Bioadhesive Properties of β-Limit Dextrin

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ABSTRACT - Purpose. β-Limit dextrin has been studied for many years as a means to investigate the internal structures of amylose and amylopectin. However its role as an excipient in the pharmaceutical industry has never been reported. This paper is the first one in a series to explore its potential use as an excipient to aid drug delivery. **Methods.** The bioadhesive properties of β-limit dextrin were studied using a texture analyser and compared with two well-known bioadhesive polymers – carbopol and chitosan (as controls). **Results.** The β-limit dextrin has significant mucoadhesive properties; similar to carbopol but superior to chitosan. **Conclusions.** The nature of β-limit dextrin (a starch derivative) makes it safe to consume and provides a natural alternative when compared with synthetic polymers. In addition, the polysaccharide can be digested by salivary amylase and thus provide a clean mouth feel.

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INTRODUCTION

Bioadhesion is the process whereby synthetic and natural macromolecules adhere to a biological tissue for an extended period of time in the body (1, 2). When a substrate is a mucosal epithelium, a bioadhesive system adheres and interacts primarily with the mucus layer; this phenomenon being referred to as mucoadhesion (1). Materials with mucoadhesive properties have received considerable attention as excipients for various drug delivery systems (Table 1). When mucoadhesive materials are incorporated into pharmaceutical formulations, the residence time of dosage forms on the mucosa can be prolonged significantly, allowing a sustained drug release at a given target site. Furthermore, mucoadhesive materials can guarantee an intimate contact with the adsorption membrane, providing the basis for a high concentration gradient as a driving force for passive drug uptake.

Various classes of materials have been investigated in order to meet the requirements for a mucoadhesive polymer, such as proper hydrogen-bonding functional groups, suitable wetting properties, swelling/water load properties, and sufficient flexibility for entanglement with the tissue mucus network (3). Park and Robinson (4) reported that in order for mucoadhesion to occur, the desired polymers must have functional groups that are able to form hydrogen bonds. The hydrophilic functional groups usually associated with forming hydrogen bonds (especially in polysaccharides) are the hydroxyl (-OH) and carboxylic groups (-COOH). A major reason behind the selection of hydrophilic polymers for oral transmucosal drug delivery system is the water-rich environment of the oral cavity owing to the presence of saliva (5).

β-Limit dextrins are specific starch hydrolysis products obtained by treating solubilised starches with (pure) β-amylase. Starch consists of two polysaccharides, amylose and amylopectin. Both polysaccharides are based on chains of 1-4 linked α-D-glucose where amylose is essentially linear, but amylopectin is highly branched with 5% of the glycosidic bonds being α-(1-6) conformations (6). β-Amylase catalyses the hydrolysis of α-(1-4) linkages of amylose/amylopectin by the successive liberation of maltose from the non-reducing ends, but it cannot by-pass the α-(1-6) branching points. As a consequence, linear amylose is completely hydrolysed to maltose, whereas about 50-60% of amylopectin is converted into maltose, with the remaining material being β-limit dextrin. β-Limit dextrin has a high molecular weight that accounts for 40-50% the weight of its original amylopectin (10^6) (7). It is a highly branched molecule consisting of the inner core amylopectin structure with its outer chains recessed to one to three glucose units (Figure 1).

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β-Limit dextrin's large size and highly branched structure are responsible for the high viscosity of its dispersions which, together with its hydrophilic nature, makes it a suitable candidate as a bioadhesive polymer (8).

The aim of this study was to evaluate the bioadhesive properties of β-limit dextrin using a texture analyser and evaluate its possible use as a pharmaceutical excipient. An initial *in vitro* test is, therefore, described for this purpose.

Table 1. Mucoadhesive polymers for buccal delivery

Categories	Examples
Semi-natural/natural	Agarose, chitosan, gelatine, hyaluronic acid, various gums (guar, hakea, xanthan, gellan, carragenan, pectin and sodium alginate)
Synthetic	*Cellulose derivatives* [CMC, thiolated CMC, Sodium CMC, HEC, HPC, HPMC, MC, methylhydroxyethylcellulose] *Poly (acrylic acid)-based polymers* [CP, PC, PAA, polyacrylates, poly (methylvinylether-co-methacrylic acid), poly (2-hydroxyethyl methacrylate), poly (acrylic acid-co-ethylhexylacrylate), poly (methacrylate), poly (alkylcyanoacrylate), poly (isohexylcyanoacrylate), poly (isobutylcyanoacrylate), copolymer of acrylic acid and PEG] *Others* Poly (*N*-2-hydroxypropyl methacrylamide) (PHPMAm), polyoxyethylene, PVA, PVP, thiolated polymers

Abbreviations: CMC, sodium carboxymethyl cellulose; HEC, hydroxyethyl cellulose; HPC, hydroxypropyl cellulose; HPMC, hydroxypropyl methylcellulose; MC, methylcellulose; CP, carbopol; PC, polycarbophil; PAA, poly (acrylic acid); PEG, poly (ethylene glycol); PVA, poly (vinyl alcohol); PVP, poly (vinyl pyrrolidone).
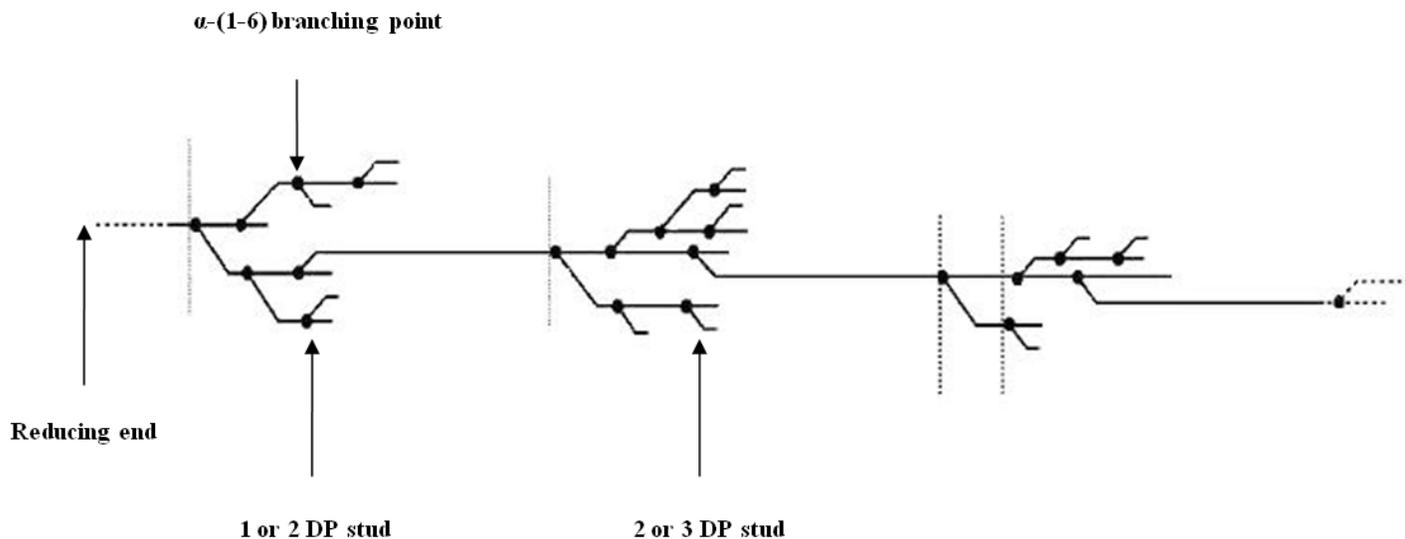

Figure 1: Schematic representation of a molecular segment of β-limit dextrin.

MATERIAL AND METHODS

Materials

Waxy maize starch (Amioca TF) was purchased from National Starch (Manchester, UK). β-Amylase (A-7005 from sweet potato, 750 units/mg) and porcine mucin (type II crude) was obtained from Sigma (Gillingham, UK). The adhesive polymer control Carbopol 934® (C934) was purchased from B. F. Goodrich (Cleveland, OH, USA). Hydroxypropylmethylcellulose (HPMC) was purchased from Colorcon (Kent, UK). Chitosan glutamate was obtained from NovaMatrix (Sandvika, Norway). Spray dried lactose (Zeparox) was included as a diluent and was purchased from Thornton and Ross (Huddersfield, UK). Polyvinylpyrrolidone 44000 (PVP, BPE431-500) was used as a binding agent and magnesium stearate (M/0955/53) as a lubricating agent. Both were obtained from Fisher (Loughborough, UK).

Preparation of β-limit dextrin

β-Limit dextrins of waxy maize starch were produced according to the general method of Hizukuri and Maehara (9) with some modifications (7). Waxy maize starches (Amioca TF) were dissolved completely in acetate buffer (0.2M, pH 5.6) to which β-amylase was added at 10,000 u g^{-1} starch. The dispersion was transferred into cellulose Visking tubing (Fisher Scientific, TWT-400-110D) and was dialysed against the same buffer at 37ºC for 36 h and then against distilled water to remove the buffer salts. The buffer was changed three times during the first 12 h and twice thereafter. The reaction was terminated by boiling the mixture and the coagulated protein was removed by centrifugation. To the supernatant, cold ethanol was added to precipitate the polysaccharides which were collected by centrifugation (5 min, 1,500 xg). The collected materials were washed by successive suspension in ethanol (recovered again by centrifugation for 5 min at 1,500 xg) and finally dissolved in water then freeze dried.

Tablet preparation

The compositions of tablets for adhesive testing are shown in Tables 2 and 3. All powdered excipients were dry blended for 5 min using a mortar and pestle to form a homogenous and directly compressible powder mix. The tablets were prepared by direct compression using a single-punch tablets press (Manesty F3, Liverpool, UK) and 6 mm diameter flat punches.

In vitro determination of mucoadhesive performance

Mucoadhesion testing was conducted *in vitro* using double strength nutrient agar (lab-Lemco powder 2g/L, yeast extract 4g/L, peptone 10g/L, sodium chloride 10g/L, agar 30g/L) coated with a 5% solution of porcine mucin over the surface. Measurements were made with a Texture Analyser (TA-XT2i, Stable Micro Systems, Surrey, UK) with a force of 0.25N and contact time of 10 minutes. Each tablet was attached to the base of an aluminium probe (using double-sided adhesive tape) fixed to the mobile arm of the texture analyser. The tablet was lowered at a rate of 0.1mm s^{-1} until contact with the agar was made. A contact force of 0.25N was maintained for 10 min, after which the probe was withdrawn from the agar at a rate of 5 mm s^{-1}. The peak detachment force (N) and the work of adhesion (area under the force/distance curve in mJ) was recorded. Triplicate determinations were made with typically a coefficient of variation (cv) of <5 %.

RESULTS

Texture analysis is a useful tool and has been extensively used as a means for mechanical characterisation of pharmaceutical mucoadhesive dosage forms. The detachment force gives an indication of the mucoadhesive strength of the testing polymer (10). In this study, the mucoadhesive effect of β-limit dextrin in a solid tablet at different concentrations was assessed using the texture analyser approach. The compositions of tablets (formulations A2, B2, C2 and D2) are shown in Table 2 and the texture analysis results are represented in Figure 2. As shown in Figure 2, it is apparent that the peak detachment force of formulations containing increasing amounts of β-limit dextrin (20 to 50%) reaches a maximum at approximately 30% (w/w). Higher concentrations of β-limit dextrin do not alter significantly the peak detachment force. This is a measure of adhesion since the peak detachment force is considered to be dependent on the formation of hydrogen bonds between the functional groups of the bioadhesive (here β-limit dextrin) and the mucus membrane (11).

Table 2. Composition of tablets containing different amount of β-limit dextrin (expressed as mg per tablet)

Ingredient	Formulation			
	A2	B2	C2	D2
β-Limit dextrin	20	30	40	50
Polyvinylpyrrolidone 44 000	6	6	6	6
Magnesium Stearate	1	1	1	1
Zeparox (lactose)	73	63	53	43

Table 3. Composition of tablets containing β-limit dextrin and other materials (expressed as mg per tablet)

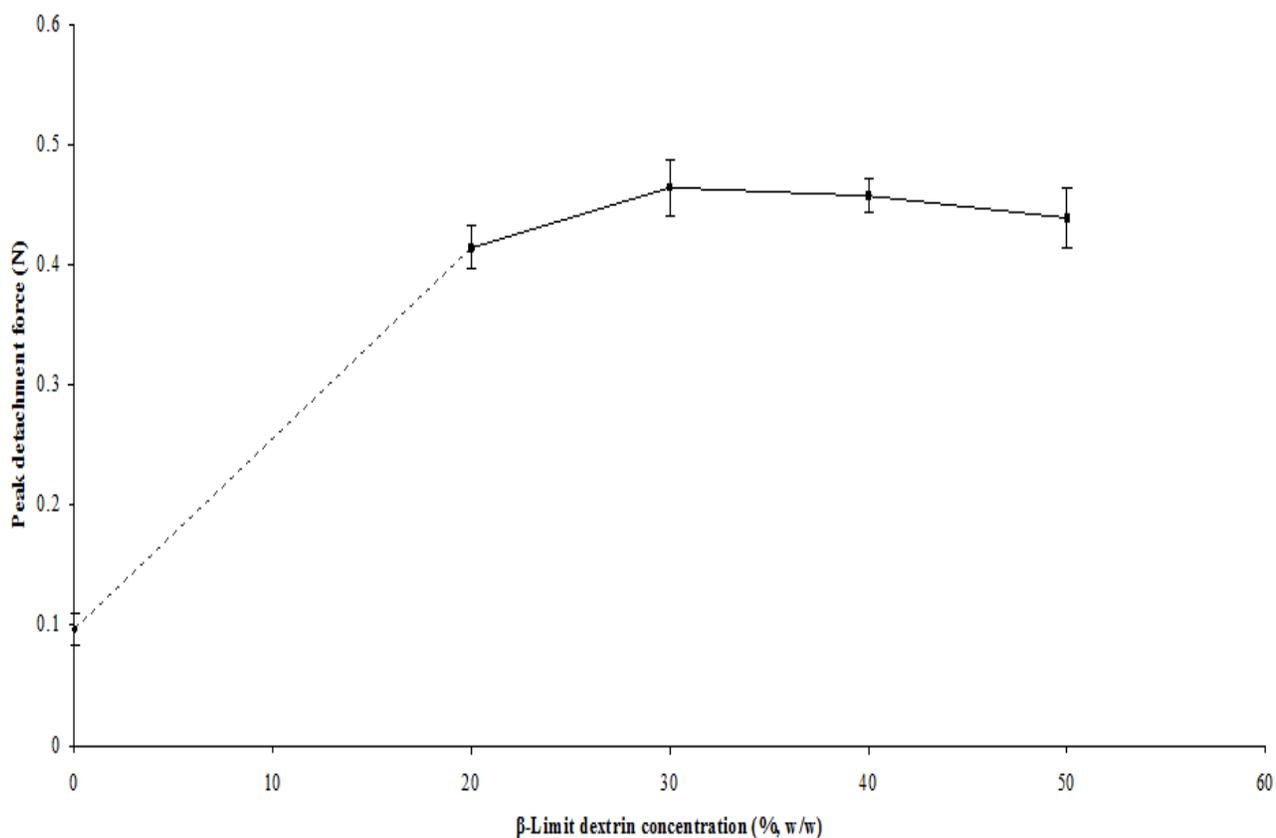| Ingredient	Formulation			
	A3	B3	C3	D3
β-Limit dextrin	30	--	--	--
Carbopol 934®	--	30	--	--
Chitosan	--	--	30	--
Hydroxypropylmethylcellulose	--	--	--	30
Polyvinylpyrrolidone 44 000	6	6	6	6
Magnesium Stearate	1	1	1	1
Zeparox (lactose)	63	63	63	63

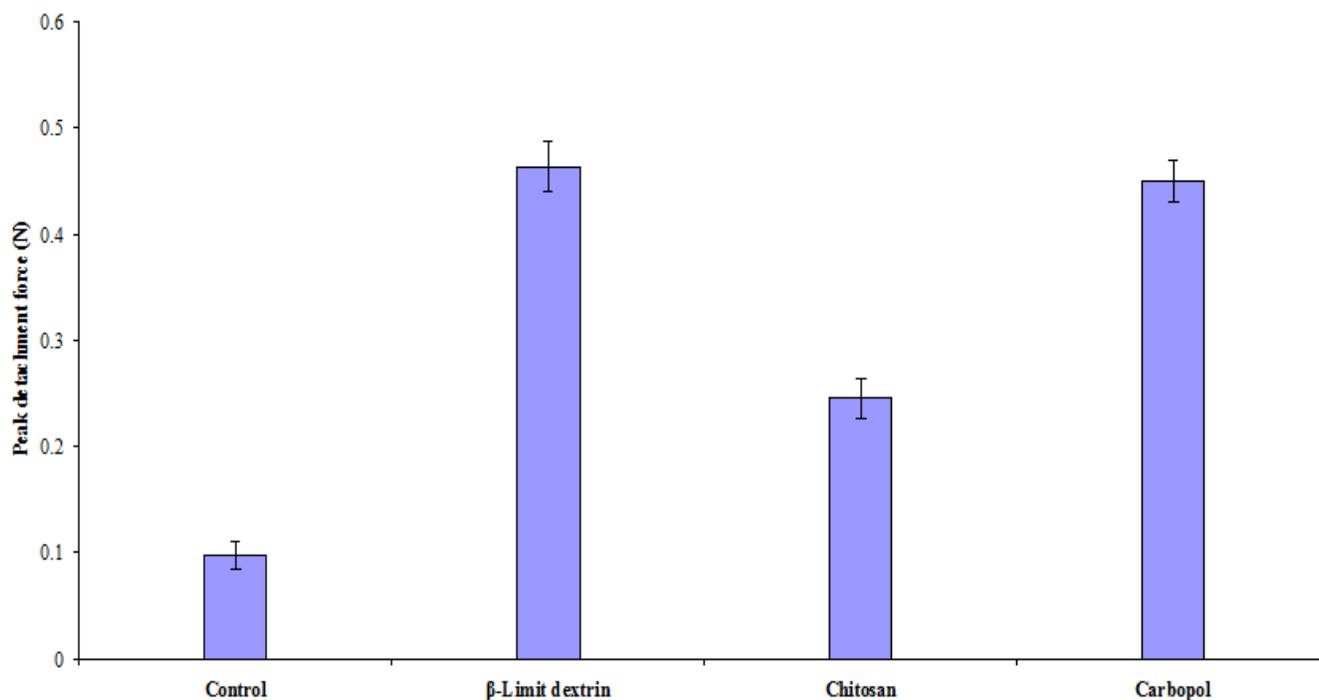**Figure 2.** Average peak detachment force (N) versus β-limit dextrin concentration (% w/w) (±S.D., n=3)

Figure 3. Comparison of peak detachment force (N) between β-limit dextrin and other materials (±S.D., n=3)

At 30% w/w β-limit dextrin, the potential for hydrogen bonding reaches a maximum due to optimization of interactions of the functional groups (hydrogen bonding) at the tablet/mucin (representing the mucus membrane) interface. Therefore, the peak detachment force cannot be increased by the addition of more adhesive polymer beyond this concentration.

Formulation B2 was chosen for further study to compare the mucoadhesive property of β-limit dextrin to two well-known mucoadhesive polymers - carbopol 934 and chitosan (Table 3). A placebo tablet was also prepared that contained no known mucoadhesive materials (Table 3). The results for the peak detachment force of the studied materials are shown in Figure 3. It is apparent that the β-limit dextrin exhibits a similar mucoadhesive force to carbopol (cross linked acrylic acid) while chitosan demonstrates lower mucoadhesive forces under the same conditions.

DISCUSSION

It has been proposed that the interaction between the mucosal surfaces and mucoadhesive polymers is a result of physical entanglement and secondary bonding, mainly hydrogen bonding and Van der Waals attractions (12, 13) as discussed above.

These forces are related to the chemical structure of the polymers. The types of surface chemical groups that contribute to this type of adhesion include hydroxyls, carboxyls, amines and amides (14, 15). Physical properties such as the rate of hydration and rheological properties of the polymeric formulations are likely to have a major impact on their bioadhesion and consequently their eventual duration of retention (16, 17).

Although starches (when gelatinised especially) have the capacity to hydrogen bond with other materials and surfaces, their gelling/interactive properties are modified by re-crystallisation (retrogradation) with time (18). This formation of double helices and their association into arrays, limits the availability of interactions within solutions and with surfaces. Hence, the potential applications of amylose and amylopectin for bioadhesive applications are severely restricted. Furthermore, due to this tendency to retrograde rapidly in aqueous environments (especially amylose), the bioavailability of any pharmaceutically active ingredients in starch based systems may be compromised.

β-Limit dextrin, is similar to amylopectin and amylose, in that it is built from glucose residues rich with hydroxyl groups capable of hydrogen bonding with other residues. The β-limit dextrin

structure, however, is not prone to exterior chain helical formation due to the hydrolysis of these regions by β-amylase (7). This prevents retrogradation of the polymer. The hydroxyl groups of β-limit dextrin thus provide a rich source of potential hydrogen bonding sites with other surfaces and structures. This capability makes it highly desirable for oral delivery.

The β-limit dextrin polymer is currently being developed as an oral delivery excipient in the format of lyophilised 'wafers'. These wafers have proven to be robust in terms of structure and provide efficient delivery matrices for different active pharmaceutical ingredients (APIs) in the mouth. The wafer formats are part of an ongoing programme of development to commercialise the technology for oral drug delivery applications. Key competitive products in the field (oral delivery wafers) are gelatine based. Traditionally the gelatine has been sourced from animals for this purpose although fish gelatine is also available. Gelatine is an effective excipient for lyophilised oral delivery wafers although as a protein, there are functional, processing and potential pharmacological challenges which impact on usage. Furthermore, gelatine is generated from animal sources (dextrins from starches and hence plants) with associated ethical issues but more importantly related to potential contamination with animal derived infective agents (e.g. BSE). In terms of mouth feel, the dextrins provide a very clean (non-gummy) oral sensation .

In addition to the desirable physical properties of the β-limit dextrin (described above), β-limit dextrin can be hydrolysed readily by α-amylase, such as salivary α-amylase. This provides an additional benefit for oral drug delivery in that the α-glucan polymeric structure is removed from the mouth and no residues are detected. For other polysaccharides such as pectin etc. this cannot happen due to the absence of relevant enzymes in the saliva.

CONCLUSION

β-Limit dextrin has significant mucoadhesive activity and may be used as an excipient for a mucoadhesive drug delivery system. It compares favourably to synthetic polymers like carbopol, chitosan etc. for this purpose. Because it is a 'natural' derivative of starch, this makes it safe to be consumed, with the advantage that removal of the polymer from the mouth post drug delivery by the saliva, leaves a desirable oral sensation.

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