Regular Article

Preparation and Characterization of Hydroxypropyl Methylcellulose/Polycarbophil Mucoadhesive Blend Films Using a Mixture Design Approach

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The objectives of this study were to prepare the hydroxypropyl methylcellulose (HPMC)/polycarbophil (PC) mucoadhesive blend film and to investigate the main and interaction effect of HPMC and PC mixtures on the physicochemical and mechanical properties of blend films using a simplex lattice mixture design approach. The cubic and quadratic models were selected to analyze mucoadhesive properties in terms of work of adhesion and maximum detachment force, respectively. It was shown that HPMC/PC blend film had higher mucoadhesive properties than pure HPMC film. The suitable models for analyzing swelling index of blend films at various times were assessed. The puncture strength, % elongation and hydrophilicity of films were also examined. The pure HPMC film displayed more homogeneous and smoother structures compared with the blend film, as observed by scanning electron microscope and atomic force microscopy. Intermolecular hydrogen bonding between HPMC and PC was detected using Fourier transform infrared and X-ray diffraction. Therefore, the blend film shows high potential for use as a buccal delivery system.

Key words hydroxypropyl methylcellulose (HPMC); polycarbophil; blend film; mixture design; mucoadhesive film; buccal drug delivery

Buccal drug delivery systems have several advantages such as good accessibility, avoiding hepatic first pass metabolism and drug degradation in the gastrointestinal tract.1,2) However, it is very difficult to apply the conventional pharmaceutical dosage forms onto the buccal mucosa, and their effects are expected to remain for a long period of time. Consequently, many bioadhesive buccal formulations such as tablets, patches and films have been investigated using mucoadhesive polymers.3) Mucoadhesive films are a very interesting dosage form for the buccal delivery of drugs owing to their small size and reduced thickness, which improves patient compliance compared to tablets, for example.3) Additionally, mucoadhesive films should be flexible, elastic, soft and sufficiently strong to withstand breakage due to stress from mouth movements. It must also possess a strong adhesive contact with the buccal mucosa in order to increase the desired duration of action and to optimize drug bioavailability.3) Several polymers have been studied for application as mucoadhesive buccal films, especially hydrophilic polymers.2,4,5)

Hydroxypropylmethyl cellulose (HPMC) is one of the most general hydrophilic polymers, which is extensively used in many fields owing to its ease of use, excellent film-forming ability, good biocompatibility and biodegradability.5,7) In addition, HPMC has been displayed to possess hydrogel-forming properties, which are essential for mucoadhesion, and it also has rapid swelling due to high water absorbing capacity, including displaying moderate mucoadhesive properties as well.5,8) Hence, HPMC is a popular hydrophilic polymer used in the formulation of mucoadhesive films. To improve the mucoadhesive and physical properties of HPMC films, one efficient and interesting technique is blend films. Blend films are made from mixing two different polymers, so it is expected that their properties should therefore be better.1,7,9) The mucoadhesive and physical properties of blend films strongly depend on the constituting polymer characteristics and their compatibility.9)

Polycarbophil (PC) is a high molecular weight acrylic acid polymer cross-linked with divinyl glycol, therefore possessing a large number of carboxyl groups on the molecular structure.10,11) The pK_a of PC is around 6.0±0.5 and it can change from sol to gel at a pH of the environment that is higher than its pK_a.10) It is a good water absorbable polymer and has been displayed to have excellent bioadhesive properties.10,12) Hence, PC has been vastly used in bioadhesive formulations for various mucus membranes such as buccal, vaginal, nasal, rectal, and ophthalmic membranes.10,12–14)

As both HPMC and PC possess good biological and mucoadhesive properties, some studies have mixed HPMC and PC for the preparation of HPMC/PC blend films used in bioadhesive drug delivery systems.8) They were compared with other polymers (carbopol, polyvinyl alcohol (PVA) and chitosan) and the physical characteristics of blend films were evaluated using thickness, swelling properties, bioadhesive properties, etc. They found that the successful delivery by blend films as mucoadhesive drug delivery systems depends on the amount of PC in the formulations being optimized. However, there is little information on the comprehensive characterization of the HPMC/PC blend films including various ratios in their formulations. Additionally, they used a traditional method that involves trial and error methodology, by keeping one variable constant while

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changing others one at a time.\textsuperscript{15,16} This approach is quite time consuming, expensive, laborious and unpredictable.\textsuperscript{15,16} Statistical methods, like design of experiment (DoE), are systematic and scientific approaches for studying the relationship between variables.\textsuperscript{15} They not only resolve the problems of traditional methods efficiently, but also provide a method for obtaining mathematical models for characterizing formulations and processes.\textsuperscript{16} DoE analysis utilizes a response surface methodology (RSM) to estimate the main effect, the interaction effect and the outline of the response surface; many studies have successfully utilized RSM.\textsuperscript{16–18} The simplex lattice mixture design is one type of RSM design that is used when the response changes as a function of the relative proportions of the components. All components must be entered in the same units of measure and each run must sum to the same total.\textsuperscript{19}

The aims of this study were to prepare the HPMC/PC mucoadhesive blend film and to investigate the main and interaction effect of HPMC and PC mixtures on the physicochemical and mechanical properties of blend films using a simplex lattice mixture design approach.

**Experimental**

**Materials** Hydroxypropyl methylcellulose, HPMC K15M (Methocel\textsuperscript{\textregistered} K15M) was manufactured by Dow Chemical Company (MI, U.S.A.) and kindly supported by Rama Production Co., Ltd. (Bangkok, Thailand). Polyacrylphile (Noveon\textsuperscript{\textregistered} AA-1) was produced by Lubrizol Company (OH, U.S.A.) and supplied by Namsiang Co., Ltd. (Bangkok, Thailand). All other chemicals used were of analytical grade and used as received.

**Preparation of HPMC/PC Blend Film** HPMC/PC blend films were prepared by the film casting technique. Briefly, at a ratio of HPMC:PC at 0.95:0.5 (HPMC95/PC5), HPMC solution was prepared by dispersing HPMC powder (0.95 g) in 70 mL of deionized water and constantly stirred using a magnetic stirrer at room temperature until a clear solution was obtained. PC powder (0.05 g) was dispersed in 30 mL of deionized water and stirred until a clear solution was obtained. The PC solution was slowly poured into the HPMC solution and mixed for 30 min until the mixture was homogeneous. Then, 0.5 g of glycerol used in a plasticizer was added to the composite solution and further mixed for 30 min. The composite solution was poured onto a glass plate and allowed to evaporate at 50°C for 10 h in a hot air oven. The dried film was peeled off and kept in a vacuum desiccator prior to testing. Various ratios of blend films (HPMC and PC), at 1.0:0.0 (HPMC100), 0.975:0.025 (HPMC97.5/PC2.5), 0.925:0.075 (HPMC92.5/PC7.5) and 0.9:0.1 (HPMC90/PC10) by weight, were prepared as described above.

**Film Thickness** The film thickness was measured at five points with a thickness gauge Mini Test 600 (ElektroPhysik Dr. Steingroever GmbH & Co., KG, Germany).

**Experimental Design and Statistical Analysis** A simplex lattice mixture design was used to evaluate the effect of HPMC ($X_1$) and PC concentrations ($X_2$) on the physicochemical properties of the HPMC/PC blend films. Based on the initial screening, the concentrations of PC and HPMC were found so that the HPMC/PC blend films could be formed as films; the upper and lower limits of concentration levels of each component were set as follows:

- $90 \leq X_1 \leq 100\%$ (1)
- $0 \leq X_2 \leq 10\%$ (2)
- $X_1 + X_2 = 100\%$ (3)

Consequently, the components of the HPMC/PC blend films were studied by changing their concentration concurrently, and the total concentration of the two components was adjusted to 100%, as described in Eqs. 1–3.

The characteristics of HPMC/PC blend films such as contact angle ($\gamma_f$), surface free energy ($\gamma_f^p$), puncture strength ($\gamma_p^\alpha$), % elongation ($\gamma_p^\beta$), work of adhesion ($\gamma^d$), maximum detachment force ($\gamma^L$), and swelling time at 10 min ($\gamma^p_1$), 20 min ($\gamma^p_2$), 30 min ($\gamma^p_3$) and 60 min ($\gamma^p_4$) were used as the response (dependent variables). The responses of all model formulations were evaluated by Design-Expert\textsuperscript{\textregistered} Software (version 9; Stat-Ease, Inc., MN, U.S.A.). In total, 16 experiments (with 5 proportions of polymers) were designed by the software and were run in a random order. The suitable models chosen for the design were linear (Eq. 4), quadratic (Eq. 5), cubic (Eq. 6) and quartic models (Eq. 7). Several statistical parameters were used to compare with the best fitting mathematical model, verified by Design-Expert\textsuperscript{\textregistered} Software.

$$Y = \beta_1 X_1 + \beta_2 X_2$$

(4)

$$Y = \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_1 X_2$$

(5)

$$Y = \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_1 X_2 + \beta_4 X_1 X_2 (X_1 - X_2)$$

(6)

$$Y = \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_1 X_2 + \beta_4 X_1 X_2 (X_1 - X_2) + \beta_5 X_1 X_2 (X_1 - X_2)^2$$

(7)

**Wettability and Surface Free Energy Measurement** The wettability of the HPMC–PC blend films was determined by contact angle measurement which was carried out by sessile drop technique using a drop shape instrument FTA 1000 (First Ten Angstroms, U.S.A.). The surface free energy of the film samples were calculated using the Wu harmonic mean equation (Eqs. 8, 9), which were determined based on measurements of the contact angle of three distinct standard liquids i.e., distilled water, formamide and ethylene glycol at 25°C.

$$\gamma_s = \gamma_s^p + \gamma_s^d$$

(8)

$$(1 + \cos \theta) \gamma_s = 4 \left( \frac{\gamma_s^p \gamma_f^d}{\gamma_s^p} + \gamma_f^d \right) + 4 \left( \frac{\gamma_f^p \gamma_f^d}{\gamma_s^p} + \gamma_f^d \right)$$

(9)

where $\gamma_s$ is total surface free energy, $\gamma_s^p$, $\gamma_s^d$ are polar and dispersive forces of the blend film, respectively, $\gamma_f^p$, $\gamma_f^d$ are polar and dispersive forces of standard liquid, respectively. $\theta$ is the contact angle between the blend film and the standard liquid.

**Mechanical Properties** The mechanical properties of the HPMC/PC blend films were determined using a Texture Analyzer (TA.XT.plus Texture Analyzer, Stable Micro Systems, U.K.) as previously described in Limmatvapirat & Pongjanyakul.\textsuperscript{20,21} Briefly, the blend films were cut into 3.5×0.6 cm rectangles. The blend films were placed in the film holder between two mounting plates, following which the holding screws were tightened to prevent slippage of the films. The probe was slowly driven through the film with a
speed of 0.1 mm/s and force-displacement curves were recorded through a 500 N loaded cell. The maximum force and maximum displacement of films were measured, and then converted to puncture strength and elongation at puncture. The parameters were calculated using the following equations:

\[ \text{Puncture strength} = \frac{F}{A} \]  
(10)

where \( F \) is maximum force for film puncture, and \( A \) is the cross-sectional area of the film located in the path of the cylindrical area of the edge of the film holder and

\[ \text{Elongation} = \frac{\sqrt{r^2 + D^2} - r}{r} \times 100 \]  
(11)

where \( r \) is the radius of the film exposed in the cylindrical hole of the film holder and \( D \) is the displacement of the probe from the point of contact to the point of film puncture.

**Mucoadhesive Properties** Porcine buccal mucosa was used in this study as a biological membrane because of the similarity to human buccal tissue.21 The mucoadhesive property studies were approved by the Laboratory Animal Center Ethical Committee of Thammasat University (No. 018/2559, Thailand). Porcine buccal mucosa was obtained from freshly killed pigs after slaughter at a local slaughterhouse (Nakhon Pathom, Thailand). The buccal tissues were washed with deionized water then placed in normal saline solution at 4°C and immediately used for tests. The underlying connective tissue was subsequently removed to isolate the mucosal membrane.22

The mucoadhesive properties of the HPMC/PC blend films were measured using a texture analyzer (TA.XT.plus, Stable Micro Systems Ltd.) with 50 N load cell equipped with mucoadhesive holder.22 A HPMC/PC blend film was attached to a 10 mm diameter cylindrical probe by double-sided adhesive tape. The tissue (about 2.0 × 2.0 cm) was equilibrated for 15 min at 37.0 ± 0.5°C before placing onto the holder stage of mucoadhesive holder with the mucosal surface facing up. Before testing, 100 µL of simulated saliva pH 6.80 was dropped onto the mucosa. The probe attached film speed was 1.0 mm/s and made contact with the tissue with a contact force of 0.05 N for 60 s. The probe withdrawal speed from the tissue was 0.5 mm/s. The relationship between force and film displacement was plotted. Maximum detachment force (\( F_{\text{max}} \)) and work of adhesion (\( W_{\text{ad}} \)), the area under the force versus distance curve were computed using the Texture Exponent 32 software.23

**Swelling Index** The swelling properties of HPMC/PC blend films were evaluated by determining the swelling index (SI).11 Each blend film was cut into 2.0 × 2.0 cm and simulated saliva fluid pH 6.80 (containing 16.748 g Na2HPO4·12H2O and 4.609 g KH2PO4 per liter of distilled water adjusted to pH 6.80 with NaOH or HCl) was used as medium in this study. Each blend film was weighed previously and placed on a pre-weighed stainless steel wire mesh. It was then dipped into simulated saliva fluid for predetermined periods of time. The blend films were wiped off from the excess surface water using filter paper and weighed. Swelling index was calculated using the following equation

\[ \text{Swelling index} = \frac{(W_t - W_0)}{W_0} \]  
(12)

where, \( W_t \) is the weight of film at time \( t \), and \( W_0 \) is the weight of film at time zero.

**Fourier Transform Infrared (FTIR) Spectroscopy** FTIR spectra of samples were measured by a FTIR spectrophotometer ( Nicolet, Magna 750, U.S.A.) using the KBr disc method. Each sample was pulverized, blended with KBr powder, and then compressed using a hydraulic press at a pressure of 5 t. The sample disc was placed in the sample holder and scanned from 4000 to 400 cm⁻¹ at a resolution of 4 cm⁻¹.

**Powder X-Ray Diffractometry** X-Ray diffraction (XRD) patterns of the samples were measured on a MiniFlex II (Rigaku, Japan) at 30 kV and 15 mA. The relative intensities were studied within the range of 5–40° 2θ using a CuKα radiation wavelength of 1.5406 Å.

**Morphology Examination (Scanning Electron Microscope (SEM))** The morphological examination of selected HPMC/PC blend films was performed by SEM (model JSM-5410LV, Jeol, Japan) with an accelerating voltage of 10 kV. The samples were mounted on metal stub with double-sided adhesive tape and coated with a fine gold layer under vacuum before obtaining the micrographs.

**Atomic Force Microscopy (AFM)** The morphology of selected HPMC/PC blend films was observed by AFM (SPA400, Seiko, Japan) in tapping mode at room temperature (ca. 25°C). The films were imaged by scanning 1.5 × 1.5 μm areas using HA HR cantilever with resonant frequencies at 230–380 kHz and a constant forces in the range from 17–34 N/m. A scan speed of each sample was 1 Hz and the data were analyzed using SPI4000 version 4.08F (SII Seiko Nanotechnology, Tokyo, Japan).

**Results and Discussion**

**Film Thickness** The thickness of HPMC/PC blend films with different concentrations varied from 0.019 to 0.032 mm, as shown in Table 1. The optimum film thickness had to be considered since it should not be too thick or too thin, potentially causing problems while handling or dissolving quickly.8

**Contact Angle (\( Y_i \)) and Surface Free Energy (\( Y_f \)) Measurement** Contact angle measurements were used to investigate the wettability and hydrophilicity of surface properties of films. The suitable models chosen for the design were linear, quadratic, cubic and quartic models. From Table 2, the quartic model was found to have a maximum R² (0.9873), adjusted R² (0.9827), predicted R² (0.9741), and also displayed low p-values (<0.05). Additionally, low standard deviation (S.D.) (0.58) and the smallest PRESS (7.65) data confirm that the quartic model was appropriate for analyzing response \( Y_i \).19,24,25 The response could be described by the following equation:

\[ Y_i = 73.1X_1 + 62.41X_2 - 0.42X_1X_2 \]
\[ - 19.26X_1X_2^2 - 94.15X_1X_2^3 - X_2^2 \]  
(13)

The contact angle of the HPMC/PC blend films is shown in Fig. 1A. It varied from 61.8 to 73.9 and exhibited the highest

Table 1. The Thickness of Various Ratios of HPMC/PC Blend Films

| Ratio of HPMC/PC blend films | Thickness (mm)*  |
|-----------------------------|-----------------|
| HPMC90/PC10                | 0.019 ± 0.002   |
| HPMC92.5/PC7.5             | 0.031 ± 0.003   |
| HPMC95/PC5                 | 0.029 ± 0.002   |
| HPMC97.5/PC2.5             | 0.032 ± 0.002   |
| HPMC100                    | 0.026 ± 0.002   |

*Values expressed as the mean ± S.D., n=5.
value in the HPMC100 film. The association of Eq. 13 and Fig. 1A shows that the coefficient of 73.1 for $X$ in the HPMC100 film. The regression equation, the positive sign of a factor shows a synergistic effect and vice versa in the negative sign.16,27) The higher coefficients mean that the independent variable has an obvious effect on the response. 25) The negative coefficient on nonlinear blending terms suggests that the mixture of the two components makes a response that is less than that which would be expected from linear blending.19) This means that the interaction in a mixture of HPMC and PC for film-forming may lead to reduction in the contact angle of blend films which results in lower wettability of their films. These results suggested that related factors do not depend on the amount of HPMC and PC only, but also depend on the interaction occurring between both compounds.

For response $Y_2$, the Quartic model was also found to be the most fitting model as shown in Table 2. The equation of this

Table 2. Regression Analysis for Response $Y_1 - Y_{10}$

| Response | $R^2$ | Adjusted $R^2$ | Predicted $R^2$ | Prob>F | $p$-Value | S.D. | PRESS | Model |
|----------|------|----------------|----------------|--------|------------|------|-------|-------|
| $Y_1$    | 0.7460 | 0.7278 | 0.6816 | <0.05 | 2.31 | 93.94 | Linear |
|          | 0.7823 | 0.7488 | 0.7036 | 0.1644 | 2.22 | 87.43 | Quadratic |
|          | 0.8216 | 0.7770 | 0.6475 | 0.1299 | 2.09 | 104.01 | Cubic |
|          | 0.9873 | 0.9827 | 0.9741 | <0.05 | 0.58 | 7.65 | Quartic* |
| $Y_2$    | 0.1548 | 0.0945 | -0.0164 | 0.1316 | 1.51 | 38.16 | Linear |
|          | 0.5425 | 0.4721 | 0.3734 | <0.05 | 1.13 | 23.52 | Quadratic |
|          | 0.5520 | 0.4400 | 0.1186 | 0.6226 | 1.18 | 33.09 | Cubic |
|          | 0.9435 | 0.9229 | 0.8695 | <0.05 | 0.44 | 4.90 | Quartic* |
| $Y_3$    | 0.4185 | 0.3770 | 0.2626 | <0.05 | 3.09 | 169.67 | Linear |
|          | 0.8454 | 0.8216 | 0.7758 | <0.05 | 1.65 | 51.57 | Quadratic* |
|          | 0.8606 | 0.8257 | 0.7712 | 0.2752 | 1.64 | 52.63 | Cubic |
|          | 0.8734 | 0.8274 | 0.7428 | 0.3134 | 1.63 | 59.18 | Quartic |
| $Y_4$    | 0.1463 | 0.0854 | -0.0942 | 0.1436 | 4.65 | 387.54 | Linear |
|          | 0.8291 | 0.8028 | 0.7384 | <0.05 | 2.16 | 92.65 | Quadratic |
|          | 0.8298 | 0.7872 | 0.6937 | 0.8349 | 2.24 | 108.49 | Cubic |
|          | 0.8878 | 0.8470 | 0.7941 | <0.05 | 1.90 | 72.91 | Quartic* |
| $Y_5$    | 0.6867 | 0.6643 | 0.5980 | <0.05 | 5.405E-003 | 5.247E-004 | Linear |
|          | 0.8453 | 0.8216 | 0.7727 | <0.05 | 3.941E-003 | 2.968E-004 | Quadratic |
|          | 0.9143 | 0.8928 | 0.8547 | <0.05 | 3.054E-003 | 1.897E-004 | Cubic* |
|          | 0.9252 | 0.8981 | 0.8669 | 0.2301 | 2.978E-003 | 1.738E-004 | Quartic |
| $Y_6$    | 0.7425 | 0.7241 | 0.6667 | <0.05 | 3.076E-003 | 1.714E-004 | Linear |
|          | 0.9562 | 0.9494 | 0.9328 | <0.05 | 1.317E-003 | 3.456E-005 | Cubic |
|          | 0.9562 | 0.9452 | 0.9184 | 0.9444 | 1.370E-003 | 4.200E-005 | Quartic |
|          | 0.9617 | 0.9478 | 0.9114 | 0.2339 | 1.338E-003 | 4.557E-005 | Quartic |
| $Y_7$    | 0.8376 | 0.8260 | 0.7917 | <0.05 | 0.030 | 0.016 | Linear* |
|          | 0.8695 | 0.8494 | 0.8079 | 0.0978 | 0.028 | 0.015 | Quadratic |
|          | 0.8746 | 0.8432 | 0.7763 | 0.5000 | 0.028 | 0.017 | Cubic |
|          | 0.8876 | 0.8468 | 0.7406 | 0.2822 | 0.028 | 0.020 | Quartic |
| $Y_8$    | 0.9257 | 0.9204 | 0.9002 | <0.05 | 0.030 | 0.017 | Linear* |
|          | 0.9328 | 0.9225 | 0.8941 | 0.2602 | 0.030 | 0.018 | Quadratic |
|          | 0.9411 | 0.9264 | 0.9012 | 0.2184 | 0.029 | 0.017 | Cubic |
|          | 0.9413 | 0.9200 | 0.8928 | 0.8555 | 0.030 | 0.018 | Quartic |
| $Y_9$    | 0.7202 | 0.7002 | 0.6646 | <0.05 | 0.062 | 0.065 | Linear |
|          | 0.7917 | 0.7596 | 0.7131 | 0.0546 | 0.056 | 0.056 | Quadratic |
|          | 0.8287 | 0.7859 | 0.6730 | 0.1334 | 0.053 | 0.063 | Cubic |
|          | 0.9627 | 0.9491 | 0.9282 | <0.05 | 0.026 | 0.014 | Quartic* |
| $Y_{10}$ | 0.7474 | 0.7294 | 0.6748 | <0.05 | 0.052 | 0.049 | Linear |
|          | 0.7599 | 0.7230 | 0.6529 | 0.4259 | 0.053 | 0.052 | Quadratic |
|          | 0.8979 | 0.8724 | 0.8207 | <0.05 | 0.036 | 0.027 | Cubic* |
|          | 0.8997 | 0.8632 | 0.7960 | 0.6714 | 0.037 | 0.031 | Quartic |

*The model was selected for fitting to that response.
response could be explained by:

\[ Y_2 = 40.98X_1 + 42.44X_2 - 12.34X_1X_2 - 3.38X_1X_2(X_1 - X_2) + 51.62X_1X_2(X_1 - X_2)^2 \]  

Equation 14 shows that the amount of HPMC (X_1) and PC (X_2) have a positive effect on Y_2 \((p < 0.05)\) and interaction terms \((p < 0.05)\) have a significant effect on Y_2. The result correlated with contact angle, as the interaction of HPMC and PC in film-forming had an impact on surface free energy. From Eq. 14, the coefficient of \(X_1X_2(X_1 - X_2)^2\) showed positive influence on response Y_2 and vice versa in the negative sign of \(X_1X_2\) and \(X_1X_2(X_1 - X_2)\). The magnitude of coefficient indicating the term of \(X_1X_2(X_1 - X_2)^2\) has a more favorable effect on response Y_2 than \(X_1X_2\) and \(X_1X_2(X_1 - X_2)\). It may be implied that the interaction between HPMC and PC molecules caused the reduction in surface free energy of blend films. Additionally, the conformation and surface molecular orientation of the both compounds may be effect on these results^6 and confirmed by using FTIR, XRD, SEM and AFM in further experiment.

The corresponding residual plot between run number and internally studentized residuals of Y_1 and Y_2, as shown in Figs. 2A and B, respectively. This was used to check the reliability of the response variables. In the entirely randomized run, the vertical spread of the internally studentized residuals was in line from bottom-to-top, suggesting that all data points lay within the limits of a 95% confidence interval.17,18,24

**Puncture Strength (Y_3) and % Elongation (Y_4)**  
Puncture
strength and % elongation are two important factors which are generally used in examining the mechanical properties of films. The interaction between polymers will impact on the mechanical properties of the blend polymers or films. For the response $Y_3$, the statistical parameters ($R^2$, adjusted $R^2$ and predicted $R^2$) of quadratic, cubic and quartic model did not show any clear differences, as displayed in Table 2. However, the statistical analysis of the quadratic model showed the most significant result ($p<0.05$) suggesting that this model was suitable for describing the response $Y_3$. The fitted equation associating the response $Y_3$ is shown in Eq. 15.

$$Y_3 = 22.12X_1 + 15.57X_2 + 22.20X_1X_2$$

(15)

Puncture strength of HPMC/PC blend films ranged from 14.2 to 27.3 MPa, as shown in Fig. 1C. From Eq. 15, it can be seen that all terms ($X_1$, $X_2$, $X_1X_2$) of blend films have a significantly positive influence on the puncture strength ($p<0.05$) indicating that puncture strength of the blend films could be increased with the amount of HPMC and PC; the interaction between HPMC and PC was high. Increasing the amount of HPMC in the blend films showed that the puncture strength tends to increase. The maximum puncture strength was reported for blend film at HPMC95/PC5, with no significant difference at HPMC97.5/PC2.5 and HPMC100. The result could be described by the phenomenon of the formation of an intermolecular hydrogen bond between the –OH group of HPMC and the –COOH group of PC, which limits the motion of the film matrix and promotes rigidity. Moreover, the...
addition of HPMC in the blend films leads to an increase in crystallinity, and subsequently increased puncture strength of the HPMC/PC blend films.\(^{28}\)

For the response \(Y_2\), a quadratic model was also found to be the most fitting model, as shown in Table 2. The fitted equation associating the response \(Y_2\) is shown in Eq. 16.

\[
Y_2 = 41.49 X_1 + 36.80 X_2 + 39.33 X_1 X_2 + 2.70 X_1^2 + 36.05 X_2^2 (16)
\]

% Elongation of HPMC/PC blend films ranged from 34.8 to 51.6, as shown in Fig. 1D. From Eq. 16, two main effects (\(X_1, X_2\)) can be seen and showed that significantly positive impact on the response \(Y_2\) (\(p<0.05\)) indicating % elongation of the blend films could be increased with the increasing amount of HPMC and PC. The interaction term \((X_1 X_2, X_1 X_2 - X_2 X_2)\) of blend films also has a positive influence on the % elongation excepting in terms of \(X_1 X_2 (X_1 - X_2)\), which showed a negative impact on response \(Y_2\). However, only \(X_1 X_2 (X_1 - X_2)\) and \(X_2 X_2\) of nonlinear blending terms were found to be highly significant (\(p<0.05\)), but the term of \(X_1 X_2 (X_1 - X_2)\) was ignored due to insignificant statistics for the response \(Y_2\). However, the coefficient of \(X_1 X_2 (X_1 - X_2)\) and \(X_2 X_2\), was different with the fact that the coefficient of \(X_1 X_2\) showed a synergetic effect and vice versa in the negative sign of \(X_1 X_2 (X_1 - X_2)\). The magnitude of coefficient indicates that the term of \(X_1 X_2 (X_1 - X_2)\) has a more favorable effect on response \(Y_2\) than \(X_2 X_2\). It may be implied that the interaction between HPMC and PC molecules caused % elongation of blend films to reduce. The lowest % elongation of blend films was exhibited at the minimal concentration of PC, at HPMC90/PC10. However, the highest % elongation happened at HPMC95/PC5. Therefore, the increase in the appropriate amount of PC increased the % elongation of blend films. This result supported the suggestion that the increase in % elongation of blend films compared with pure HPMC film is probably due to the intermolecular interactions between HPMC and PC.\(^{29,30}\) Therefore, the blend film at HPMC95/PC5 was found to be the most suitable by considering puncture strength and % elongation that determines the mechanical properties of the blend films. From Figs. 2C and D, the suitable model was analyzed by comparing run number versus internally studentized residuals and showing that the data points fall within the limits.

Mucoadhesive Properties; Work of Adhesion (\(Y_3\)) and Maximum Detachment Force (\(Y_6\))\(^{11}\) Mucoadhesion is a specific term explaining the interaction between mucus membrane and surface of the dosage form or substrate.\(^{9}\) In this study, \(F_{\text{max}}\) and \(W_{\text{ad}}\) were used as parameters for the investigation of mucoadhesive properties.

The model selection for analyzing the response \(Y_3\) was dependent on \(R^2\) adjusted \(R^2\), predicted \(R^2\), S.D., and PRESS; however, only the cubic and quartic model had higher values for these parameters than the other models, as seen in Table 2. ANOVA of the data revealed that only the cubic model was significant (\(p<0.05\)) and it was suggested to use this to analyze the response \(Y_3\). The fitted equation associating the response \(Y_3\) is shown in Eq. 17.

\[
Y_3 = 0.034 X_1 + 0.057 X_2 - 0.032 X_1 X_2 + 0.054 X_1 X_2 (X_1 - X_2) (17)
\]

For response \(Y_6\), the statistical parameters (\(R^2\), adjusted \(R^2\) and predicted \(R^2\)) of the quadratic, cubic and quartic models did not have a significant difference, as displayed in Table 2. However, statistical analysis showed that the quadratic model was the most significant (\(p<0.05\)) indicating that this model was suitable for describing response \(Y_6\). The response could be described by the following quadratic equation:

\[
Y_6 = 0.021 X_1 + 0.034 X_2 - 0.023 X_1 X_2 (18)
\]

\(W_{\text{ad}}\) of HPMC/PC blend films ranged from 0.029 to 0.058 N·mm and \(F_{\text{max}}\) ranged from 0.0191 to 0.0347 N, as shown in Figs. 1E and F, respectively. From Eqs. 17 and 18, the cubic and quadratic models were used to analyze mucoadhesive properties in terms of \(W_{\text{ad}}\) and \(F_{\text{max}}\), respectively. The main effects (\(X_1, X_2\)) have a significantly positive impact on both responses and the higher coefficients of factor \(X_2\) than \(X_1\) suggest that the amount of PC in the blend film has a more desirable effect on both dependent variables compared to the amount of HPMC (\(p<0.05\)). Increasing amounts of PC caused an increase in the responses \(Y_3\) and \(Y_6\). Interaction term \((X_1 X_2)\) of each equation shows a significantly negative coefficient (\(p<0.05\)) resulting in a mixture of HPMC and PC for film-forming, which results in a response that is less than expected from linear blending and then \(W_{\text{ad}}\) and \(F_{\text{max}}\) of HPMC/PC blend films reduced.\(^{29}\) The result of \(F_{\text{max}}\) has a characteristic pattern, the same as the effect of \(W_{\text{ad}}\) where an increase of HPMC induces a reduction of \(F_{\text{max}}\); it remains consistent from HPMC95/PC5 to HPMC100. The increase in amount of HPMC from HPMC90/10 to HPMC95/5 caused a reduced level of adhesion of the blend film and it gradually decreased when the amount of HPMC was higher than at HPMC95/5. The combination of HPMC and PC in blend films showed that good bioadhesive properties may be due to a synergistic effect on the bioadhesive strength.\(^{8}\) In addition, an important factor of mucoadhesion is hydrogen bonding polymers and the mucus membrane.\(^{11}\) PC has a large number of carboxyl groups on the molecular structure which are employed for binding with the mucosal surfaces through hydrogen bonding interactions to produce bioadhesive strength.\(^{11}\) Therefore, polymers containing a high density of available hydrogen bonding groups could combine more strongly with mucin.\(^{13}\) It could be concluded that the blend films showed higher mucoadhesion than pure HPMC film due to the hydrogen formation between HPMC and PC, and FTIR was used to confirm that in the next experiment. In Figs. 2E and F, the suitable model was analyzed by comparing run number versus internally studentized residuals and showed that the data points fall within the limits.

Swelling Index at 10 min (\(Y_7\)), 20 min (\(Y_8\)), 30 min (\(Y_7\)) and 60 min (\(Y_{10}\))\(^{8}\) Swelling properties are an important factor for the study of the bioadhesive properties of the films, since swelling of the polymer results in relaxation of the polymer chains and penetration through the mucus membrane during adhesion.\(^{8}\)

For response \(Y_7\), the statistical parameters (\(R^2\), adjusted \(R^2\), predicted \(R^2\), S.D. and PRESS) of all models were not significantly different, as displayed in Table 2. However, statistical analysis of the linear model showed the most significant result (\(p<0.05\)), indicating that this model was suitable for describing response \(Y_7\). The fitted equation associating the response \(Y_7\) is shown in Eq. 19.

\[
Y_7 = 0.43 X_1 + 0.26 X_2 (19)
\]

For response \(Y_{10}\), the linear model was selected for analyzing
the response for the same reason described in the selection of response $Y_7$ that only the statistical data of the linear model were significant ($p<0.05$). The fitted equation associating the response $Y_8$ is shown in Eq. 20.

$$Y_8 = 0.61X_1 + 0.34X_2$$

(20)

The maximum $R^2$ (0.9627), adjusted $R^2$ (0.9491), predicted $R^2$ (0.9282), and low $p$-values ($<0.05$) also suggest choosing the quartic model for response $Y_9$. Additionally, low S.D. (0.026) and the smallest PRESS (0.014) data confirm that the model was appropriate for analyzing response $Y_9$. The response could be described by the following quartic equation:

$$Y_9 = 0.55X_1 + 0.32X_2 + 0.42X_1X_2 + 0.48X_1X_2(X_1 - X_2) - 2.17X_1X_2(X_1 - X_2)^2$$

(21)

The model selection for analyzing the response $Y_{10}$ was dependent on $R^2$ adjusted $R^2$, predicted $R^2$, S.D., and PRESS; however, only the cubic and quartic model had higher val-
ues for these parameters than other models, as displayed in Table 2. Further ANOVA of the data revealed that only the cubic model has a statistically significant ($p<0.05$) effect on response $Y_{10}$ and it was suggested to use this to analyze the response. The fitted equation associating the response $Y_{10}$ is shown in Eq. 22.

$$Y_{10} = 0.50X_1 + 0.31X_2 + 0.097X_1X_2 + 0.81X_1X_2(X_1 - X_2)$$

(22)

The SI of HPMC/PC blend films at 10 to 60 min ranged from 0.225 to 0.649, as shown in Figs. 3B, D, F and H. The linear model is suitable for analyzing SI at 10 and 20 min, as shown in Eqs. 19 and 20, respectively. It could be suggested that only the main effect ($X_1$, $X_2$) has a significant effect on the swelling index at 10 and 20 min ($p<0.05$). The increase in amount of HPMC resulting in the SI was higher as well. The result was quite different from the data of contact angle that showed the highest result at HPMC100, indicating that it had more hydrophobic properties than other blend films. Hence, the SI of at HPMC100 should be lower than other formulations. This result may be explained by the incorporation of PC into blend films, leading to entanglement of the polymer chains. Two major factors with an important role of the initial swelling of films are the surface area of the film and the diffusivity of water into the polymer. Therefore, the entanglement of polymer chains disturbs water diffusion into the blend films causing decreased water content. In the following experiment, using SEM and AFM, it was confirmed that the addition of PC in blend films made the film surface more rugged than the surface of the pure HPMC film. As a result, the SI of HPMC100 showed the highest value. For the SI at 30 and 60 min, the suggested models used for analyzing the swelling property were the quartic and cubic models, as shown in Eqs. 21 and 22, respectively. The two models suggested that not only did $X_1$ and $X_2$ have an influence on the SI at 30 and 60 min, but the nonlinear blending terms were also involved. The main effects ($X_1$, $X_2$) have a significantly positive impact on both responses and the higher coefficients of factor $X_1$ than $X_2$ suggest that the amount of HPMC in the blend film has a more desirable effect on both dependent variables compared to the amount of PC ($p<0.05$). The coefficient on nonlinear blending terms ($X_1X_2$, $X_1X_2(X_1-X_2)$, $X_1X_2(X_1-X_2)^2$) in Eq. 21 has a statistically significant effect on $Y_5$ ($p<0.05$). The higher negative coefficient of $X_1X_2(X_1-X_2)^2$ compared to the coefficient of the other interaction terms with a mixture of HPMC and PC for film-forming results in a response that is less than expected from linear blending and then probably reduces SI of HPMC/PC blend films. Oppositely, in all terms of Eq. 22 showed a positive effect on the response $Y_{10}$, indicating the increase in the amount of HPMC, PC and effect of interaction of them resulting in increasing SI at 60 min, significantly ($p<0.05$). Compared with the same concentration at varying times of SI, it was shown that SI at 20 min was higher than at 10 min. However, the SI at 30 and 60 min showed a trend to be constant or reducing in some formulations of blend films. The results suggest that HPMC/PC blend films have reached an equilibrium state of swelling at 30 min and excessive swelling after this time may lead to erosion due to a loss of blend film weight. In Figs. 3A, C, E and G, the suitable model was analyzed by comparing run number versus internally studentized residuals; it was shown that the data points fall within the limits.

**FTIR Spectroscopy** The FTIR spectra of HPMC powder/film, PC powder and blend films are displayed in Fig. 4. The spectrum of HPMC powder in Fig. 4A shows characteristic peaks at 3460 cm$^{-1}$ (–OH stretching vibrations)$^{1,7}$ at 2931 cm$^{-1}$ (–CH stretching), at 1455 cm$^{-1}$ (–CH$_2$ asymmetric bending vibration) and at 1066 cm$^{-1}$ (–CO stretching vibration). The peak at 3460 cm$^{-1}$ of HPMC powder broadened in HPMC film, as shown in Fig. 4B, indicating that the formation of a hydrogen bond between HPMC and glycerol had occurred.$^{31,32}$ The spectrum of PC in Fig. 4E displays a broad band at 3100 cm$^{-1}$ (–OH stretching vibrations), a weak band at 2939 cm$^{-1}$ (–CH$_3$ asymmetric stretching), and a strong band at 1697 cm$^{-1}$ (–C=O stretching).$^{14,31}$ The weak band at 1415 cm$^{-1}$ is due to the symmetrical stretching of carboxylate anion, while bands 1235 and 1165 cm$^{-1}$ are attributed to the –CO stretching.$^{14,31}$ In FTIR spectra of HPMC/PC blend films, as seen in Figs. 4C and D, two new peaks are shown at 1653 and 1716 cm$^{-1}$ when compared with characteristic peaks of both HPMC and PC. The occurrence of new peaks will appear in changes on the spectra of the polymer mixture since intermolecular interactions may take place.$^{31}$ Therefore, this result supported the interaction of hydrogen bonds between HPMC and PC in the blend film.$^{7}$
PC had an influence on the formation of hydrogen bonding as shown in Fig. 4D, that a peak at 1716 cm$^{-1}$ of HPMC90/PC10 is sharper than the same peak of HPMC95/PC5 in Fig. 4C. It could be assumed that the intermolecular hydrogen bonds were formed between HPMC and PC in the blend film.

**Powder XRD** The XRD patterns of HPMC powder/film, PC powder and blend films are shown in Fig. 5. The HPMC powder shows a characteristic diffraction peak at $2\theta=10.9$ and 20.8°, and the HPMC film shows a peak at 20.8°, as shown in Figs. 5A and B, respectively. The intensity of HPMC film peak at 20.8° was increased, indicating an increase in the crystallinity of film due to the addition of glycerol and suggesting that the hydrogen bonding between HPMC and glycerol was formed. Therefore, this result was associated with the FTIR analysis. In Fig. 5E, the PC powder shows a characteristic diffraction peak at $2\theta=18.8°$. Compared with the peak of HPMC film, the diffraction peak of blend films at $2\theta=10.9$ and 20.8° in Figs. 5C and D became flatter and broader, indicating decreased crystallinity of the blend films. Therefore, the blend films of mixtures of HPMC and PC were amorphous.

**Morphology Examination (SEM)** SEM images were employed to investigate the surface morphology of films, as shown in Fig. 6. The HPMC100 film exhibited smooth and homogeneous surface indicating good properties of film-forming (Fig. 6C). However, the addition of PC in blend films showed a partially smooth surface and wrinkles appeared (Fig. 6B). The increase in amount of PC in HPMC90/PC10 film meant that the film surface was more rugged than the surface of
other films (Fig. 6A). This could be due to differences between their chemical polymer structures.\(^{30}\) It indicated that PC was partially miscible with HPMC, although interaction was proved by the FTIR and XRD analyses.\(^{28}\)

**AFM**

AFM AFM topography was used to further explore the films surface morphology. Figures 7A–C and Figs. 7D–F represent the AFM phase images and three dimensional (3D) images, respectively. The HPMC100 film surface was quite uniform and smooth, as shown in Figs. 7C and F, which correlated with the SEM observations. Increasing PC in the blend films showed a quite rough surface and granular structure embedded in blend films. It was also confirmed by 3D images (Figs. 7D, E). Therefore, the addition of PC into the blend films makes the surface film rough and then AFM topography of PC and HPMC blend film confirmed the SEM results that PC was partially miscible with HPMC. Therefore, the incorporation of PC in blend films resulted in entanglement of the polymer chains, leading to the physical properties of the blend films described in the previous section.

**Conclusion**

In this study, a simplex lattice design was employed to investigate the effect of HPMC and PC on the physical properties of the mucoadhesive blend film. The suitable models chosen for the design were linear, quadratic, cubic and quartic models, depending on several statistical parameters to select the best fitting mathematical model. It was demonstrated that HPMC/PC blend film had higher mucoadhesive properties than pure HPMC film. However, SI at 10 and 20min of pure HPMC film was higher than that of HPMC/PC blend films, possibly due to the entanglement of polymer chains disturbing water diffusion into the blend films. It was proved by SEM and AFM that the addition of PC in blend films made the film surface more rugged than that of the pure HPMC film. The mechanical properties of blend film at HPMC95/PC5 were higher than other proportions of blend film. FTIR and XRD were used to confirm the intermolecular hydrogen bonds between HPMC and PC in the blend film. In summary, HPMC and PC are potentially useful polymers for preparing mucoadhesive blend film as a buccal drug delivery system.

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**Conflict of Interest** The authors declare no conflict of interest.

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