ELSEVIER

International Journal of Pharmaceutics 160 (1998) 61–74

international journal of pharmaceutics

Evaluation of possible mechanism(s) of bioadhesion

Khalid Mahrag Tur, Hung-Seng Ch'ng *

School of Pharmaceutical Sciences, Universiti Sains Malaysia, Minden, 11800, Penang, Malaysia

Received 24 June 1997; received in revised form 1 September 1997; accepted 10 September 1997

Abstract

The possible bioadhesion mechanism(s) of a series of 0.3% cross-linked polyacrylic and polymet acrylic acids to the mucin and animal tissue was investigated in vitro. The zeta potential and extent of hydration of the polymers were examined in the presence of three physiologically relevant test fluids (isotonic saline, simulated gastric and simulated intestinal fluids). Our results indicated that for a same series of polymers such as either cross-linked polyacrylic acid or polymethacrylic acid, the increase in the negative zeta potential resulted in an increase in bioadhesion (force of detachment) between the polymer and rabbit stomach tissue for both isotonic saline and simulated gastric fluid. Isotonic saline produced higher zeta potential and bioadhesion for a given series of polymers in comparison with simulated gastric fluid. The acidic condition of gastric fluid suppressed the ionization of the polymers hence their possession of lower zeta potential. All polymers swelled extensively in simulated intestinal fluid, leading to difficulties in determining their zeta potential hence no comparison was possible. The adhesive performance, measured by colloidal gold staining technique, between polymer and bovine submaxillary mucin in gastric medium revealed the same ranking order as in the detachment experiment except for PMDB and PMDG polymers. No comparable data were available for saline and intestinal media due to the possible surface charge repulsion dominating at the polymer–mucin interface. The relationship found between the zeta potential and the bioadhesive strength of polymers showed that the bioadhesion was linked to the extent of ionization that occurred during the interaction of the polymer with the mucin/epithelial cell surface. A suitable extent of ionization was required for the initial adhesion of polymer to the biological tissue. However, extensive ionization and swelling could lead to the formation of very loose particles or dissolution and therefore difficulty in interacting with the animal tissue.

Keywords: Swelling; Zeta potential; Bioadhesion mechanism(s); Bioadhesive polymers; Adhesion test

1. Introduction

Bioadhesive polymers have received considerable attention as absorption promoters due to their ability to adhere to the mucin/epithelial cell

* Corresponding author.

PII S0378-5173(97)00297-4

surface and thereby to anchor a dosage form at the site for optimum drug absorption (Hui and Robinson, 1985; Longer et al., 1985). Different molecular theories have been postulated to explain the molecular events which occur during bioadhesion (Peppas and Buri, 1985). Nevertheless, the interaction between polymers and the mucin/epithelial cell surface is still rather complicated. It was suggested by several workers that the initial interaction between the polymer and the biological surface is through electrostatic interaction followed by mechanical interlocking of the polymer chains, Van der Waal's attraction and other factors. Lehr et al. (1993) have modelled this phenomenon by considering the polar and dispersive components of surface energy of the polymer and of mucus. No correction was made for eventual interpenetration in a later stage of the mucoadhesive contact, nor for other relevant surface energy parameters such as possible ionic interactions, hydrogen bonding and acid–base interactions. The latter parameter has been considered by Rillosi and Buckton (1995) and proved to be a valuable tool in studies of mucoadhesive mechanism. However, no information on the surface charge density of polymers was given to correct for electrostatic behaviour during bioadhesion process.

Although the role of charge density of polymers has been explored in gastric condition by Park and Robinson (1987), the mechanism of bioadhesion in the presence of different fluids has not been systematically studied. In the present work we aim to correlate the zeta potentials of water-insoluble polymers and their bioadhesive behaviours in the presence of three simulated physiological fluids. To achieve this aim the physicochemical properties of polymers such as hydration and zeta potential will be determined and correlated with adhesiveness of the polymer with mucin (colloidal gold staining) and the animal tissue (detachment force). The study would also allow us to establish the rank order of bioadhesive performance of polymers in the three physiological fluids.

2. Experimental procedures

2.1. Materials

Acrylic acid, methacrylic acid, benzoyl peroxide and sodium sulfite were purchased from E. Merck, Darmstadt, Germany. Bovine submaxillary mucin (Type I), gold tetrachloride and bovine albumin were from Sigma (St. Louis, MO). Magnesium sulfate heptahydrate from BDH (Poole, UK), divinylbenzene and 2,5-dimethyl-1,5-hexadiene from TCI (Kasei, Tokyo, Japan) and divinylglycol from Polysciences (Warrington, PA). Isotonic saline, simulated gastric and simulated intestinal solutions were prepared from distilled water according to the relevant USP monographs, but without adding any enzymes. The pH was 6.0 for isotonic saline, 1.3 for gastric fluid and 7.5 for intestinal fluid.

2.2. Synthesis of polymers

Polymers of acrylic and methacrylic acids cross-linked (0.3% w/w) with 2,5-dimethyl-1,5-hexadiene (PADH; PMDH), divinyl glycol (PADG; PMDG), and divinyl benzene (PADB; PMDB) were synthesized by the method of Ch'ng et al. (1985). The synthesized polymers were extensively washed with distilled water to remove unreacted monomers, cross-linking agents and initiators. The washed cross-linked polymers were completely dried in a hot air oven at 90°C for 24 h before being ground to the required size of 400–630 μm in diameter.

2.3. Swelling of polymers

100 mg of each polymer was placed in a 10-ml graduated measuring cylinder. 10 ml of either isotonic saline, simulated gastric, or simulated intestinal solution was added to the cylinder and the polymer was allowed to swell at room temperature (26°C). The volume of the polymer was measured from the meniscus of the interface between the hydrated polymer and the test solution. The volume measurement was taken at different time intervals until equilibrium swelling was achieved. Three replications of each test were carried out.

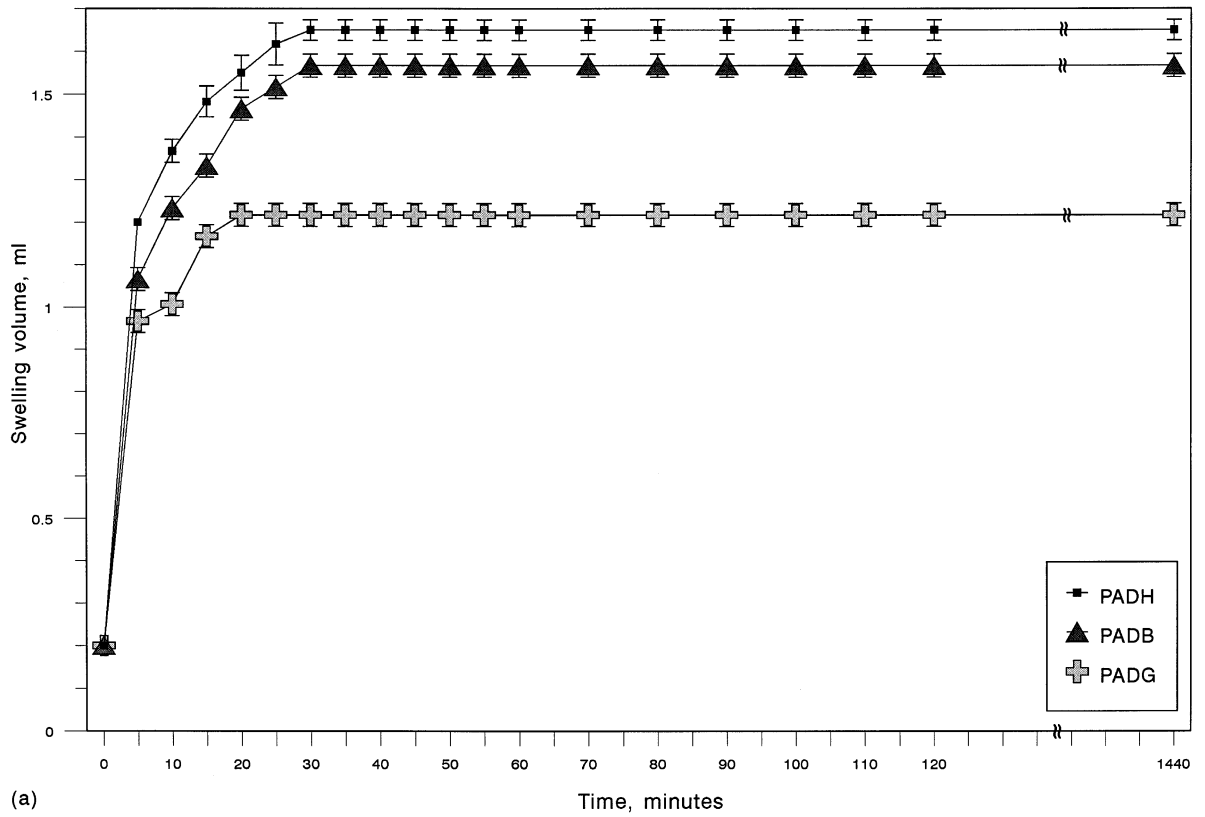

Fig. 1. Swelling profiles of acrylic (a) and metahacrylic polymers (b) in isotonic saline solution. Each point represents the mean ± S.E. of triplicate experiments. Note: The S.E. is less than the point size for some points.

2.4. Determination of zeta potential

The electrophoretic mobility (EPM) of the polymers dispersed in either isotonic saline, simulated gastric or simulated intestinal solutions was measured at 25°C using a Rank mark II microelectrophoresis apparatus (Rank, Bottisham, Cambridge, UK). 100 mg of the polymer (less than 50 μm in diameter) was dispersed in 20 ml of the medium, and a known amount of this suspension was added into the flat cell filled with the tested medium. The time required for the particle to travel across a fixed distance (50 μm) either from right to left or left to right, depending on the charge of electrodes, was recorded at an applied voltage of 10 V. For statistical purposes, at least ten readings were recorded for each polymer for each direction of movement. The zeta potential was calculated using Helmholtz–Smoluchowski equation.

2.5. In vitro evaluation of bioadhesion

Two different methods were used for the purpose of screening the bioadhesive strength of polymers. In the first, the interaction was determined by measuring the force required to detach a polymer from the animal tissue. In the second method, the bioadhesive strength was measured from the interaction with mucin using colloidal gold staining technique.

2.5.1. Tensile testing

The force required to separate a polymer specimen from freshly excised rabbit stomach was measured in isotonic saline, simulated gastric and

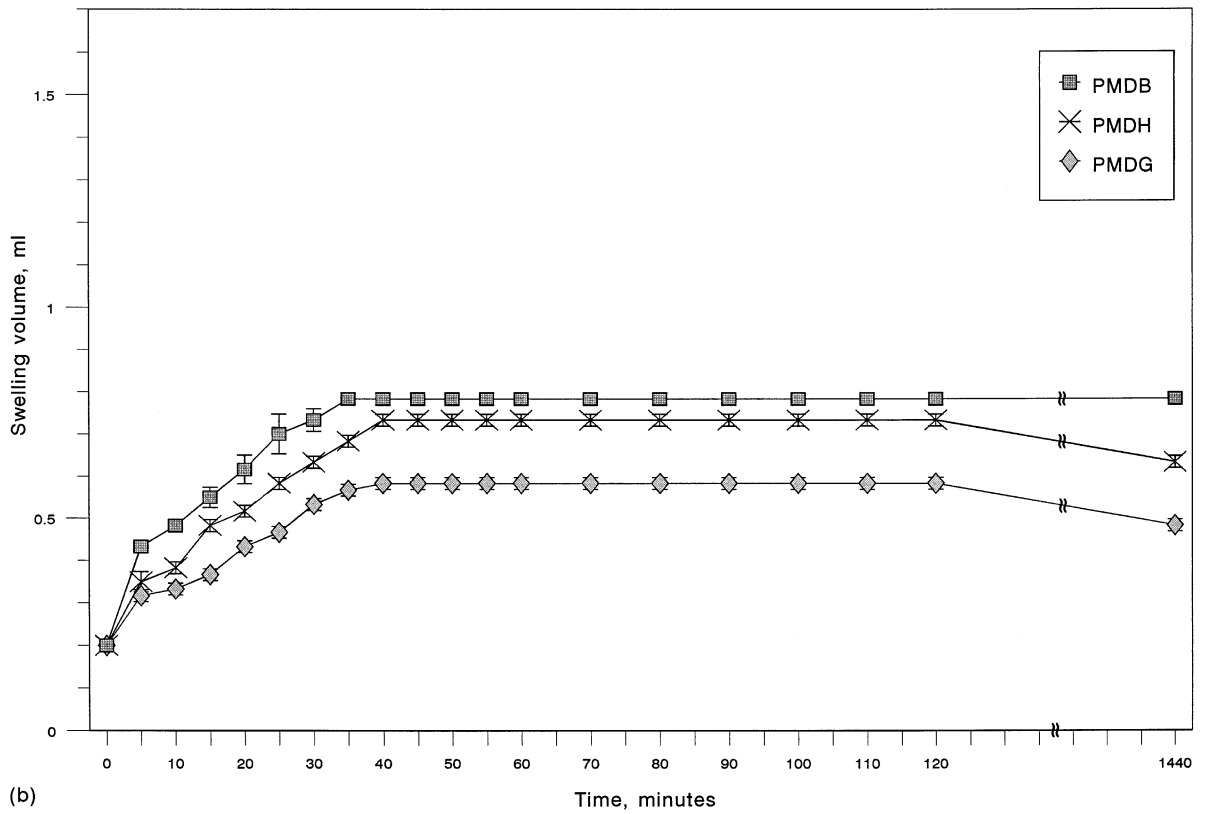

Fig. 1. (*Continued*)

simulated intestinal solutions using a modified method of Ch'ng et al. (1985) on a TA.XT2 Texture Analyzer (Stable Micro Systems, Haslemere, Surrey, UK) connected to a personal computer and running with a XTRA Dimension software package. A section of tissue was cut from the fundus of a healthy rabbit stomach and secured, mucosal side out, to a tissue mounting device. The tissue was equilibrated for 15 min before and maintained during the test in 200 ml of the medium at 37°C. Another section of the stomach was placed, mucosal side out, over the instrument probe and secured with an aluminium cap with a hole of 10 mm diameter in its center. Four milligrams of the fully hydrated polymer was carefully spread into a uniform monolayer over the exposed tissue on the probe. The hydrated polymer was brought into contact with the tissue with a 2 g force and maintained for 1 min. At the end of this time, the probe was withdrawn at a rate of 0.1 mm/s for 15 cm and the force/time curve was recorded until the polymer became detached from the mucus layer. The maximum force required for detachment was determined directly from the recorded curve. The influence of applied force (2, 5, 10 g) on the bioadhesion was investigated in a separate part of the study.

2.5.2. Colloidal gold staining

The dried mucin powder was dissolved in deionized distilled water in a concentration of 1% (w/v). The mucin solution was dialyzed against deionized distilled water at 4°C for 24 h using spectra/Por®7 dialysis membrane (Spectrum, Houston, USA). The mucin-gold conjugates were freshly prepared by the method of Park (1989), then diluted with either isotonic saline, simulated gastric, or simulated intestinal solutions to give an

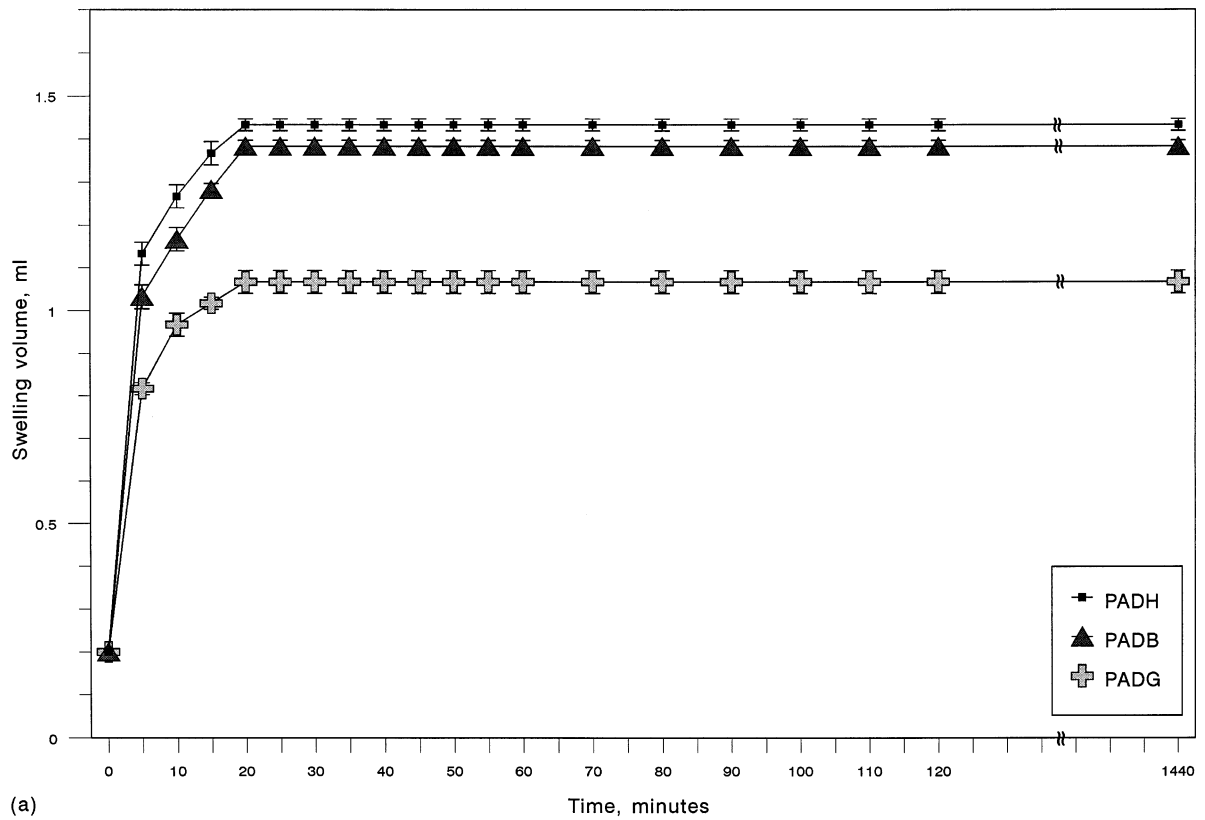

Fig. 2. Swelling profiles of acrylic (a) and metahacrylic polymers (b) in simulated gastric fluid. Each point represents the mean ± S.E. of triplicate experiments. Note: The S.E. is less than the point size for some points.

UV absorbance of 1.331 at 525 nm. 10 ml of this diluted mucin-gold conjugates solution was added slowly to 100 mg of fully hydrated polymer particles. The intensity of the red colour on the polymer surface was quantified by measuring the decrease in the absorbance value of the bulk solution from the initial value at 525 nm by using spectrophotometer model U-2000 (Hitachi, Tokyo, Japan).

3. Results and discussion

3.1. Swelling studies

Some physico-chemical properties of the above polymers were previously determined by several workers (Ch'ng et al., 1985; Park and Robinson, 1985). However, the swelling profiles of various polymers in the three physiologically relevant fluids were not determined and therefore it would be useful to obtain these data in order to help to interpret the mechanism of bioadhesion in these fluids. The swelling profiles are shown in Figs. 1–3. All polymers hydrate quickly and reach an equilibrium after 20–30 min in an acidic condition, 30–40 min in saline solution and 40–60 min in intestinal medium. At the initial stage, the swelling occurs very rapidly due to the entry of water via metastable pores in the polymer. This mechanism is known as hysteresis of the swelling that followed by swelling as a result of diffusion processes (Garcia-Gonzalez et al., 1993).

As expected, the swelling increases as a function of pH, being minimal in acidic, intermediate in saline and maximal in intestinal solutions. This

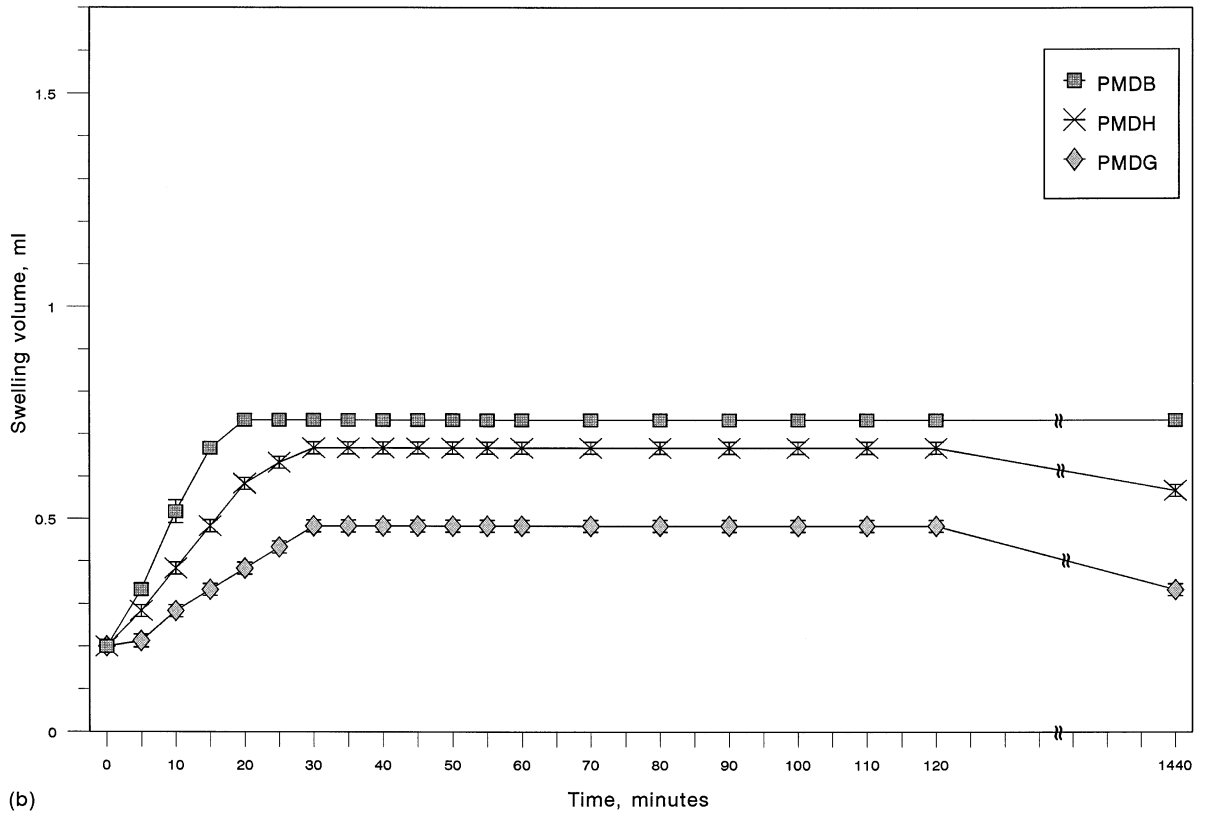

Fig. 2. (*Continued*)

behaviour is related to the increase in ionization as a result of increased pH that leads to increase in electrostatic repulsion between adjacent carboxylic groups and the subsequent formation of an expanded polymer network. Two-way ANOVA applied to the swelling experiments indicates that the effects on swelling of polymer (P), medium (M) and the interaction between them ($P \times M$) are all significant ($P{:}F_{5,36} = 2461.16$, $\alpha < 0.0001$; $M{:}F_{2,36} = 6584.48$, $\alpha < 0.0001$; $P \times M{:}F_{10,36} = 1138.55$, $\alpha < 0.0001$).

Although saline solution has a higher pH than gastric fluid, there is less significant difference ($\alpha < 0.05$) between their swelling profiles. This can be explained based on the results published by Kriwet and Kissel (1996). They observed that the particle size of polymer (carbophil) decreased with the addition of sodium chloride (NaCl). The presence of NaCl was found to cause dehydration of polymer and compensation of the negative charges through binding of sodium counterions to the carboxylic groups. Both effects will result in the decrease of swelling of polymers in saline. In buffered intestinal medium, the above effect of counterions is less obvious and the polymer becomes more hydrophilic and extensive swelling occurs as a result of increased ionization at higher pH 7.5.

The swelling of methacrylic polymers is significantly less ($\alpha < 0.001$) than that of acrylic polymers. This behaviour is mainly attributed to the cohesion of the hydrophobic methyl groups that resist expansion of the polymer chain.

It is worth mentioning that during the swelling studies, two polymers (PMDG, PMDH) showed a decline in their swelling after achieving their maximum volumes (Fig. 1b, Fig. 2b and Fig. 3b). This behaviour was mainly related to the tendency of

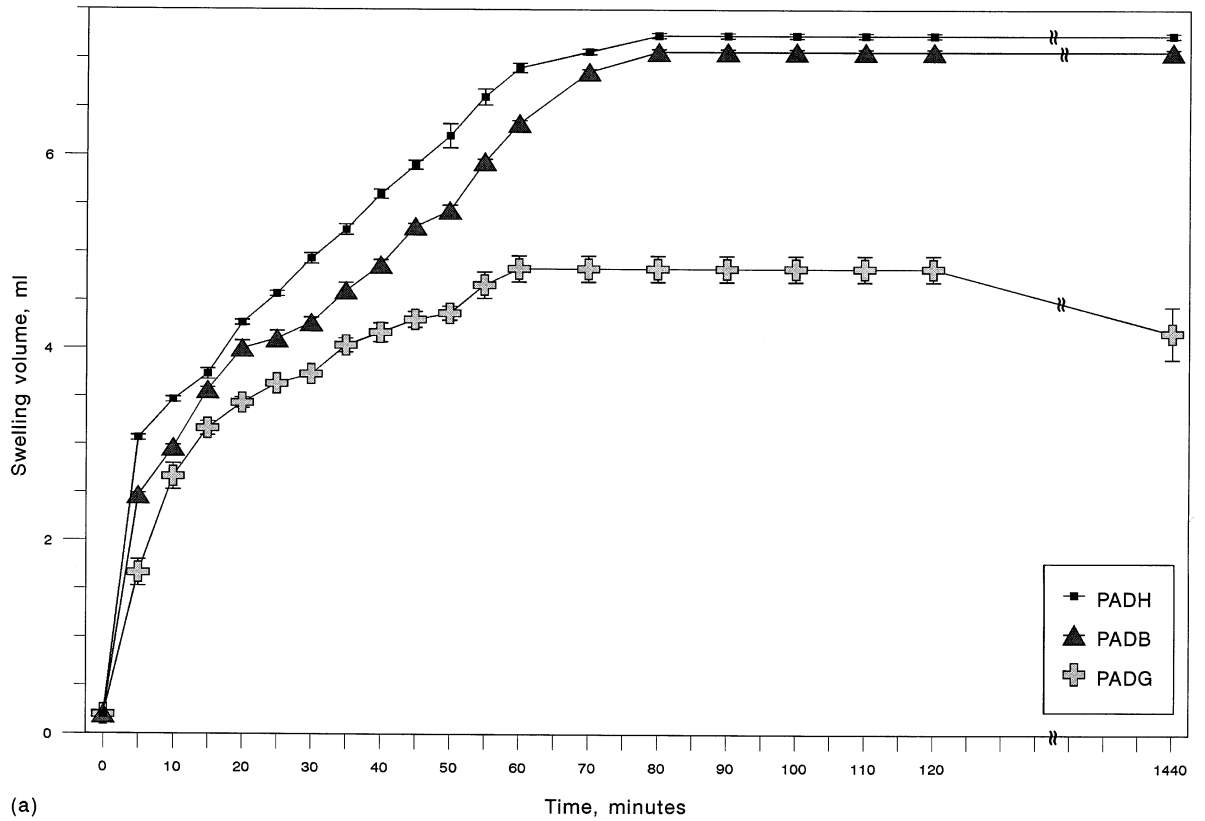

Fig. 3. Swelling profiles of acrylic (a) and metahacrylic polymers (b) in simulated intestinal fluid. Each point represents the mean ± S.E. of triplicate experiments. Note: The S.E. is less than the point size for some points.

these two polymers to dissolve in these media. The same behaviour was also noted for PADG in intestinal media (Fig. 3a). This finding may present some practical difficulties in generating some results. However, it may offer other advantages for formulating these polymers into film forms for buccal and wound applications.

3.2. Zeta potential

The present attempt to determine the zeta potential of polymer particles must be seen in the light of the presence of some experimental obstacles in obtaining accurate and meaningful data due to the swelling characteristic of polymers. The effect of the swelling on the EPM cannot be neglected since increased hydration tends to decrease the mobility of the polymer particle as a result of increased particle size. This in turn exerts a pronounced effect upon the calculation of the zeta potential; for example, in calculating the zeta potential for polymer dispersed in intestinal fluid, compensation for the swelling effect needs to be determined. In the present study we were unable to set a factor to correct for the equivalent zeta potential. Thus assumption had to be made when true data were neither available nor accessible by measurements.

The results of the zeta potential for various polymers in the presence of the three different fluids are given in Table 1. All the polymers exhibited a negative mobility due to the presence of carboxylic groups at the polymer surface. The magnitude of the zeta potential was varied and found to be dependent on the pH of the medium ($\alpha < 0.0001$, two-way ANOVA) and the extent of

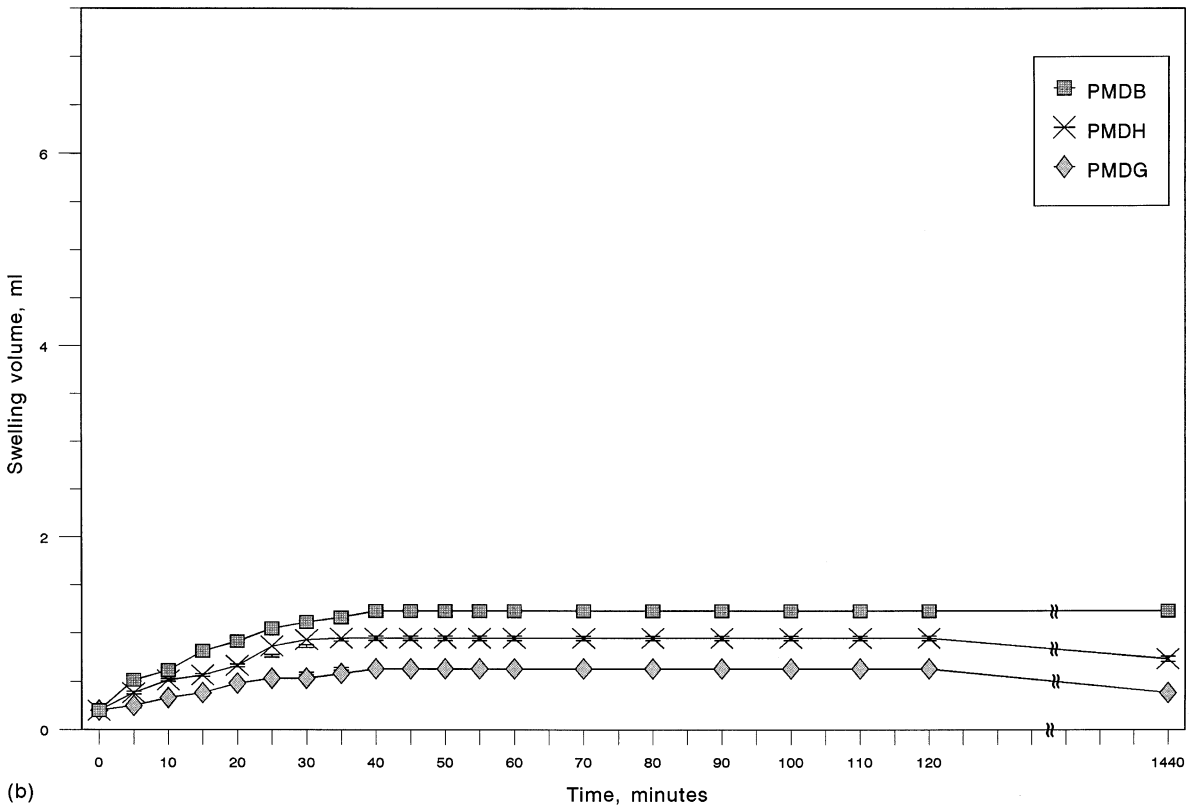

Fig. 3. (*Continued*)

swelling. The higher the pH the more ionization occurred until full ionization was achieved. Thus the surface charge density increased with the degree of ionization, leading to a more negative zeta potential.

Table 1
The zeta potential of various crosslinked polymers in the presence of different physiologically relevant fluids[a]

Polymer	Zeta potential (mV)		
	Isotonic saline	Gastric fluid	Intestinal fluid
PADH	169.4 ± 1.5	153.4 ± 1.4	50.5 ± 0.3
PADB	126.1 ± 0.9	114.5 ± 1.0	45.8 ± 0.4
PADG	100.7 ± 2.3	80.3 ± 0.8	39.9 ± 0.9
PMDG	130.7 ± 1.3	118.3 ± 0.8	80.0 ± 0.6
PMDH	122.3 ± 0.4	112.4 ± 0.4	75.7 ± 0.1
PMDB	116.2 ± 0.7	102.5 ± 1.6	72.1 ± 0.4

[a] Data are presented as mean ± S.E. of 20 readings.

In this study, polymers exhibit higher negative zeta potential in saline solution compared to acidic medium ($\alpha < 0.05$) due to greater ionization of carboxylic groups. However, the increase is not as much as expected despite the greater increase in pH when compared with acidic medium. The main reason is probably due to the presence of sodium counterions in close proximity of the polymer surface that interacts with the ionized carboxylic groups. The above effect and the additional swelling at pH 7.5 are thought to be the reasons behind the reduced negative zeta potential and the consequent slower movement of the particles in the intestinal medium. Thus, it is difficult to rank the two groups of polymers for their negative zeta potentials in the same medium. Instead, the ranking order is found between each group of polymers i.e. acrylic and methacrylic polymers in all three media: PADH > PADB > PADG and PMDG > PMDH > PMDB.

Table 2
In vitro evaluation of polymer bioadhesion to rabbit stomach tissue in the presence of isotonic saline solution[a]

Polymer	Weight required for detachment (mg)	Force (dyne)	Force/area (dyne/cm^2)
PADH	1010 ± 24	991 ± 21	1262 ± 27
PADB	920 ± 12	903 ± 11	1150 ± 14
PADG	820 ± 12	804 ± 11	1025 ± 14
PMDG	740 ± 19	726 ± 16	925 ± 21
PMDH	670 ± 12	657 ± 11	837 ± 14
PMDB	500 ± 16	491 ± 14	625 ± 18

[a] Data are presented as mean $\pm$S.E. of five determinations.

Table 3
In vitro evaluation of polymer bioadhesion to rabbit stomach tissue in the presence of simulated gastric fluid[a]

Polymer	Weight required for detachment (mg)	Force (dyne)	Force/area (dyne/cm^2)
PADH	860 ± 19	844 ± 16	1075 ± 21
PADB	800 ± 16	785 ± 14	1000 ± 18
PADG	730 ± 20	706 ± 11	900 ± 14
PMDG	660 ± 19	647 ± 16	825 ± 21
PMDH	600 ± 16	589 ± 14	750 ± 18
PMDB	370 ± 25	363 ± 22	462 ± 28

[a] Data are presented as mean $\pm$S.E. of five determinations.

3.3. Bioadhesion studies

An initial investigation using 2, 5 and 10 g contact forces to test their influence on bioadhesion revealed a significant, measurable resistance to detach the polymer from the rabbit tissue. There was a significant increase in the force required for detachment when the applied force was increased from 2 to 5 g and from 5 to 10 g. Although the latter two applied forces provided a greater surface area for interaction between the two surfaces, its use to compare acrylic and methacrylic polymers that had different hydration profiles was not recommended. The methacrylic polymers that were less hydrated would be pressed into the cell surface instead of just interacting with the mucin layer and consequently it could result in a higher force required for detachment than acrylic polymers. This may lead to difficulties in obtaining a consistent ranking order between the two groups. For this reason, an applied force of 2 g was chosen. This force caused insignificant deformation of the mucosa but was found to provide enough surface area of contact between the hydrated polymer particle and the topmost layer of the mucus, with the polymer particles being in contact with more than the 'peaks' of the rough mucus layer. Furthermore, in a separate study to test the effect of the applied forces on the adhesion between mucin layers of two stomach tissues, the 2 g force showed negligible adhesion compared to the other two forces.

The mean maximum forces required to break the adhesive bond between various polymers and the rabbit gastric mucosa in the presence of different media are shown in Tables 2–4. Two-way ANOVA indicated that the effects on adhesion of polymers (P), medium (M) and the interaction between them ($P \times M$) were all significant (P:$F_{5,72} = 87.32$, $\alpha < 0.0001$; M:$F_{2,72} = 1889.67$, $\alpha < 0.0001$; $P \times M$:$F_{10,72} = 83.48$, $\alpha < 0.0001$). Duncan's multiple comparison showed significant differences ($\alpha < 0.05$) in the force required for detachment among the tested polymers in the same and different media. The methacrylic polymers required a lower force to detach than acrylic polymers. The reduction in the bioadhesion behaviour is mainly attributed to the introduction of

Table 4
In vitro evaluation of polymer bioadhesion to rabbit stomach tissue in the presence of simulated intestinal fluid[a]

Polymer	Weight required for detachment (mg)	Force (dyne)	Force/area (dyne/cm^2)
PADH	—[b]	—[b]	—[b]
PADB	—[b]	—[b]	—[b]
PADG	—[b]	—[b]	—[b]
PMDG	400 ± 35	392 ± 31	500 ± 40
PMDH	302 ± 34	316 ± 40	402 ± 50
PMDB	134 ± 19	132 ± 17	167 ± 21

[a] Data are presented as mean $\pm$ S.E. of determinations
[b] Not determined due to extensive swelling.

the methyl group in the polymer structure that results in an increased hydrophobicity (Ch'ng et al., 1985). The hydrophobic structure of the polymer network will give rise to chain interactions and decreased chain flexibility. The rank order of bioadhesiveness is found to be (from highest to lowest): PADH > PADB > PADG > PMDG > PMDH > PMDB. All six polymers show strongest adhesion in the saline medium, intermediate in the acidic gastric fluid, and weakest in the buffered intestinal fluid, which agrees with the previous data obtained by Lehr et al. (1992). These results can be explained on the basis of different extent of ionization and interpenetration of molecular chains that occur as a result of pH changes and applied force. In gastric fluid, mucin carboxylic acid groups (from terminal sialic group) will be less ionized ($pK_a = 2.6$). Similarly, carboxylic groups in the polymer structure will be in protonated form with a small degree of ionization resulting in a limited amount of uncoiling of the polymer, which in turn produces only a small amount of entanglement and penetration of polymer chains with mucin molecules even when the force is applied. However, intermediate adhesion may occur through hydrogen bonding of the unionized carboxylic groups. In saline medium, both the polymer and the mucin will be more ionized with the increase in pH and hence produces suitable degree of ionization which ensures sufficient network expansion without actually being over-hydrated. This suitable amount of uncoiling of the polymer chains will produce good adhesion through strong entanglement and penetration with the mucin chains. The same explanation was also offered by Hassan and Gallo (1990). In contrast, the higher content of water absorbed by the polymer in intestinal fluid tends to change the polymer to a loosely packed structure. This extensive uncoiling of the polymer chain can lead to a reduction of its mechanical entanglement and penetration and coupled with stronger repulsion between the ionized carboxylic groups can result in insignificant adhesion with the mucin as shown in Table 4.

3.4. Mucoadhesion screening of polymers by colloidal gold technique

The preliminary study on the stability of mucin-gold conjugate in isotonic saline, simulated gastric and simulated intestinal solutions showed the formation of stable red conjugates without any aggregation (Fig. 4). The percentages of change in the colour intensity over a period of 24 h were only 2.3, 4.5 and 1.6% respectively. These insignificant changes made it possible to study the interaction between the polymer and the mucin in the presence of these media.

The adsorption of mucin-gold conjugates on the polymer surface represented the extent of adhesion and interaction between polymer chains and mucin molecules. The highest adsorption was obtained in acidic medium and to a lesser extent in isotonic saline and intestinal fluid. The colloidal gold technique was only able to measure the surface interaction between polymer and mucin in acidic condition. The increased ionization of the polymer and the mucin in saline and intestinal media would increase the possibility of

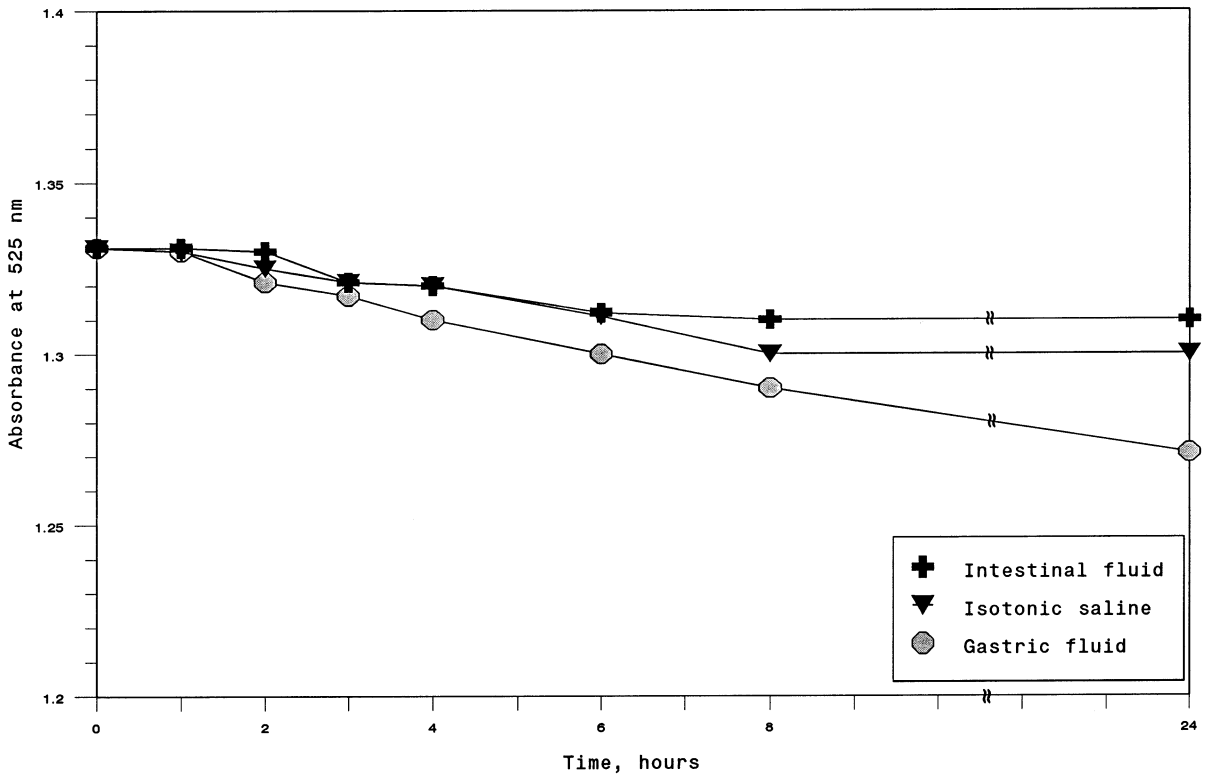

Fig. 4. Stability of mucin-gold conjugates as a function of time in different physiological fluids.

the surface charge repulsion at the mucin–polymer interface resulting in less interaction between the two surfaces.

The kinetics of the adsorption process for various polymers in simulated gastric fluid are shown in Fig. 5. The extent of mucoadhesion for all cross-linked polyacrylic acid polymers seems to increase as a function of time, reaching the maximum at about 8 h. However, for polymethacrylic acid polymers the adsorption is slow and no saturation values are obtained after 24 h. One-way ANOVA applied to the mean results indicates that the amount of mucin adsorbed on the surface of polymers is significant ($\alpha < 0.0001$). Duncan's multiple range test performed at $\alpha = 0.05$ indicates that all the polymers are significantly different, except between PMDG and PMDH. Examination of the results at different time periods show that the extent of adhesion is in the following order (from highest to lowest):

PADH > PADB > PADG > PMDB > PMDH > PMDG, being the same ranking generated by using gastric tissue except for PMDB and PMDG polymers which are in reverse positions (Table 3). The decreased adhesion of PMDG can be seen as a direct result of its solubility behaviour in the tested media as shown in the swelling studies (Fig. 2b).

The methacrylic polymers show lower interaction than acrylic polymers. This may be due to the increased hydrophobicity of methacrylic polymers and the tendency of its chains to interact to each other. This in turn leads to a decrease in chain flexibility and also to prevent the carboxyl groups from dissociation.

The mechanism of adhesion in gastric condition can be explained based on the studies conducted by Gandhi and Robinson (1991) on the permselective characteristics of the rabbit buccal mucosa. They reported that there is a cationic selectivity of

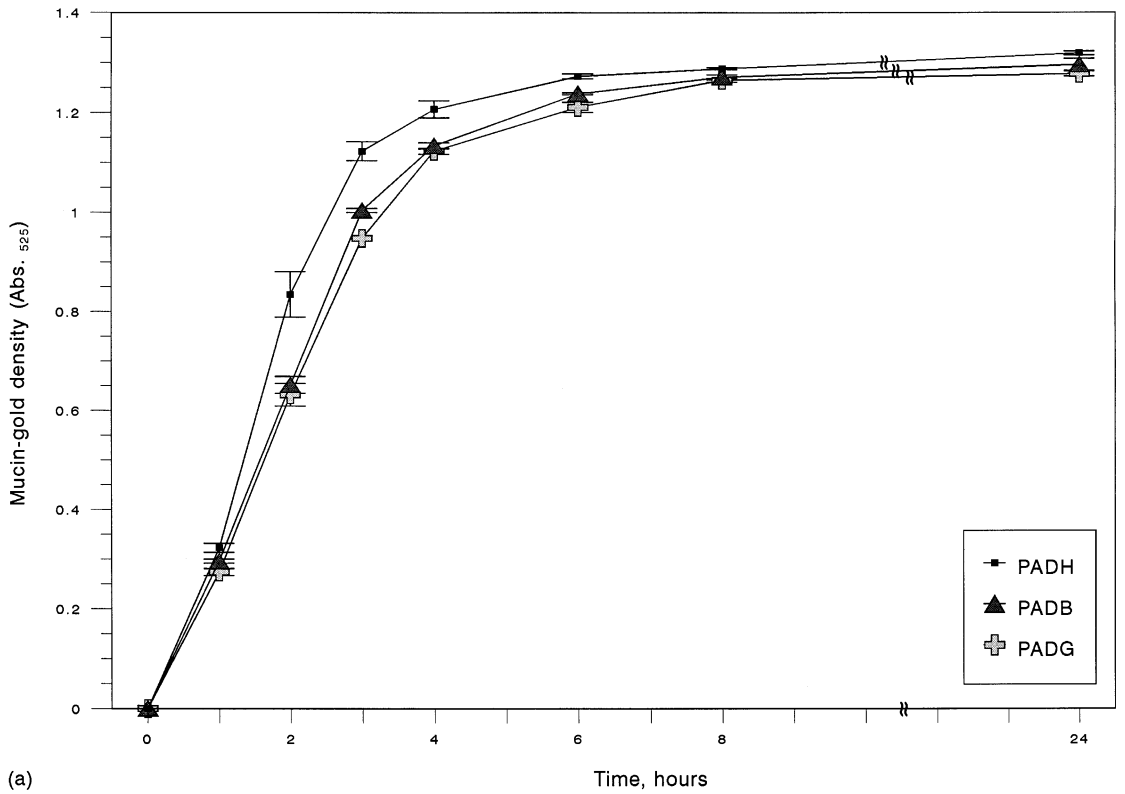

Fig. 5. The density of mucin-gold conjugates on acrylic (a) and methacrylic polymers (b) as a function of time in the presence of simulated gastric fluid. Each point is an average $\pm$S.E. of triplicate experiments. Note: Staining was carried out using mucin-gold concentration of $Abs_{525} = 1.331$.

the membrane above pH 2.6 and anionic selectivity below that pH. In this study, we expect the same behaviour for mucin as a function of pH. Changes in pH will effect ionization of the sialic acid (pK_a of 2.6) and amino acids in the peptide backbone of the mucin making the mucin negatively charged above that pH (Longer and Robinson, 1986). At acidic pH below the pK_a, the mucin becomes protonated and will bear a net positive charge (Gandhi and Robinson, 1991). Similarly, carboxylic groups in the polymer structure will undergo a different degree of ionization at a different pH. At acidic pH, Park and Robinson (1987) reported the presence of about 80% of the carboxylic groups in the protonated form. This percentage was quoted by the same investigators as the critical carboxyl-group necessary to favour adhesion. Thus the high binding ability in acidic media is mainly due to the presence of this number of carboxylic groups in an undissociated form that favours hydrogen bonding. It seems that the availability of this critical percentage capable of hydrogen bonding is less in the other two media due to ionization. At higher pH such as in saline and intestinal media, both mucin and polymer become ionized and the negative charge repulsion will be considerably increased in the mucin–polymer interaction. This behaviour could be the reason behind the reduced interaction between the polymer and the mucin in these media in our experiments. In this study, adhesion increases at gastric pH 1.3 which is below the pK_a of the mucin molecule. At this pH, mucin carries a net positive charge on the peptide chain while, in contrast, polymers carry a net negative charge as indicated by the zeta potential

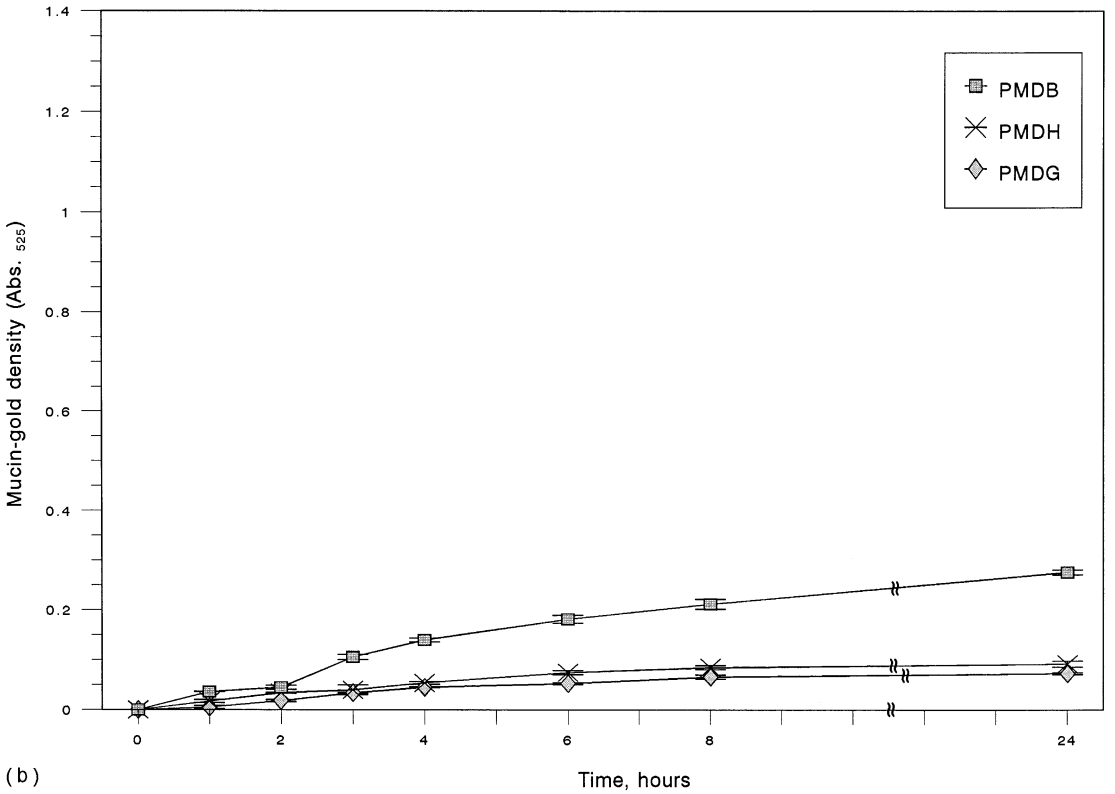

Fig. 5. (*Continued*)

measurement. Hence, an opposite charge favours adhesion. The mechanism of adhesion is therefore predominantly by primary electrostatic interaction followed by secondary hydrogen bonding.

The results obtained using colloidal gold staining were different from that of Texture analyzer. We observed a sharp drop in adhesion of polymers in saline solution using colloidal gold staining, an effect that is not observed using gastric tissue. The disagreement between the results could be due to the difference between the methods used. It can be argued that the Texture analyzer method measures all interaction involved during adhesion such as chain entanglement and interpenetration rather than the ionic interaction which may participate in the binding of the polymer to the mucin in colloidal-gold staining method. In the latter case, the individual mucin-gold conjugate interacts only with the surface chains of the polymer. Thus, the surface area available for interaction is more important than the entanglement of the mucin molecules. In Texture analyzer method, the applied force, although small, is sufficient to overcome the repulsive force generated from the ionization of carboxylic groups in the saline medium and allows the mechanical interlocking of the molecular chains to occur; hence the higher force required for detachment.

4. Conclusion

The zeta potentials of different polymers in saline and gastric media show reasonable correlation to their bioadhesive performance determined by the detachment experiment. The zeta potential of polymers can provide the initial driving force for bioadhesion, followed by mutual interpenetration and subsequent establishment of secondary

bonds through non-specific physical entanglement when pressure is applied. It is also inferred that the role of electrostatic bonding becomes insignificant when charge repulsion dominates at higher pH such as in simulated intestinal fluid.

The present study indicates that in order to deliver a bioadhesive dosage form effectively to the targeted tissue, one must control the degree of ionization of the polymer by manipulating the pH of the media, or alternatively choose a bioadhesive with a pK_a that can provide a suitable extent of ionization. This information is essential in the design of bioadhesive dosage forms since external pressure cannot be applied to them in the GIT. Therefore, what is required under these conditions is a mucoadhesive which demonstrates significant adhesion at low contact pressure. Acrylic polymers are suitable candidates for this purpose.

Acknowledgements

The authors are deeply indebted to the School of Pharmaceutical Sciences, USM for providing the equipment and facilities needed for this work and to the Libyan government for granting the scholarship for K.M.T.

References

Ch'ng, H.S., Park, H., Kelly, P., Robinson, J.R., 1985. Bioadhesive polymers as platforms for oral controlled drug delivery II: Synthesis and evaluation of some swelling, water-insoluble bioadhesive polymers. J. Pharm. Sci. 74, 399–405.

Gandhi, R.B., Robinson, J.R., 1991. Permselective characteristics of rabbit buccal mucosa. Pharm. Res. 8, 1199–1202.

Garcia-Gonzalez, N., Kellaway, I.W., Blanco-Fuente, H., Anguiano-Igea, S., Delagado-Charro, B., Otero-Espinar, F.J., Blanco-Mendez, J., 1993. Design and evaluation of buccoadhesive metoclopramide hydrogels composed of poly(-acrylic acid) crosslinked with sucrose. Int. J. Pharm. 100, 65–70.

Hassan, E.E., Gallo, J.M., 1990. A simple rheological method for the in vitro assessment of mucin–polymer bioadhesive bond strength. Pharm. Res. 7, 491–495.

Hui, H.W., Robinson, J.R., 1985. Ocular delivery of progesterone using a bioadhesive polymer. Int. J. Pharm. 26, 203–213.

Kriwet, B., Kissel, T., 1996. Interactions between bioadhesive poly (acrylic acid) and calcium ions. Int. J. Pharm. 127, 135–145.

Lehr, C.M., Bouwstra, J.A., Bodde, H.E., Junginger, H.E., 1992. A surface energy analysis of mucoadhesion: contact angle measurements on polycarbophil and pig intestinal mucosa in physiologically relevant fluids. Pharm. Res. 9, 70–75.

Lehr, C.M., Bodde, H.E., Bouwstra, J.A., Junginger, H.E., 1993. A surface energy analysis of mucoadhesion II. Prediction of mucoadhesive performance by spreading coefficients. Eur. J. Pharm. Sci. 1, 19–30.

Longer, M.A., Ch'ng, H.S., Robinson, J.R., 1985. Bioadhesive polymers as platforms for oral controlled drug delivery III: oral delivery of chlorothiazide using a bioadhesive polymer. J. Pharm. Sci. 74, 406–411.

Longer, M.A., Robinson, J.R., 1986. Fundamental aspects of bioadhesion. Pharm. Int. 7, 114–117.

Park, K., 1989. A new approach to study mucoadhesion: colloidal gold staining. Int. J. Pharm. 53, 209–217.

Park, H., Robinson, J.R., 1985. Physico-chemical properties of water insoluble polymers important to mucin/epithelial adhesion. J. Control. Release 2, 47–57.

Park, H., Robinson, J.R., 1987. Mechanisms of mucoadhesion of poly(acrylic acid) hydrogels. Pharm. Res. 4, 457–464.

Peppas, N.A., Buri, P.A., 1985. Surface, interfacial and molecular aspects of polymer bioadhesion on soft tissues. J. Control. Release 2, 257–275.

Rillosi, M., Buckton, G., 1995. Modelling mucoadhesion by use of surface energy terms obtained by Lewis acid–Lewis base approach. Int. J. Pharm. 117, 75–84.

.