

SAGO STARCH BASED HYDROGEL PREPARED USING ELECTRON BEAM IRRADIATION TECHNIQUE FOR CONTROLLED RELEASE APPLICATION

(Hidrogel berasaskan kanji sagu disediakan menggunakan teknik radiasi alur elektron untuk aplikasi pelepasan terkawal)

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Abstract

Carboxymethyl Sago starch (CMSS) is one of the natural polymers with high value as a polymeric device for medical application due to its potential as biocompatible materials. The monomer blending with modified natural polymers by irradiation technique has received wide attention due to the simple method of preparation, more promising material and free chemical residue in the product. CMSS/methacrylic acid (MAA) hydrogels were synthesized by Electron Beam irradiation at 2meV, 10mA. The objective of this study was to evaluate the ability of hydrogels from modified carboxymethyl sago starch CMSS for oral drug delivery. The hydrogels were characterized by FT-IR, TGA and DTG. The effects of the preparation conditions such as the, monomer composition and irradiation dose on the gel fraction of the synthesized hydrogel were investigated. The swelling properties of the hydrogel were carried out in acidic media, neutral and alkaline media at different temperatures (27°C – 60 °C). The pH sensitive properties of CMSS/MAA hydrogel showed that it can be used as drug delivery devices due to suitability of pH response to the environment in gastrointestinal tract.

Keywords: carboxymethyl sago starch, hydrogels, controlled release

Abstrak

Karboksimetil kanji sagu (CMSS) adalah salah satu polimer semula jadi yang mempunyai nilai tinggi sebagai peranti polimer untuk aplikasi perubatan kerana berpotensi sebagai bahan bioserasi. Adunan monomer dengan polimer semulajadi yang dimodifikasi dengan teknik penyinaran telah menerima perhatian yang meluas kerana kaedah penyediaan yang mudah, bahan yang lebih berkualiti terhasil dan tiada sisa kimia dalam produk tersebut. Hidrogel CMSS/asid metakrilik (MAA) disintesis menggunakan penyinaran alur elektron pada kondisi 2meV, 10mA. Objektif kajian ini adalah untuk menilai kemampuan hidrogel daripada karboksimetil yang telah diubahsuai daripada kanji sagu, untuk penghantaran ubat secara oral. Hidrogel telah dicirikan menggunakan FT-IR, TGA dan DTG. Kesan daripada keadaan persediaan seperti komposisi monomer dan dos sinaran ke atas hidrogel yang disintesis telah diselidik. Sifat pembengkakan hidrogel dalam media berasid, neutral dan alkali pada suhu yang berbeza (27 °C - 60 °C) juga diuji. Sifat sensitif yang ditunjukkan oleh hidrogel terhadap pH telah menjadikannya sesuai untuk digunakan sebagai alat pembawa ubat kerana kesesuaian respon pH kepada persekitaran dalam saluran gastrousus.

Kata kunci: karboksimetil-kanji sagu, hidrogel, pelepasan terkawal

Introduction

Hydrogels can be defined as three dimensional polymer networks that can absorb large amount of water or biological fluids and can retain the fluid without dissolution [1]. It widely use in various applications especially in

controlled release for drug delivery. Controlled release is a system that delivers desirable substances or agents in the controlled rate in term of time, pH or temperature. There are few ways and routes for drug to deliver in human body, but the most complex and most popular route of drug delivery is gastrointestinal (GI) tract due to its huge surface area for systemic absorption and good facility of administration of drugs for compliant therapy [2]. In order for a polymeric material to be use as controlled release devices, it should be inert, biocompatible, bio adhesive, capable of achieving high drug loading at targeting condition and comfortable for the patient [3]. Carboxymethyl sago starch (CMSS) was selected because it is abundant, cheap and a renewable biopolymer. It was synthesized from sago palms (Metroxylon sago) which abundantly grow well in swampy areas mostly in Sarawak State in Malaysia [4]. The uniqueness of starch based hydrogels is, the properties of the hydrogel can be adjusted depending on desired type of devices by altering the structure of the hydrogels.

Addition of Methacrylic acid (MAA) into CMSS via electron beam irradiation is one of the good techniques in order to achieve the desirable pH sensitive properties for the hydrogels. MAA is a good candidate to get the pH sensitive of the hydrogel. This vinyl monomer was extensively used in previous research [5, 6, 7, 8, 9] by using different methods and successfully showed the pH sensitive properties of the hydrogels. The presence of MAA, improved the ability of the hydrogels to swell optimally in intestine pH condition and keep it minimum swell in the acidic condition of the stomach. This is due to the existence of polar functional groups which is responsible for pH-sensitive properties of the polymer and other properties as responsible for bioadhesive [10]. Electron beam irradiation technique was selected because, this method does not required any addition of additives, initiators, cross-linkers or catalysts to initiate the cross linking in order to modify the material. Furthermore, the hydrogels that synthesized using this method are residue-free, pure, sterile product and also protect the inherent of biodegradability and biocompatible properties of the natural polymer [11]. In addition, electron beam irradiation technique will eliminate the present of possible toxicity element due to the formation of radioactive particle because this technique does not involve the radioactive source. Moreover, the gel fraction and swelling behavior of the synthesized hydrogels are easily manage and controlled by varying the dose of irradiation [12, 13]. Because of that, this method is found to be very suitable in producing hydrogels to be applied in pharmaceutical field, which even low level of contamination being avoided [14].

The aim of this paper is to study the ability of modified carboxymethyl sago starch (CMSS) for oral drug delivery by using optimum amount of MAA monomer via electron beam irradiation method. In order to achieve these objectives, the effects of the preparation conditions such as the monomer composition and irradiation dose on the gel fraction as well as the swelling behavior of the synthesized hydrogels were investigated.

Materials and Methods

Materials

Carboxymethyl Sago Starch (D.S 0.75) was prepared from sago starch from Song Ngeng Sago Ind. Sibu Sarawak. The CMSS had been synthesized, purified and characterized as described in [4, 15] with minor modification. The degree of substitution was analyzed as followed in [4]. Methacrylic acid (MAA) 99.5% stabilized was supplied by Acros Organics and pH buffer solution was supplied by Eutech Instrument. Hydrochloric Acid (Fuming 37%) was obtained from Sigma- Aldrich USA. Distilled water and deionized water were used at various stages of the experiment, while (11.0 cm×4.5 cm × 0.30 cm) plastic trays were used as the mold for the preparation of the hydrogels.

Preparation of CMSS/MAA hydrogels

The hydrogels were prepared according to the method suggested by [14,16] with minor modification. The CMSS powder was used after dried in oven at 60 °C. Various amounts of MAA monomer to CMSS (Table 1) were added to a 40% (w/v) dispersion of CMSS in distilled water to make CMSS/MAA mixtures. To ensure homogeneity of the mixture, each mixture was stirred for 15 minutes and was left overnight. Then, the homogenous paste was poured into a plastic molds and inserted into plastic bags. The plastic bags were sealed by vacuum pump to remove air. The samples were then sent for irradiation purpose by electron-beam radiation (EPS-3000, Japan) at the Malaysian Nuclear Agency with beam current of 10mA and acceleration energy of 2 MeV and irradiated at different doses (5, 10, 15, 20, and 25 kGy). While, duration length of irradiation depends on the doses required and control by speed of sample pass under electron beam scanner.

Table 1. Feed composition for the preparation of CMSS/MAA hydrogels

Hydrogel formulation	CMSS (g)	MAA (g)
CMSS/MAA0	40.0	0.0
CMSS/MAA05	40.0	0.5
CMSS/MAA5	40.0	5.0
CMSS/MAA10	40.0	10.0
CMSS/MAA15	40.0	15.0

Gel fractions of hydrogels

The measurement of the non soluble part of the hydrogels, after immersion in water for 48 hours, estimated the gel content of the hydrogels. The irradiated hydrogels were weighed and placed in a tea bag. Then, the tea bag was immersed in distilled water at room temperature. The soluble parts, which were uncross linked of CMSS and MAA, were dissolved and extracted into the water. The successful cross linked hydrogels remained inside the tea bag. The samples were removed from water and oven dried at 60 °C. The percentage of gel content of the hydrogel was calculated by Equation 1 shown below:

$$\text{Percentage of gel content} = (W_b / W_a) \times 100 \quad (1)$$

where W_a is the initial weight of CMS after irradiation, and W_b is the weight of the insoluble part after extraction with water.

Swelling studies at different pH and temperature

To measure the degree of swelling (w/w), approximately 1.0 g of hydrogels was put into a tea bag and was immersed in 100 mL of distilled water. The degree of swelling for the hydrogels was calculated by weighing the swollen sample after 48 hours of immersion. Before it was weighed, the sample was extracted out from the distilled water, the hydrogels was filtered by using stainless steel mesh and the excess water was removed by blotting with filter paper. The degree of swelling was calculated using Equation 2 below:

$$\text{Degree of swelling} = (W_s - W_i) / W_i \quad (2)$$

where, W_s is the weight of swollen sample, and W_i is the weight of dried hydrogel.

The influences of pH were tested by using buffer solution at different pH levels, pH (2, 4, 7, and 10). Meanwhile, the effects of temperature on swelling behavior were tested by using distilled water (pH 7). The temperatures of the solution were controlled at 10 °C, 27 °C, 40 °C, and 60 °C.

Fourier Transform Infrared analysis

The structure of sago, CMSS, and the hydrogels were identified by FTIR analysis using a Spectrum BX FTIR (Perkin Elmer England). Sago starch and CMSS were analyzed in powder form, while the hydrogels were analyzed in the form of thin film. The hydrogels were cut into small pieces and scanned within a range of 4000–500 cm^{-1} using diamond attenuated total reflection (ATR).

Thermogravimetric analysis

Thermal stability of the hydrogels was evaluated using a Metler Toledo machine. The TGA and the differential thermogravimetric (DTG) of the sago, CMSS, MAA, and the hydrogels were interpreted and derived using STAR SW 10.00 software program. Approximately, 10 mg of samples were placed in the sample pan for analysis. The analysis was done over temperature range of 30 °C to 600 °C at 10 °C min^{-1} heating rate under nitrogen gas.

Results and Discussion

Gel fraction of the hydrogels

Figure 1 shows the gel fraction of the CMSS/MAA hydrogels. The result showed that the gel fraction increased with increase amount of MAA until optimum amount, which is 5.0 g. However with further addition of the MAA, the gel fraction started to decrease. Similar trend also can be observed among hydrogels for the same composition but exposed to different dose of irradiation. It seems that, the gel fraction of hydrogels is highly dependent on the electron beam irradiation dose and amount of MAA used [13, 14]. Higher dose of irradiation yielded higher gel fraction. This is because, at higher dose of irradiation, the samples were exposed to the radiation source at a longer time. As a result, it prolonged the propagation step of the polymerization process, which contributed to higher degree of conversion and crosslink of the hydrogels [17]. In addition, the decrements of the gel fraction occur with further increase of MAA. This is because, with excess amount of MAA, non grafted monomer tends to homopolymerize with each other, and then, dissolves into water. At the same time, exposure of electron induced, leads to the degradation process in the polymer network as a result from the chain scission of the hydrogel network [18]. This suggests that dose of irradiation and amount of MAA added play a big role that affected the gel fraction of the hydrogels.

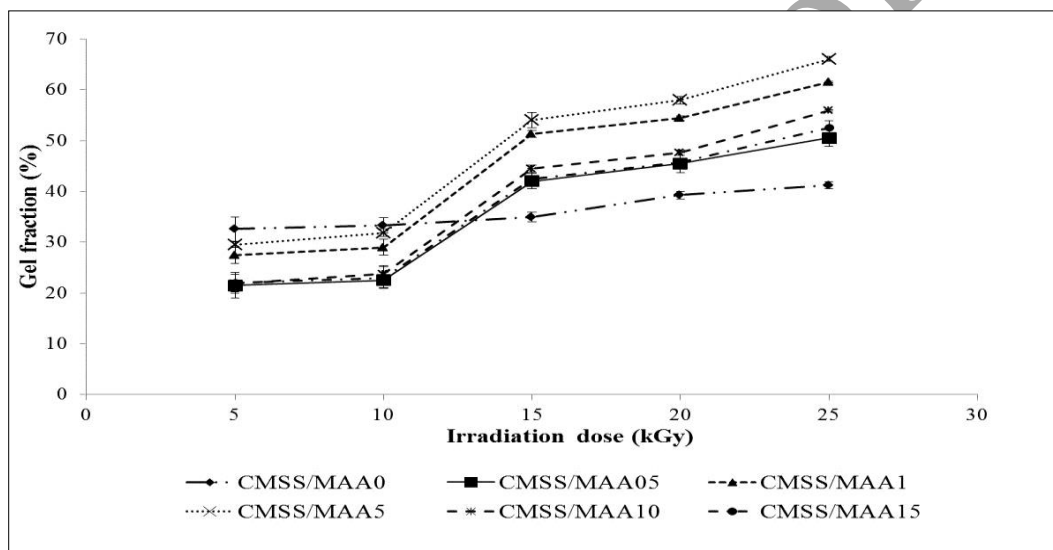


Figure 1. Gel Fraction of CMSS/MAA hydrogels at different dose and different composition.

FT-IR analysis

FT-IR analysis is an effective technique to study the structure of the polymer. The FT-IR spectra of sago, CMSS and the hydrogels were shown in Figure 2. The spectrum of sago starch shows the absorption band at 3310 cm^{-1} , which is due to O-H stretching vibration, as well as intramolecular and intermolecular hydrogen bonds in the glycosidic bond in the sago starch molecule. The appearance of absorption band at 2929 cm^{-1} is due to C-H stretching. Absorption peak at 1637 cm^{-1} most probably is assigned for water molecule, which tightly binds at the sago starch molecule [15]. The absorption band at 1343 cm^{-1} belongs to CH_2 symmetrical band, while the broad band at the range of 999 cm^{-1} refers to the stretching of C-O in C-O-C and C-O-H in the glycosidic ring of starch molecule [15]. The carboxymethylation of sago starch gave some additional new peaks to the FTIR spectrum. New absorption peak is observed at 1590 cm^{-1} . This proves the substitution of carboxyl methyl ether ($-\text{COO}-\text{Na}^+$) group on the sago starch chains. Then, the absorption bands around 1412 cm^{-1} and 1323 cm^{-1} are respectively due to $-\text{CH}_2$ scissoring and $-\text{OH}$ bending vibration, while $\text{OCH}-\text{O}-\text{CH}_2$ stretching is represented by the absorption band at 1007 cm^{-1} . The blank CMSS hydrogel have nearly same FTIR spectrum as CMSS powder but varying from each other by

means the intensity of the absorption band. Meanwhile, CMSS/MAA hydrogel exhibits new peak at 1698 cm^{-1} . The new peak with low intensity at 1698 cm^{-1} corresponds to the C=O in carboxylic groups of MAA [13], nearly similar as the MAA monomer at 1690 cm^{-1} . This appearance of new C=O peak indicates that MAA took part in the CMSS/MAA hydrogels formation.

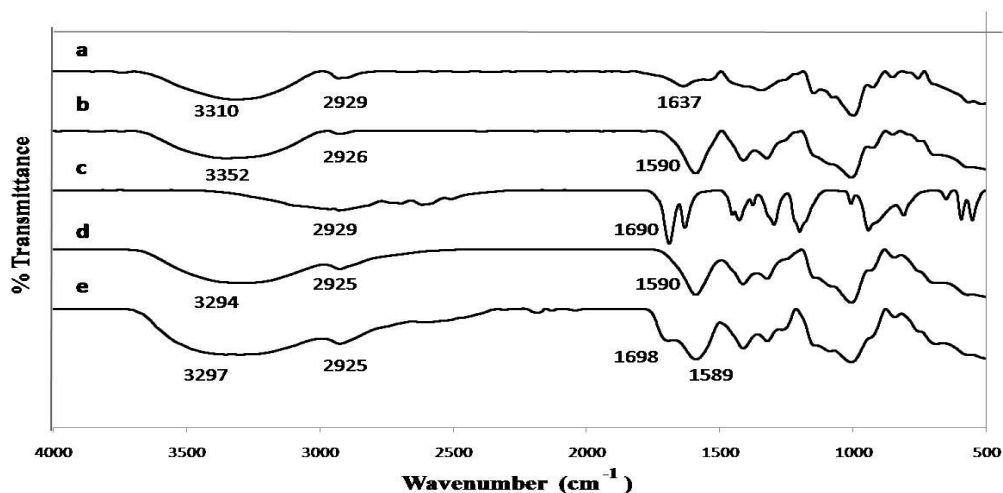


Figure 2. IR spectra for a) Sago starch, b) CMSS, c) MAA monomer, d) CMSS hydrogel e) CMSS/MAA5 hydrogel irradiated at 25 KGy.

Thermogravimetric analysis

Thermogravimetric analysis is a convenient analysis to study the thermal stability and thermal decomposition of polymer. The thermal properties and the derivatives of CMSS, MAA and CMSS/MAA hydrogel which irradiated at 25 KGy dose of irradiation are shown in Figure 3. The thermogram showed the percentage weight loss at decomposition temperature for the sago starch, CMSS, MAA, CMSS hydrogel and CMSS/MAA hydrogel. The TGA and DTG from the figures show that the thermal decomposition for CMSS DS (0.75) occurred with the maximum temperature of $285.1\text{ }^{\circ}\text{C}$, which lower than sago starch before modification, which was $298.8\text{ }^{\circ}\text{C}$. It shows the miscibility between sago starch and other materials in the reaction to produce CMSS powder. As for MAA, it underwent a single step of decomposition with maximum decomposition temperature occurred at $140.4\text{ }^{\circ}\text{C}$. Meanwhile, from the figures, it is obvious that CMSS/MAA hydrogels underwent three steps of decomposition. The first step belongs to the decomposition of free water and the remaining free MAA molecule, which is grafted to the CMSS. Then, the second step belongs to CMSS decomposition peak, and the last one comes from the full breakage of the poly(methacrylic acid) similarly reported by [19] in the range of temperature from $346.8\text{ }^{\circ}\text{C}$ to $546.4\text{ }^{\circ}\text{C}$. In addition, the decomposition peak of CMSS/MAA is observed shifted to higher temperature, which is $301.9\text{ }^{\circ}\text{C}$ compared to $285.1\text{ }^{\circ}\text{C}$ for CMSS hydrogels without the addition of MAA. This is due to the high cross linked structure and compact networks of hydrogels, which are influenced by the higher quantity of MAA. This produced higher degree of stability to the hydrogels, in which higher temperature is needed to decompose them [14].

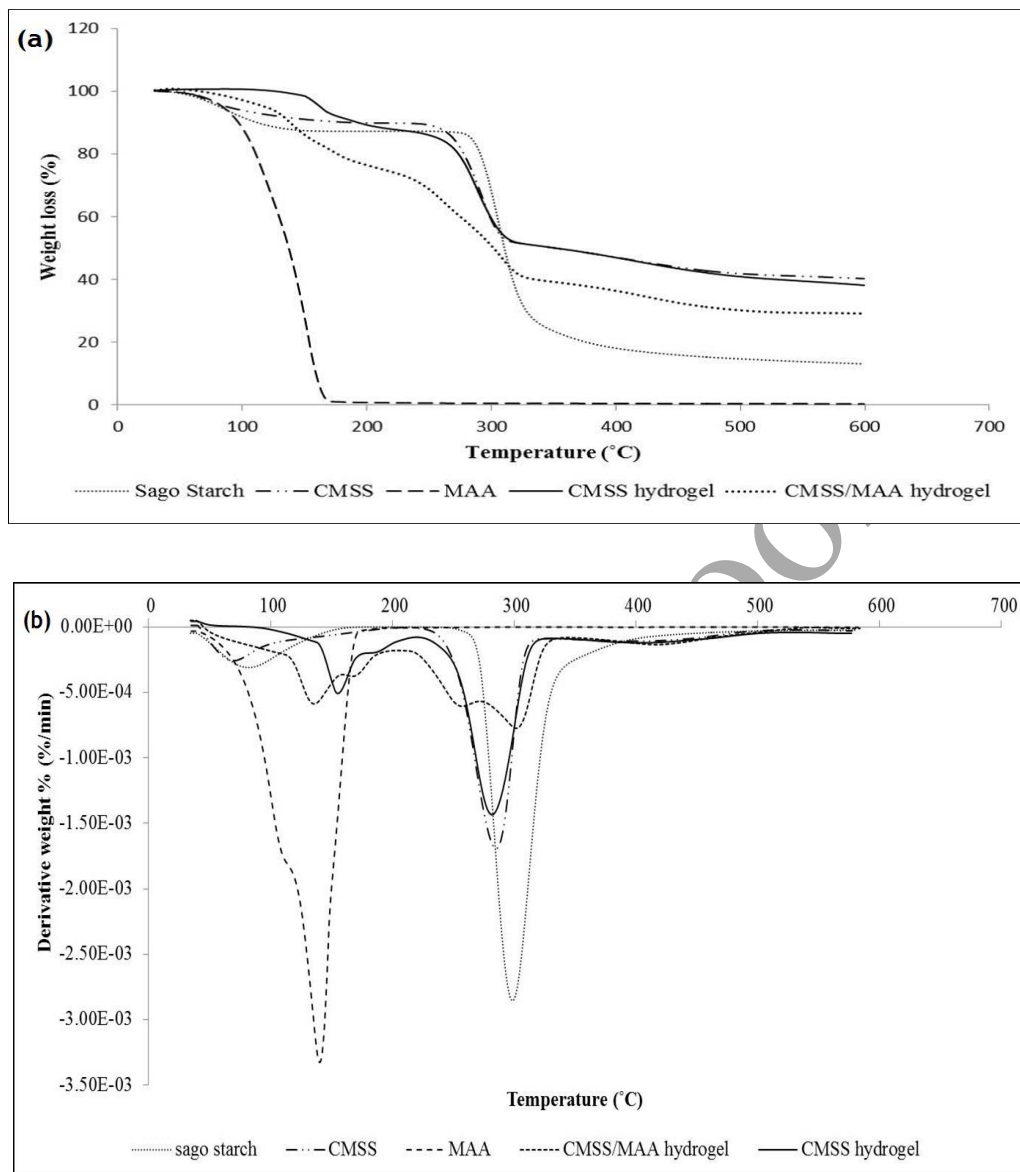


Figure 3. TGA (a) and DTG (b) of sago starch, CMSS, MAA, and hydrogels irradiated at 25 Kgy

Swelling behaviour

The study in swelling would suggest a good prediction on the ability of the hydrogels to be used as drug delivery devices in controlled release application. The swelling behavior of CMSS/MAA5 hydrogels in different media is shown in Figure 4. From the figure, all hydrogels, even though irradiated at different doses (5 - 25 kGy), showed the same pattern of swelling as a response to the pH. The hydrogels exhibited low degree of swelling within acidic medium (pH 2-4) and swelled slightly high with increased pH until reaching the neutral pH (pH 7). Then the gels showed a decreasing degree of swelling in the alkaline medium (pH 10). The swelling behavior is influenced by hydrophilicity of the carboxylic group in the structure of the hydrogels. In an acidic medium (pH 2), most -COOH groups are kept in the form of COOH , which induce the formation of hydrogen bond of -COOH groups with CMSS

segment. This interaction between the polymer chains might be attributed to form more hydrogen bonds, leading to a more compact network in hydrogels, which causes a decrease in the swelling degree in the hydrogels [7]. Meanwhile, with increasing pH (4-7), the MAA is deprotonated to a negatively charged carboxylate ion [20, 21], and caused the breaking of hydrogen bonds between the chains of the polymers. Moreover, with the presence of electrostatic repulsion, leading to the polymeric network expansion, attracts more water into the network of hydrogels. It directly causes the degree of swelling to become higher [7, 22]. However, in alkaline medium (pH 10), the reduction of swelling degree was observed. The hydrogels have more chance to interact with basic species in this medium. This restricted the interaction of acid molecules with water molecules. The restricted interaction between acid molecules and water, and also the non ionized condition of the carboxylic groups, contributed to the reduction of the swelling ratio [23, 24].

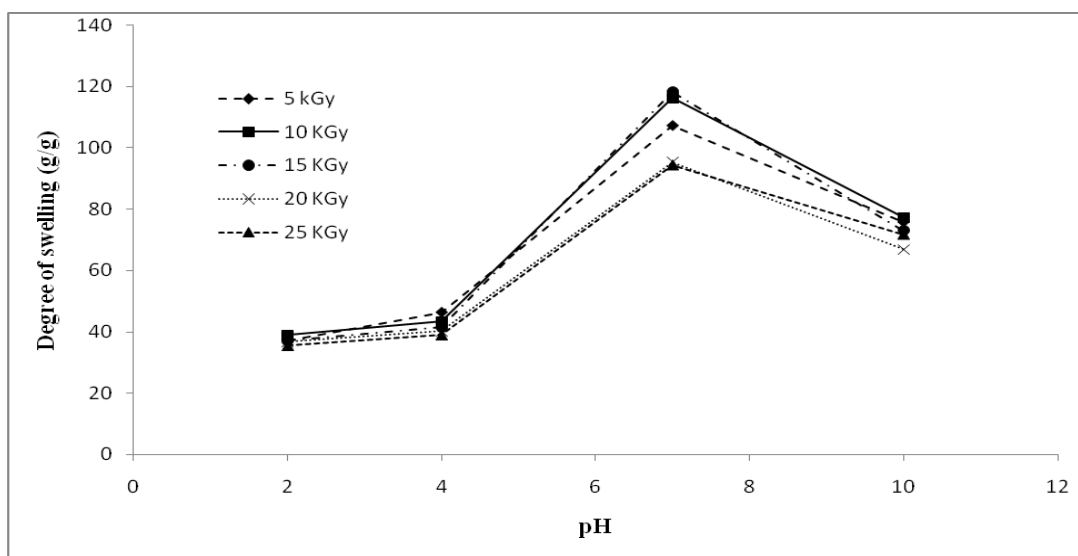


Figure 4. The effects of various pH levels on swelling behavior of CMSS/MAA5 hydrogels, irradiated at 5-25 kGy

The swelling behavior of the hydrogels at various temperatures is shown in Figure 5. The swelling study showed that the highest degree of swelling exhibited by hydrogels irradiated at 15 kGy and the lowest was at 25 kGy. It can be observed that all hydrogels gave optimum swelling at 27 °C, and then decreased with further increase of temperature. This is because, above from the optimum temperature, strong aggregation forces between the network of hydrogels occur, which lead to increase void volume, resulting in rapid release of water [18]. While at high temperature, the amount of bound fluid in hydrogels is increased [25]. Furthermore, when the temperature keeps increasing, the force of hydrogen bonding is reduced and causes the bond fluid to become non binding fluid, and this free fluid can move out from the polymeric network with the favor of the higher kinetic energy of the solution [26]. Furthermore, from the results obtained, higher doses contributed to the lower swelling degree, and this is because, higher dose of irradiation leads to denser network. This rigid and denser structure restricts the mobility within the network and causes lower swelling for higher irradiated doses of hydrogels [12].

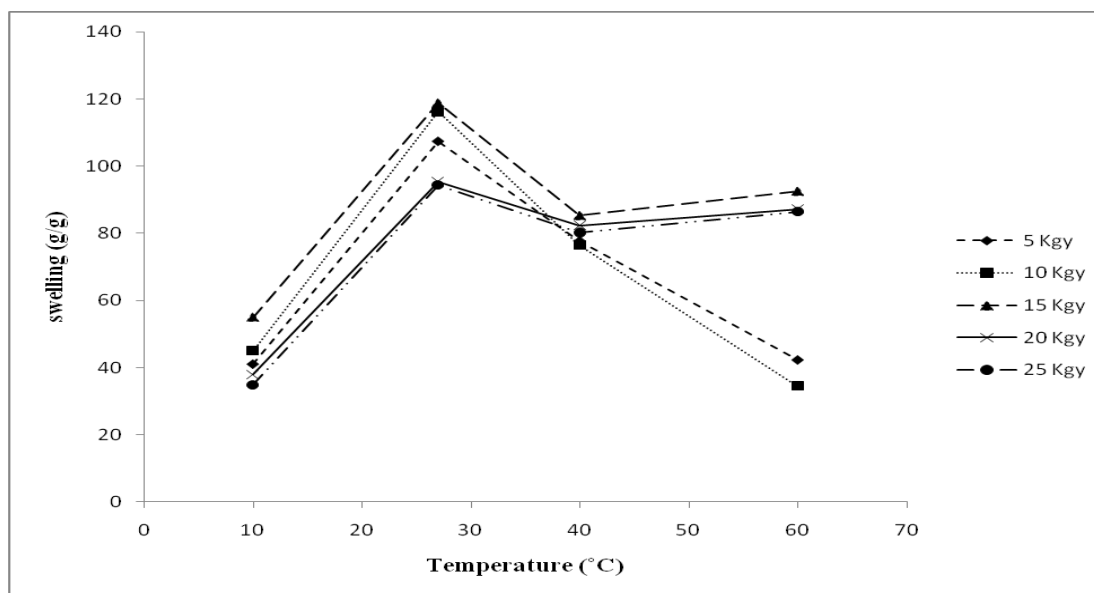


Figure 5. The effects of irradiation dose on the swelling behavior in distilled water (pH=7) for hydrogels composition CMSS/MAA5 at different temperatures.

Conclusion

This study showed the potential of hydrogels from CMSS/MAA for controlled release in drug-delivery application using electron beam irradiation technique. The gel fraction of the hydrogels depends on the CMSS amount, amount of MAA added and also the irradiation dose. The optimum amount of MAA added was 5.0 g at irradiation dose of 25 kGy due to the maximum value of gel fraction at this condition. The result of FTIR revealed that MAA successfully grafted onto the CMSS. Furthermore, TGA and DTG results showed that the addition of MAA into hydrogels, improved the thermal stability of the hydrogel. The results of swelling behavior studies suggested that the CMSS/MAA hydrogels are pH responsive. The CMSS/MAA hydrogel exhibited low swelling at acidic pH (pH 2) and the swelling increased with increasing pH until maximum swelling at neutral pH (pH 7). The pH and temperature responsive behavior of the CMSS/MAA hydrogel are suitable for retaining the minimum swelling of the drug carrier devices at gastric media and swell optimally at intestine area. Findings from this study suggested that CMSS/MAA hydrogels could be used as a controlled release device for drug delivery.

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