samples preparations. Aliquots of 100 µl plasma were transferred to polyethylene tubes after addition of 20 µl IS solution (10 ng/ml), followed by vortex mixing for 1 min. Then, 3 ml ether-dichloromethane (3:2, v/v) was added and the mixture was mixed for 10 min. Following centrifugation and separation, the supernatant organic layer was evaporated to dryness at 40°C in a centrifugal concentrator (Labconco Corp., Missouri, USA). The residue was reconstituted in 120 µl methanol and an aliquot of 5 µl was injected into the UPLC-ESI-MS/MS system for analysis.

Chromatography was performed using an ACQUITY™ UPLC system (Waters Corp., Milford, MA, USA) with a conditioned autosampler at 4°C. An ACQUITY UPLC™ BEH C_{18} column at 35°C (50 mm × 2.1 mm i.d., 1.7 µm; Waters Corp., Milford, MA, USA) was employed for the chromatographic separation of the analytes. The analysis was carried out with linear gradient elution using (A) water (containing 0.1% formic acid) and (B) acetonitrile as the mobile phase. The flow rate was 0.2 ml/min. The gradient conditions started with solvent A decreasing from 80% to 10% within 1.0 min and then maintained at this ratio for 1.0 min. Then, Solvent A was increased to 80% within the next 0.5 min and maintained at this level from 2.5 to 3.0 min for column equilibration. The total run time was 3.0 min. The injection volume was 5 µl and the partial loop mode was used for sample injection.

Mass spectrometric detection was carried out on a Waters ACQUITY™ TQD triple-quadrupole tandem mass spectrometer (Waters Corp., Manchester, UK) equipped with an electrospray ionization (ESI) interface in the positive ionization mode with the capillary voltage set at 0.6 kV. The cone voltage was set at 50 V. The extractor and RF lens voltages was 3.0 and 0.2 V, respectively. The temperature of the source and desolvation was 100°C and 450°C, respectively. The flow rate of nitrogen as desolvation gas and cone gas was at 450 L/h and 50 L/h, respectively. Argon was used as the collision gas at a flow rate of 0.20 ml/min for collision-induced dissociation (CID). Quantification was achieved using multiple reaction monitoring (MRM) mode. The *m/z* transitions were 336→320 for BBR and 356→192 for IS. All data collected in centroid mode were obtained using Masslynx™ NT4.1 software (Waters Corp., Milford, MA, USA). An A QuanLynx™ program (Waters Corp., Milford, MA, USA) was employed for the post-acquisition quantitative analysis. The method was validated over the concentration range of 0.1–200 ng/ml, with a lower limit of quantification of 0.1 ng/ml.

Pharmacokinetics analysis

Pharmacokinetic parameters were obtained by non-compartmental analysis using the drug and statistics (DAS) version 2.0 software (Mathematical Pharmacology Professional Committee of China, Shanghai, China). The peak concentration (C_{max}) and the time to reach C_{max} (T_{max}) were determined directly from the plasma concentration-time profiles. The area under the curve (AUC) was calculated by the trapezoidal method from the beginning of administration to the final sampling time. All values are expressed as the mean ± SD. Differences in pharmacokinetics were analyzed by the Student's *t*-test. A value of $p < 0.05$ was considered statistically significant.

BBR solubility study

The BBR equilibrium solubility in water, 1.0% acetate acid, 0.1 N HCl, chitosan and chitosan hydrochloride solutions was investigated. Chitosan and chitosan hydrochloride solutions were prepared at concentrations of 0.5%, 1.5%, and 3.0%. Excess BBR was added to the solution and shaken for 3 days in a thermostatic shaking water bath (Zhicheng, PR China). Samples were passed through a 0.45 µm microporous filter film (Xinya, PR China) and diluted to suitable concentrations for HPLC analysis. The HPLC analysis of BBR was performed using a modified version of a method previously reported[21].

Histopathological evaluation of local toxicity

The local toxicity of chitosan and chitosan hydrochloride was studied by comparing the histopathological changes in the test and control groups. The ileums of the control group, 3.0% chitosan group and 3.3% chitosan hydrochloride group were removed at 5 h after oral gavage administration of drugs and washed using saline. Segments were removed and immersed in a 10% aqueous solution of formalin. A vertical section was prepared, stained using hematoxylin-eosin and examined by light microscopy. The histopathological evaluation was performed by an experienced veterinary histopathologist.

Results

Effect of chitosan on absorption of BBR in rats

The plasma BBR concentration-time profiles of the control group and three test groups after administration BBR and formulations containing different chitosan doses to rats are shown in Figure 2. The main pharmacokinetic parameters are shown in Table 1. There was very little BBR absorbed from the GI tract in the control group without any absorption enhancers after oral administration of BBR at a dose of 100 mg/kg in rats with C_{max} and $AUC_{0\text{-}36\,h}$ values of 3.93 ± 1.56 ng/ml and 36.01 ± 4.19 ng h/ml, respectively. Addition of

0.5% chitosan to BBR preparation resulted in increased absorption of BBR with a C_{max} value of 13.15 ± 6.61 ng/ml and an $AUC_{0-36\ h}$ value of 66.75 ± 15.84 ng h/ml. Addition of 1.5% chitosan to BBR solution enhanced the absorption of BBR with C_{max} and $AUC_{0-36\ h}$ values of 29.05 ± 23.74 ng/ml and 78.35 ± 28.73 ng h/ml, respectively. When the chitosan solution concentration was increased to 3.0%, the C_{max} and $AUC_{0-36\ h}$ were increased to 13.27 ± 8.95 ng/ml and 91.13 ± 51.29 ng h/ml, respectively. With the addition of 0.5%, 1.5%, and 3.0% chitosan to the BBR solutions, the $AUC_{0-36\ h}$ of BBR was increased by about 1.9, 2.2, and 2.5 times compared with the control group, respectively. This indicated that chitosan produced a dose-dependent enhancement of BBR absorption.

Soluble chitosan hydrochloride was also investigated with regard to enhancing BBR GI absorption in rats. The plasma BBR concentration-time curve following administration of BBR solution (control) and formulations containing 2.0% and 3.3% chitosan hydrochloride are shown in Figure 3. The main pharmacokinetic parameters are listed in Table 2. Unexpectedly, the absorption of BBR was not increased but decreased to a degree following administration of BBR formulations containing 2.0% and 3.3% chitosan hydrochloride in rats. The C_{max} and $AUC_{0-36\ h}$ values in the control group were 3.48 ± 1.76 ng/ml and 42.44 ± 13.90 ng h/ml, respectively. After addition of 2.0% chitosan hydrochloride, the C_{max} and $AUC_{0-36\ h}$ values were 2.55 ± 1.95 ng/ml and 31.75 ± 20.36 ng h/ml, respectively. When the chitosan hydrochloride concentration in the formulation was increased to 3.3%, the C_{max} and $AUC_{0-36\ h}$ were 3.46 ± 3.21 ng/ml and 32.32 ± 15.77 ng h/ml, respectively. The $AUC_{0-36\ h}$ values were 0.75 and 0.76 times that of the control group.

BBR solubility study in different solutions

The solubility of BBR in water, 1.0% acetate acid, 0.1 N HCl, saline, chitosan, and chitosan hydrochloride at concentrations of 0.5%, 1.5%, and 3.0% were studied. The results of the BBR solubility in different solutions are shown in Figure 4 and Figure 5. As indicated in Figure 4, BBR had a similar solubility in chitosan solutions of different concentrations. The solubility of BBR in chitosan hydrochloride was significantly lower than that in chitosan solutions of all concentrations. The results shown in Figure 5 showed that the BBR solubility in water, and 1.0% acetate acid was higher than that in 0.1 N HCl, and saline. These results indicate that the BBR solubility is reduced by chloride ion.

Histopathological evaluation

Photomicrographs of the intestinal mucosa exposed to BBR and BBR combined with chitosan and chitosan hydrochloride are shown in Figure 6. The ileum cavity was chosen as the site of testing because it is very sensitive to chitosan. As indicated in Figure 6, the epithelium of each group was undamaged, and villus structure was intact. There was no significant difference between BBR alone and chitosan and chitosan hydrochloride.

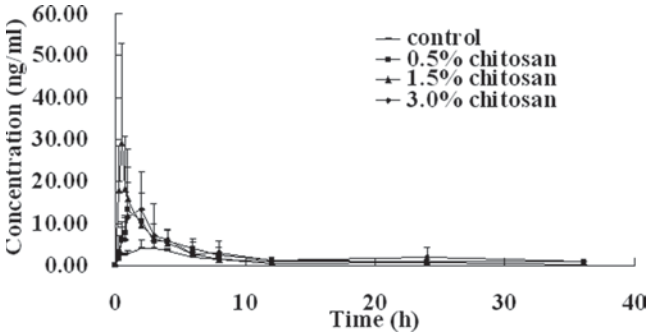

Figure 2. Plasma BBR concentration-time profile after oral administration of BBR alone (control) and in combination with chitosan of different dose in rats. Data are shown as mean concentration, and error bars represent SD (n=5). The BBR dose was 100 mg/kg. The concentration of chitosan in the formulations was 0.5%, 1.5%, and 3.0%, respectively.

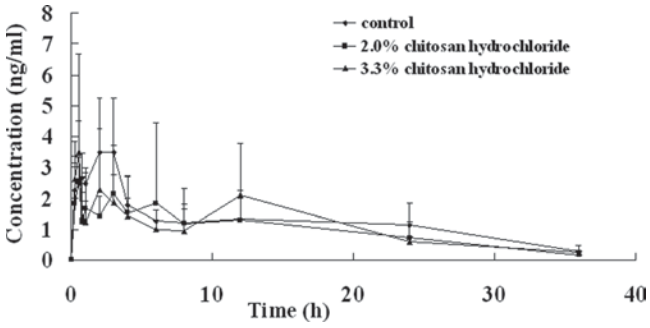

Figure 3. Plasma BBR concentration-time profile after oral administration of BBR alone (control) and in combination with chitosan hydrochloride of different dose in rats. Data are shown as mean concentration, and error bars represent SD (n=5). The BBR dose was 100 mg/kg. The concentration of chitosan in the formulations was 2.0% and 3.3%, respectively.

Table 1. Mean (± SD) pharmacokinetic parameters of BBR after administration of control preparation and test preparations containing chitosan in rats (n = 5).

Formulation	C_{max} (ng/ml)	T_{max} (h)	AUC0-36h (ng h/ml)
Control	3.93 ± 1.56	2.80 ± 1.30	36.01 ± 4.19
0.5% Chitosan	13.15 ± 6.61*	1.40 ± 0.55	66.75 ± 15.84*
1.5% Chitosan	29.05 ± 23.74	0.95 ± 1.15	78.35 ± 28.73*
3.0% Chitosan	13.27 ± 8.95	3.00 ± 3.92	91.13 ± 51.29*

BBR was dissolved in water or chitosan solutions with different dose.
*Significantly different from control group, p< 0.05.

Table 2. Mean (± SD) pharmacokinetic parameters of BBR after administration of control preparation and test preparations containing chitosan hydrochloride to five rats.

Formulation	C_{max} (ng/ml)	T_{max} (h)	AUC0-36h (ng h/ml)
Control	3.48 ± 1.76	1.90 ± 1.14	42.44 ± 13.90
2.0% Chitosan Hydrochloride	2.55 ± 1.95	3.65 ± 3.29	31.75 ± 20.36
3.3% Chitosan Hydrochloride	3.46 ± 3.21	2.45 ± 3.30	32.32 ± 15.77

BBR was dissolved in water or chitosan hydrochloride solutions with different dose.

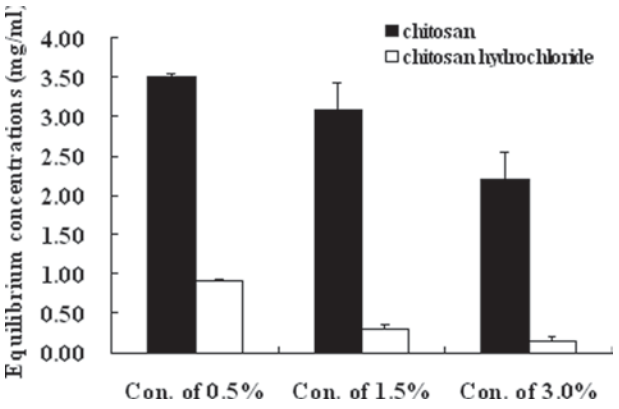

Figure 4. BBR equilibrium concentration in chitosan and chitosan hydrochloride solution with different concentration. Each point is the mean value (± SD) of three experiments. The Con. represents concentration.

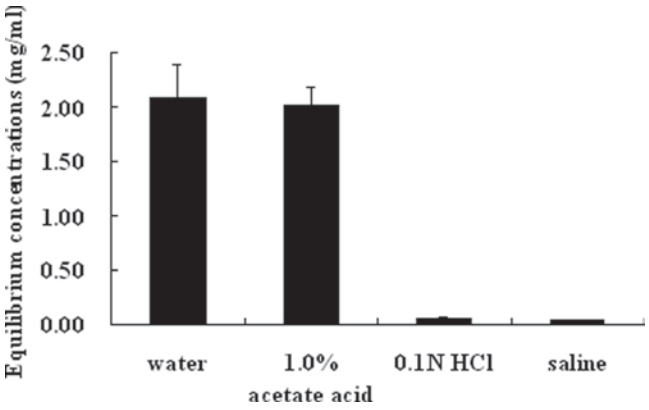

Figure 5. BBR equilibrium concentration in water, 1.0% acetate acid, 0.1 N HCl and saline. Each point is the mean value (± SD) of three experiments.

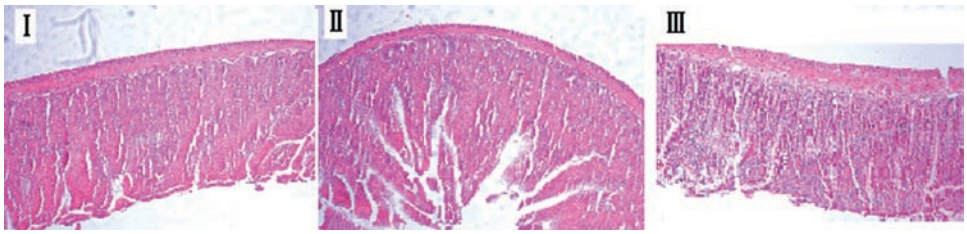

Figure 6. Histopathological comparison between test and control rats. Light micrographs sample were taken at 5 h after oral gavage administration of drugs (original magnificent × 100). 1 control, oral gavage administration of BBR; II oral gavage administration of BBR formulation containing 3.0% chitosan; III oral gavage of BBR formulation containing 3.3% chitosan hydrochloride. The BBR doses were all 100 mg/kg.

Discussion

BBR is an isoquinoline alkaloid with a temperature-dependent aqueous solubility which is increased with an increase in temperature. Battu et al. reported that BBR has a log P value of –1.5 and it is considered as hydrophilic[11]. The main method to improve the absorption of BCS class III drugs such as BBR is to regulate the paracellular permeability and increase the transport of drug into the systemic circulation. The paracellular route is aqueous pathway along the intercellular space between adjacent cells and is restricted by tight junctions[12]. Compared with the transcellular pathway, the paracellular pathway is a very limited route for drug absorption and accounts for only about 0.1% of the intestinal epithelium surface[22]. The rate-limiting barrier of the paracellular pathway is the tight junction and the paracellular permeability of drugs is primarily regulated by the gate. An important method used to increase drug absorption is to regulate the drug paracellular permeability[13,14]. The most efficient method to achieve this effect is to regulate tight junctions by using absorption enhancers. The present work investigated whether the membrane permeability of the lipophobic drug BBR could be enhanced by the application of chitosan as an absorption enhancer.

Based on the studies performed in the past decade, it has become clear that chitosan and chitosan salts are able to enhance the paracellular permeability of a number of compounds. Many *in vitro* studies have been conducted to investigate the absorption-enhancing ability of chitosan. Artursson et al. first reported the effect of chitosan salts on intestinal permeability across tight junctions[15]. These authors found that chitosan glutamate and chitosan hydrochloride were able to increase the permeation of low molecular weight compounds (mannitol) and large hydrophilic compounds (PEG-4000 and fluorescein dextrans)[23]. Schipper et al. studied the relationship between chitosan structural characteristics and their absorption enhancing capacity using chitosan

hydrochloride and found that the absorption enhancing abilities is related to the molecular weight and degree of deacetylation. Chitosans with a high degree of deacetylation were efficient permeation enhancers at both low and high molecular weights, and chitosans with a low degree of deacetylation were only efficient at a high molecular weight[24]. Other authors found that the transport of buserelin was increased markedly by chitosan hydrochloride[25]. Several *in vivo* investigations have also been conducted to investigate the absorption-enhancing ability of chitosan. It is reported that the drug bioavailability of buserelin was enhanced greatly by the intraduodenal application of buserelin and chitosan hydrochloride in a gel formulation in rats[25] . Thanou et al found that intraduodenal application of octreotide acetate and chitosan hydrochloride 1.5% w/v at pH 5 produced a 3-fold increase in drug bioavailability compared with a control[26].

The results of the *in vivo* study indicated that different doses of chitosan all have a BBR absorption-enhancing efficacy in rats. The enhancing ability of chitosan was dose dependent. The higher the chitosan concentration used, the more efficient was the BBR absorption from the GI tract. When the concentration of chitosan reached 3.0%, chitosan had the most efficient enhancing ability with an $AUC_{0-36\ h}$ of 91.13 ± 51.29 ng h/ml, which was 2.5 times ($p < 0.05$) that of the control group. The dose-dependent profile of the chitosan absorption-enhancing ability was mentioned by Schipper et al[27]. It is reported that the positive charge of chitosan and its binding to the naked epithelial apical membrane are essential for increasing absorption[24,28]. Mucus layers covering the intestinal epithelial cell membranes may act as a diffusion barrier and reduce the absorption-enhancing effects of chitosan[27]. Therefore, a high concentration of chitosan was proposed to enhance drug absorption. It was speculated that an increased effective concentration of chitosan could deplete the mucosa of mucins released from goblet cells by a charge interaction and reduce the effective thickness of the mucus layers on the surface of the epithelial cell membrane. Also, the remaining available chitosan could bind directly to the exposed membrane and increase the absorption. Another advantage of chitosan as a high molecular weight polymer is its mucoadhesive properties, which prolong the contact of the polymer with the epithelium surface and increase the time of action as an absorption enhancer. By using sufficiently high chitosan concentrations, transmucosal absorption of drugs can be improved taking advantage both of the ability of chitosan to increase the epithelial permeability and of the mucoadhesive nature of chitosan resulting in increased retention of the drug at the mucosal surface[27].

Since chitosan should be dissolved in acidic solutions and chitosan hydrochloride is soluble in water after agitation, chitosan hydrochloride was also investigated as an absorption enhancer in this study. Compared with the chitosan formulations dissolved in 1.0% acetate acid, chitosan hydrochloride formulations dissolved in distilled water did not show any absorption-enhancing ability but produced a decreased C_{max} and $AUC_{0-36\ h}$ ($p > 0.05$). The precise reason for obtaining such results with the addition of chitosan hydrochloride is not known. It may be due to a solubility change of BBR in formulation solutions. The equilibrium solubility of BBR in different vehicles was examined. As indicated in Figure 4 and Figure 5, the solubility of BBR was sharply reduced when chloride ions were present in the solution, which imply a common ion effect of chloride on the solubility of BBR. The common ion effect is used to describe the effect on a solution of two dissolved solutes that contain the same ion or ions. BBR dissolved in solutions in an equilibrium condition with a solubility product constant. According to Le Chatelier's principle, the addition of chloride ion will suppress the ionization of BBR and reduce its solubility. It is speculated that the reduced solubility of BBR in chitosan hydrochloride produced a limited BBR solution concentration in the GI tract and, therefore, resulted a reduced absorption. However, further studies are needed to confirm this hypothesis.

As shown in Table 1, the test group with chitosan has bigger variability than that of the control group in the study. This may be due to the individual variation of response to chitosan among rats. When the BBR and chitosan mixtures were administered to the rat GI tract, the epithelial cell membrane was covered with mucus layer with different physiological properties and in different lumen conditions which possibly influence the mixture property and may interact with chitosan differently. It is necessary to develop an appropriate chitosan-drug formulation or drug delivery system to reduce variability and obtain a more stable drug absorption enhancement. Most papers published earlier have reported chitosan as an absorption enhancer of macromolecular drugs[29]. In this study, chitosan was used to increase the absorption of the smaller molecule BBR. The results showed that chitosan produces an improvement in the GI absorption of BBR. Chitosan, as a high molecular weight polymer with few systemic side effects, has many advantages, such as biodegradability, biocompatibility, and FDA approval, and has attracted great interest as an excipient in the pharmaceutical field[16–18]. The present study suggests that chitosan can be used as an absorption enhancer of small molecular weight drugs such as BBR and produce efficient enhancement.

Conclusion

Various concentrations of chitosan and chitosan hydrochloride were studied to enhance the GI absorption of BBR. Chitosan showed a dose-dependent absorption-enhancing ability. Addition of 3.0% chitosan produced the highest BBR $AUC_{0-36\ h}$. Chitosan hydrochloride did not show an absorption increasing effect and this may be due to the common ion effect of chloride. Histopathological evaluation performed after applying chitosan and chitosan hydrochloride did not show any significant

morphological changes. This study showed that chitosan is a good pharmaceutical excipient for use as an absorption enhancer to improve the absorption of BBR.

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Declaration of interest

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