THE DEVELOPMENT OF A TECHNOLOGY PLATFORM FOR PRODUCING NOVEL COSMETIC HYDROGELS CONTAINING ACTIVE AGENTS

MAYTINEE YOOYOD

A Thesis Submitted to the Graduate School of Naresuan University
in Partial Fulfillment of the Requirements
for the Doctor of Philosophy in Chemistry
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By Maytinee Yooyod

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Title	THE DEVELOPMENT OF A TECHNOLOGY PLATFORM FOR PRODUCING NOVEL COSMETIC HYDROGELS CONTAINING ACTIVE AGENTS
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ABSTRACT

The cosmetic industry is a market that is constantly increasing in size and scope with the products offered. This thesis focuses on the development of a technology platform to produce novel cosmetic hydrogels that incorporate active ingredients. The hydrogels were produced from poly(N-vinylformamide) (PNVF) and NVF-copolymers through photopolymerization to create cosmetic hydrogel patches. In order to fully investigate these hydrogels the work was separated into two approaches. The first approach was to investigate a series of PNVF hydrogels and various copolymers, including N-hydroxyethyl acrylamide (HEA) and 2-carboxyethyl acrylate (CEA). For a guide in the selection of the most suitable properties for cosmetic hydrogels, a series of commercial cosmetic hydrogel masks were first studied. The PNVF-based materials were evaluated and then compared with the commercial products. The method used to produce all hydrogels in this work was photopolymerization using a UV-LEDs light source, which has a high throughput and short processing time. This is the major advantage of this method with the ability to simply and effective scale the fabrication of the hydrogels from batch to continuous production on an industrial scale. The study evaluated the impact of varying the ratio of ingredients in the formula for producing the hydrogels and the effect this had on the base gel properties such as equilibrium water content (EWC), state of water, wettability, and mechanical properties. These were all observed in the hydrogels to determine the optimal composition for the novel cosmetic hydrogel patches. The

applied properties, such as skin adhesion and skin irritation, were also studied, along with self-perception questionnaires to determine the suitability of the cosmetic hydrogel patches. The results for the homopolymer of PNVF gave a hydrogel with an EWC of ~95%. When decreasing NVF content in copolymer hydrogels the water content also decreased. As these gels are designed for incorporation and release of active agents a series of controlled drug release studies were undertaken. Three different dyes were investigated: Orange II Sodium salt, Crystal Violet and Congo Red. The dyes were incorporated to the hydrogels and the amount of each dye released from the hydrogel was measured. Water structure played a key role in the amount of dye released from the hydrogels, with the amount released depending on two parameters: first, the ratio of freezing water (free) and non-freezing water (bound), and second, the hydrophilicity and structure of the dye. This series of hydrogels can therefore control the amount of dye released by controlling the structure and amount of water contained in the hydrogel through the monomer ratio. In terms of *in-vivo* clinical applications with adhesion ability and skin irritation after application, the results were successful in terms of adhesion, with no slippage of the hydrogels. After a single application of the cosmetic hydrogel patch on the skin, the skin erythema value showed no difference. Additionally, no signs of skin irritation were observed during this study, indicating that the developed hydrogels for novel cosmetic hydrogel patch applications are mild on the skin.

The second approach was to fabricate the pH responsive hydrogels and their potential ability as delivery carriers for cosmetically active ingredients. This was achieved by producing hydrogels that contain poly(vinylamine) (PVAm). We first prepared a poly(N-vinyl formamide)-co-poly(N-hydroxyethyl acrylamide) (poly(NVF-co-HEA)) hydrogel via photopolymerization (UV-LEDs) using N,N-methylenebisacrylamide (MBAAm) as the crosslinker. This hydrogel was then hydrolyzed to convert the vinylformamide groups in the polymer to vinyl amine, the hydrolysis was carried out under acidic conditions, at 80 °C using 0.01 M aqueous HCl. These pH sensitive hydrogels were characterized by EWC, DSC and swelling behavior in both water and ethanol. The hydrogel swelling at different pHs was observed, non-hydrolyzed hydrogels (poly(NVF-co-HEA)) swelling was consistent

across the entire pH range but after hydrolysis the hydrogels show different behavior with reduced swelling ratios observed at acidic and basic conditions (pH2 and pH 10 and 12). In our design of a controlled release system, Lactobionic acid was used as a model ingredient. All the results show that the functionalized hydrogel was successfully prepared to form the poly(NVF-co-VAm-co-HEA) hydrogels, which have the potential for use as a pH-responsive carrier for cosmetic ingredients triggered by an external pH change. The results of pH-responsive release provide an understanding of the active delivery properties and help identify the best suitable cosmetic active ingredients. PNVF-based hydrogels were synthesized over a wide range of compositions included different copolymers. This gave a wide range of useful properties for novel cosmetic hydrogel patches.

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Maytinee Yooyod

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CHAPTER I

INTRODUCTION

This research is funded by the RRi scholarship and linked with the company Amity laboratory Co., Ltd. This is a relative new SME company in the cosmetic industry. The cosmetic industry is an ever rapidly changing environment where companies must continuously adapt to the latest trends. Therefore, in order to help the company produce new products this research aims to produce a technology platform for the production and fabrication of synthetic hydrogels. The hydrogels should be able to be quickly and simply adapted to incorporate different active agents for slow release to the target area. In this way the production method and procedure remain the same for the different active agents. Hydrogel sheets can be cut into multiple shapes enabling a wide range of locations to be used, for example, full face masks, eye and lip patches (tear drop shape).

The important aspects of this research work was in the study of the production of novel cosmetic hydrogels, which were designed with a novel monomer system along with a photopolymerization process that can be easily scaled up. The research was separated into two approaches. The first approach was the fabrication of poly(N-vinylformamide; PNVF)-based materials through photopolymerization to create cosmetic hydrogel patches. The second approach is the synthesis of hydrogels containing cationic poly(vinylamine) by partially hydrolyzing the amide group under acidic conditions. This chapter gives an introduction about the research work of the PNVF based novel cosmetic hydrogel patches and the PNVF-copolymer hydrogels containing the cationic poly(vinylamine), including objectives and scope of the research.

Approach I: Poly(N-vinylformamide) based novel cosmetic hydrogel patches

The cosmetic industry is a market that is constantly increasing in dimension and products offered. The skin is the largest organ of the human body, which accounts

for about 16% of its total weight [1] and plays a major role in maintaining homeostasis and environmental protection. The skin regulates the body temperature and the body fluid content, and represents the principal barrier to the external environment, for example, it protects against pathogenic agents and UV radiation [2]. Skin appearance and texture are highly influenced by the water content of the epidermis and dermis, defined as skin hydration. The water in the skin can be present in the form of free or bound water molecules and in the stratum corneum, free water is able to diffuse from the skin to the outer environment, while bound water is associated with other molecules, called “Natural Moisturizing Factors” [3].

Decreased hydration of the skin leads to a dry and scaly skin surface and it may cause skin irritation and inflammation, and different dermatological conditions [4-6]. Reduced moisture will also lead to loss of skin radiance and skin elasticity. The reduction in elasticity decreases skin’s ability to retain its shape and this causes the formation of fine lines and wrinkles. Skin hydration can be affected by endogenous factors, such as genetic components, hormonal changes and intrinsic ageing [7-8]. Skin hydration can also be influenced by the diet and lifestyle factors, i.e., poor diet, alcohol, caffeine and cigarette consumption, each of reducing skin moisturization [9-10].

Lastly, environmental factors, such as air dryness and air conditioning, wind, cold weather and ultraviolet (UV) radiation, can also disrupt the water balance in the skin [11-14]and therefore cause dry skin. For example, exposure to UV radiation can lead to skin photo-damage and, as a consequence, to skin ageing as well as to skin cancer [15-16]. In particular, UV light is an external oxidative stressor on the skin and reactive oxygen species (ROS) are generated after UV exposure [17].

Facial skin is the most UV exposed area and it is susceptible to premature skin ageing. Recent studies suggest that sunscreen application provides only limited protection against the long-term effects of UV exposure [18]. Other strategies should be considered to attenuate ROS signaling and inflammation pathways. For example, topical application of cosmetic products containing botanical extract with antioxidant and anti-inflammatory as well as hydrating properties have proved to be effective against photo-damage and photo-ageing [19-23].

Hydrogels are three-dimensional, hydrophilic, polymeric networks capable of absorbing large amounts of water or biological fluids. Hydrogels are often chosen as facial mask ingredients because of their high-water content and their hydrophilic base which creates a matrix to the skin, thus allows high efficacy in delivering active ingredients into the skin [24]. Hydrogel masks not only have a superior moisturizing and regenerative effect on the skin but are also able to control skin temperature and enhance physiological activity [25]. Furthermore, hydrogel masks have good elasticity and are convenient to apply. Hydrogels can be either synthetic or natural in source, many synthetic hydrogels are based on hydrophilic polymers that contain hydroxyl groups. The first synthetic hydrogel was made from 2-hydroxyethyl methacrylate (HEMA) in the 1960's by Otto Wichterle [26], these materials has seen great-success and it is a testament to his pioneering work that it is still in use today in applications such as contact lenses.

In terms of materials, the hydrogels themselves will be based on a novel biocompatible system comprised of poly(N-vinylformamide) and other copolymers. N-vinylformamide (NVF) is an isomer of acrylamide (AAm) but possesses some favorable properties over AAm such as; a lower toxicity [27], is more hydrophilic, more reactive [27-28] and is liquid at room temperature [29] so has easier usage. The physical properties of poly(N-vinylformamide) (PNVF) gels are quite similar to the technologically important hydrogel of polyacrylamide (PAAm). Polyacrylamide (PAAm) hydrogels are used in electrophoresis [30], chromatography [31], cosmetics [30-31], biomedical implants [32], superabsorbent products [33] and soil conditioners [34] among numerous other applications. PNVF gels are also chemically related to poly(N-vinyl pyrrolidone) gels, another important biomedical hydrogel, widely used in contact lenses, [35] drug delivery systems, and wound dressings.

All the monomers selected in this project will contain the vinyl group and undergo free radical addition polymerization, there are many initiation methods for free radical polymerization, but this project will focus solely on photo-initiation. Photo-initiation of hydrogels has been widely used, particularly with the Irgacure series of photo-initiators, these increase the gelation rate and provide spatial and

temporal control of gelation [36]. During the past 20 years, the field of photo-polymerization has become of central importance in polymer science and technology.

In the case of synthetic hydrogels, polymerization processes are one of the most widely used chemical processes in various fields of industry [37-38]. One of the most modern and rapidly developing methods of obtaining polymers is light-induced polymerization, i.e., photopolymerization [39-40]. The use of photopolymerization processes requires an appropriate initiating system that, in biomedical applications, must meet additional criteria such as high-water solubility, non-toxicity to cells, and compatibility with visible low-power light sources [41]. Photo-initiation by UV light is recognized as a very useful synthetic tool. Photo-initiation has various advantages over other initiation methods such as being easier to control, more ease to convert into a continuous process for scaled up production, also it offers the possibility of combining both polymerization and sterilization into one technological step [42]. Photo-initiators are molecules that can be split into two or more parts by exposure to light. At least one of these parts is capable of reacting with both the monomers and binders to link them together. Photo-initiators are only sensitive to specific wavelengths of light. The spectrum of the light source used must overlap that for which the initiator is sensitive. Both lasers and other types of light sources such as arc lamps may be used. Photo-initiators comprise normally of just a few percent of the overall mixture.

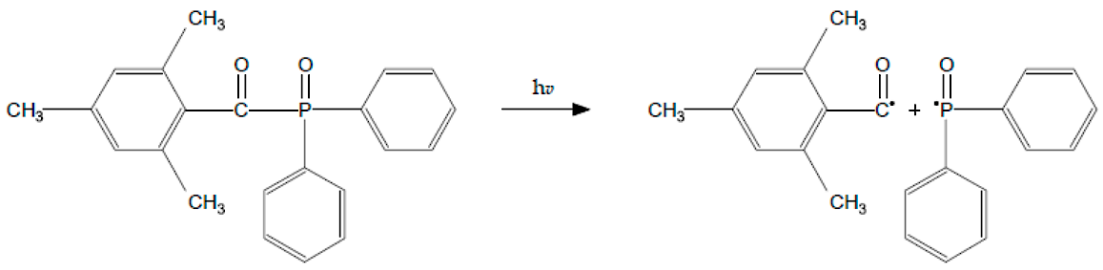


Figure 1 Schematic structure of the production of Photo-initiator by UV-LEDs radiation

For this project, the one photo-initiator is Diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide (TPO), which breaks down into free radicals via UV-LEDs light as shown in figure 1. The free radicals then attack the vinyl group of the monomers during the initial step; the macro polymer radicals then continue to propagate to form linear copolymers, during this stage the cross-linking also occurs to form 3D networks.

So, this work will focus on continuing our unique knowledge of novel hydrogel systems with the specific goal of enhancing the retention and release of active agents. The method used to produce the gels will be photo-polymerization using UV light sources, which has high throughput and processing time. This is the major advantage of this method with the ability to easily and effective scale the fabrication of the hydrogels from batch to continuous production on an industrial scale easily achieved.

Approach II: Synthesis of NVF-copolymer hydrogels containing the cationic poly(vinylamine)

Some polymers in aqueous solution can undergo a reversible phase transition upon external stimuli or modest changes of environmental conditions like temperature, pH, redox condition, or ionic concentration, etc [43]. For instance, the copolymers of poly(phenylene oxide)-poly(ethylene oxide) (PPO-PEO), poly(lactic-co-glycolic acid)-poly(ethylene oxide) (PLGA-PEO) and polycaprolactone-poly(ethylene oxide) (PCL-PEO) exited a sol-gel or fluid-gel transition upon a temperature stimulus [44-46].

Polyvinylamine (PVAm) is a cationic polyelectrolyte that has the highest density of primary amine functional groups of any polymer. PVAm adsorbs spontaneously on most surfaces in aqueous solution, generating cationic interfaces. PVAm, therefore, provides a great potent tool for the modification of macroscopic and nanoparticle surfaces [47]. Polyvinylamine (PVAm) is one of the simplest and most important polymers due to significant modification possibilities [48]. PVAm, a linear polymer with all primary amine groups that are all bonded directly to the main

chain, is of interest because of its pH-dependent polycationic nature. However, vinylamine monomer, as the simplest precursor to PVAm, is not available in the free state, because it tautomerizes so readily to acetaldehyde imine. Therefore, PVAm has been synthesized only via indirect routes. It is most convenient to produce PVAm from poly(N-vinylamide)s by hydrolysis [50-51] groups. It normally showed great pH-responsiveness: A minimum swelling ratio was normally observed at the isoelectric point (pI, the pH of zero net charge), while a much larger swelling ratio and high chain extension were shown at extreme pH values that far away the isoelectric point [51].

Here we report on the results of a comparison between the effects of PNVF-co-HEA hydrogels containing cationic poly(vinylamine) (PVAm) after undergoing acid hydrolysis and those of PNVF-co-HEA hydrogels. The expected hydrolysis product from poly(NVF) and its copolymer is PVAm. We discuss the pH responsiveness of the hydrolyzed hydrogels, covering their equilibrium water content (EWC), state of water, wettability, swelling behavior as a function of pH, and mechanical properties. Another aspect of this work if how the material will release active agents, therefore, this gel was also assessed for the uptake and release of several species across multiple pHs. The results of pH-responsive release provide an understanding of the active delivery properties and help identify the best conductive cosmetic active ingredients.

Objectives of the research

The objective of this research was separated into two approaches depending on the application of the hydrogels (novel cosmetic hydrogel patches and pH-responsive hydrogel)

Approach I

To fabricate, develop and characterize the poly(NVF)-copolymer hydrogel series, with the goal for eventual up-scaling and optimizing of the photo-polymerizable system to be a synthetic hydrogel sheet technology platform, for potential use for novel hydrogel patches in cosmetic application.

Approach II

The preparation of pH-responsive hydrogels and their potential use as delivery carriers for cosmetic active ingredients, via the synthesize of poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine) (PVAm) by partially hydrolyzing the amide group of poly(NVF-co-HEA) under acidic conditions. The effect of the resulting poly(NVF-co-VAm-co-HEA) hydrogels (hydrolyzed hydrogels) will be compared to that of the non-hydrolyzed poly(NVF-co-HEA) hydrogels. This is to confirm the presence of the pH responsive PVAm.

Scope of research

This thesis focuses on the development of a technology platform for the production of novel cosmetic hydrogels that can incorporate active agents. The scope of this research is divided into two approaches. The first approach involves the fabrication of poly(N-vinylformamide; PNVF)-based materials through photopolymerization to create novel cosmetic hydrogel patches. The second approach is the synthesis of poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine) by partially hydrolyzing the amide group of poly(NVF-co-HEA) under acidic conditions.

Approach I: The scope of this approach is summarized in the picture below (Figure 2), which is divided into three parts and these related to development of a novel hydrogel system based on N-vinylformamide and its copolymers for use as cosmetic hydrogel patches. Initially commercial cosmetic hydrogel patches were

characterized that were classified based on gel matrix and what effect this had on the base gel properties. This information was used to guide the selection of the most suitable properties for cosmetic hydrogels. For own PNVF-based hydrogels were then studied to evaluate the impact of varying the ratio of ingredients in the formula for producing hydrogels of PNVF and PNVF with its selected copolymers. The base gel properties such as equilibrium water content (EWC), state of water, wettability, and mechanical properties of the hydrogels were observed to determine the optimal composition for the novel cosmetic hydrogel patches. The application properties, such as skin adhesion and skin irritation, were also studied, along with self-perception questionnaires to determine the suitability of the cosmetic hydrogel patches.

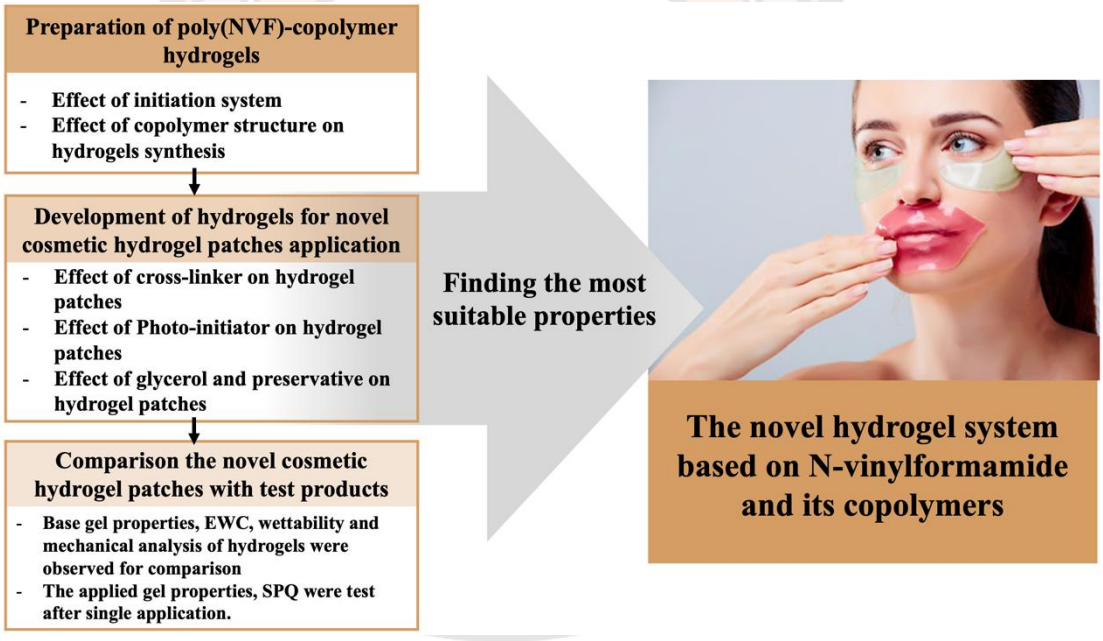


Figure 2 Schematic illustration of scope of research for approach I

Approach II: The preparation of pH-responsive hydrogels and their potential use as delivery carriers for cosmetic active ingredients, the hydrogels synthesized through UV-LED photopolymerization using a copolymer of poly(NVF) and hydroxyethyl acrylamide (HEA) as monomers. The study compares the effect of poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine), PVAm, after undergoing acid hydrolysis with that of poly(NVF-co-HEA) hydrogels. The analysis

covers equilibrium water content (EWC), state of water, wettability ability, swelling behavior as a function of pH and mechanical properties of the hydrogels. The results of pH-responsive release provide an understanding of the active delivery properties and help identify the best conductive cosmetic active ingredients. Figure 3 shows the scope of this approach.

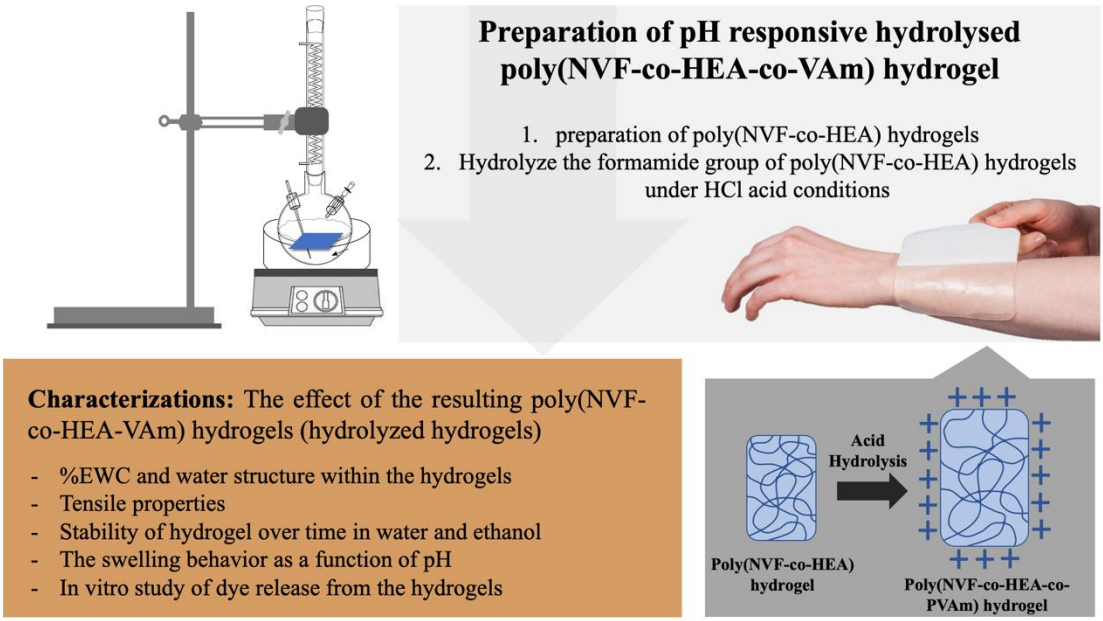


Figure 3 Schematic illustration of scope of research for approach II

CHAPTER II

LITERATURE REVIEW

This chapter contains information on the applications, research methodology, and materials and published works related to this research. This research work is divided into two approaches. The first approach involves the fabrication of poly(N-vinylformamide; PNVF)-based materials through photopolymerization to create novel cosmetic hydrogel patches. The second approach is the synthesis of poly(NVF-co-HEA) hydrogels that contain cationic poly(vinylamine) by partially hydrolyzing the amide group of poly(NVF-co-HEA) under acidic conditions. Therefore, the literature review can be classified into 6 topics as followed:

1. **Information and applications of hydrogels**
2. **Photopolymerization**
3. **N-vinylformamide (NVF) and poly(vinylamine); PVAm**
4. **Cosmetic hydrogels**
5. **pH sensitive hydrogels for delivery of active ingredients**
6. **Water states in hydrogels**

Information and applications of hydrogels

A hydrogel is a cross-linked network of a one or more hydrophilic polymer that is insoluble in water. In the presence of abundant water, a hydrogel absorbs water to swell to a size much larger than its original size. Consequently, they are soft, pliable, wet materials with a wide range of potential applications. Hydrogels are today widely used in bio-applications and play a crucial role in current strategies to remedy malfunctions in and injuries to living systems. The high-water content of hydrogels renders them compatible with most living tissue and their viscoelastic nature minimizes damage to the surrounding tissue when implanted in the host. In addition, their mechanical properties parallel those of soft tissue, making them

particularly appealing with the host tissues, assisting and improving the healing process, and mimicking functional and morphological characteristics of organ tissue.

Figure 4 Hydrogel applications/products

In ‘permanent’ or ‘chemical’ gels, the network of covalent bonds joining different macromolecular chains can be achieved by cross-linking polymers in the dry state or in solution [52]. Chemical hydrogels are commonly prepared in two different ways: ‘three-dimensional polymerization’, in which a hydrophilic monomer is polymerized in the presence of a polyfunctional cross-linking agent, or by direct cross-linking of water-soluble polymers (Figure 5) [53].

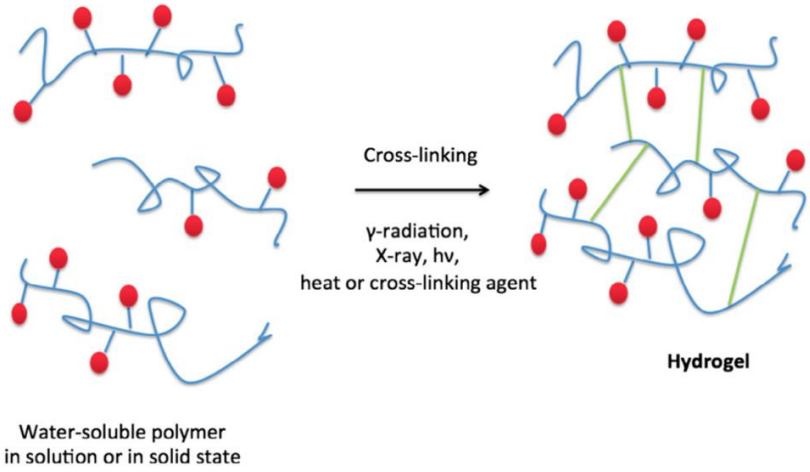


Figure 5 Synthesis of hydrogels by cross-linking of ready-made water-soluble polymers [53]

Hydrogels can be either synthetic or natural in source, many synthetic hydrogels are based on hydrophilic polymers that contain hydroxyl groups. The first synthetic hydrogel was made from 2-hydroxyethyl methacrylate (HEMA) in the 1960's by Otto Wichterle [26], these materials has seen great-success and it is a testament to his pioneering work that it is still in use today in applications such as contact lenses.

With the establishment of the first synthetic hydrogels by Wichterle and Lim [26] in 1960, hydrogels have many applications in numerous fields such as; hygienic products , agriculture [54], drug delivery systems [3-5], sealing [26], coal dewatering [56], artificial snow [26], food additives [57], pharmaceuticals [52], biomedical applications [52], [57] tissue engineering and regenerative medicines [58], [59], diagnostics [60], wound dressing [61], separation of biomolecules or cells [62] and barrier materials to regulate biological adhesions [63], and Biosensor [64].

In addition, the ever-growing range of functional monomers and macromers widen their properties and potential applications. Hydrogels were used in early agricultural water absorbents based on biopolymers through grafting of hydrophilic monomers onto starch and other polysaccharides [65], [66]. Hydrogel products for hygienic applications are mainly based on acrylic acid and its salts. Whereas acrylamide is a main component employed for preparation of agricultural hydrogel products [54].

Various publications on this subject have discussed in detail synthetic methods and applications of hydrogels. For example, a comprehensive review of the chemistry and various synthetic schemes employed for hydrogel preparation can be found in various chapters of a compilation edited by Peppas [67] . More recently, hydrogels produced by radiation polymerization and grafting have been published by Khoylou [68]. Mi-Ran Park [69] described the preparation and chemical properties of hydrogels employed in agricultural applications. Vijayalakshmi and Kenichi have reviewed the potential of hydrogels in sensor utilization [70]. Dimitrios et al. [71] discussed the tailoring of hydrogels for various applications of medical interest.

A subset of Hydrogels are gels which have functional groups that possess a charge, and these are termed ionic hydrogels. There are three types of ionic polymers;

if the functional group is negative the polymer is anionic, if positive, cationic and if contains both a negative and a positive charge, zwitterionic. This means that ionic hydrogels can contain a net positive, net negative or neutral charge depending on environmental pH and ionic strength. Increasing number of ionic groups in hydrogels is known to increase their swelling capacities. This is mainly due to the simultaneous increase of the number of counterions inside the gel, which produces an additional osmotic pressure that swells the gel [72].

For example, many anionic hydrogels have an acid functional group such as acrylic acid (AA), whereas some cationic hydrogels are based on chitosan. These cationic hydrogels based on chitosan are well-known for their potential as delivery systems for protein therapeutics and antigens [73], including vaccines [74] (as Tetanus toxoid [75]) or enzymes (lipase [26-27]).

Photopolymerization

Free radical polymerization [78]

In step-growth polymerization reactions it is often necessary to use multifunctional monomers if polymer with high molar masses is to be formed; this is not the case when addition reactions are employed. Long chains are readily obtained from monomers such as vinylidene compounds with the general structure $CH_2=CR_1R_2$. These are bifunctional units, where the special reactivity of -bonds in the carbon-to-carbon double bond makes them susceptible to rearrangement if activated by free-radical or ionic initiators. The active center created by this reaction then propagates a kinetic chain, which leads to the formation of a single macromolecule whose growth is stopped when the active center is neutralized by a termination reaction. The complete polymerization proceeds in three distinct stages:

(1) initiation, when the active center which acts as a chain carrier is created;

(2) propagation, involving growth of the macromolecular chain by a kinetic chain mechanism and characterized by a long sequence of identical events, namely the repeated addition of a monomer to the growing chain; and

(3) termination, whereby the kinetic chain is brought to a halt by the neutralization or transfer of the active center.

Typically, the polymer formed has the same chemical composition as the monomer, i.e., each unit in the chain is a complete monomer, and not a residue as in most step-growth reactions.

An effective initiator is a molecule that, when subjected to heat, electromagnetic radiation, or chemical reaction, will readily undergo hemolytic fission into radicals of greater reactivity than the monomer radical. These radicals must also be stable long enough to react with a monomer and create an active center. Particularly useful for kinetic studies are compounds containing an azonitrile group, as the decomposition kinetics are normally first order, and the rates are unaffected by the solvent environment. For this work, persulfates are useful in emulsion polymerizations where decomposition occurs in the aqueous phase, and the radical diffuses into a hydrophobic, monomer containing, micelle.

$$S_2O_8^{2-} \rightarrow 2SO_4^{-}\cdot$$

A chain carrier is formed from the reaction of the free radical and a monomer unit; chain propagation then proceeds rapidly by addition to produce a linear polymer.

$$RM_1 + M_2 \rightarrow RM_n$$
$$RM_n + M_1 \rightarrow RM_{n+1}\cdot$$

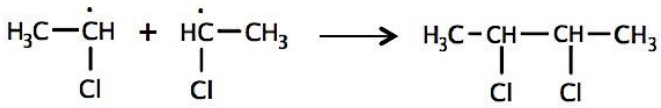
The average lifetime of the growing chain is short, but a chain of over 1000 units can be produced in 10^{-2} to 10^{-3} s. Bamford and Dewar have estimated that the thermal polymerization of styrene at 373 K lead to chains of x = 1650 in approximately 1.24 s, i.e., a monomer adds on once in every 0.75 ms.

In theory, the chain could continue to propagate until all the monomer in the system has been consumed, but for the fact that free radicals are particularly reactive species and interact as quickly as possible to form inactive covalent bonds. This means that short chains are produced if the radical concentration is high because the probability of radical interaction is correspondingly high, and the radical concentration should be kept small if long chains are required. Termination of chains

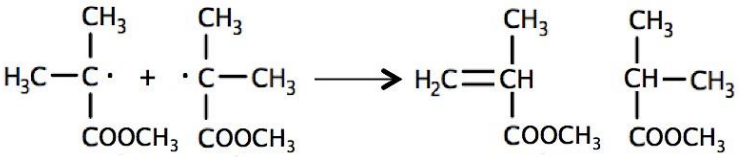
can take place in several ways by: (1) the interaction of two active chain ends, (2) the reaction of an active chain end with an initiator radical, (3) termination by transfer of the active center to another molecule which may be solvent, initiator, or monomer, or (4) interaction with impurities (e.q., oxygen) or inhibitors.

The most important termination reaction is the first, a biomolecular interaction between two chains end. Two routes are possible:

1. Combination, where two chain ends couple together to form one long chain.



2. Disproportionation, with hydrogen abstraction from one end to give an unsaturated group and two dead polymer chains.



One or both processes may be active in any system depending on the monomer and polymerizing conditions. Experimental evidence suggests that polystyrene terminates predominantly by combination, whereas poly(methyl methacrylate) terminates mainly by disproportionation when the reaction is about 333 K but by both mechanisms below this temperature. The mechanism can be determined by measuring the number of initiator fragments per chain using a radioactive initiator. One fragment per chain is counted when disproportionation is operative and two when combination occurs. Alternatively, the number-average molar mass of the product can be measured.

The majority of synthetic hydrogels are produced via free-radical polymerization. This means they require some initiation process, in industry this is normally a thermal process but, this project will develop a new technology platform that will lead to a multiple new product. Therefore, the hydrogels should be able to

quickly and simply be adapted to incorporate different active agents for slow release to the target area.

The UV Hg (UVA) system possesses good efficiency for photopolymerization; however, this light source emits harmful mercury contamination and generates a lot of heat. This weakness drives the need to use an environmentally friendly curing technology which is mercury free and at the same time can reduce heat generation. Thus, intense efforts to utilize more suitable and energy-efficient curing are being actively undertaken [79], [80]. UV LED lamps generate UV light which can tackle disadvantages of conventional UV Hg lamp technology. The utilization of UV LED is green since it does not emit light energy below 240 nm, which could prevent ozone production. In addition, it can offer a fast-curing rate and, thus, less energy is required to achieve complete polymerization. This reduction in operation downtime may lead to a significant saving in energy bills and cost effectiveness [79]. The radiation emitted from UV LED lamps is monochromatic in nature, with more energy concentrated in a restricted region at wavelengths of either 365, 385, 395 or 405-10 nm. [81]. The emission spectrum is always very specific and narrow, which can contribute to 60–80% more energy efficiency than conventional lamps at this given wavelength.

Yibo Wu. Et al. [82]. They use poly(ethylene glycol) diacrylate hydrogel (PEGda) and subsequently use interfacial polymerization to crosslink polyaniline structure within its network by UV crosslinking process. This methodology with commonly used manufacturing approach has the advantage of design and fabrication flexibility, low cost, high-speed and user-defined patterning. From figure 6, both hydrogels have low tensile strength. Note that single network hydrogels normally have low mechanical strength since they possess a high-water content.

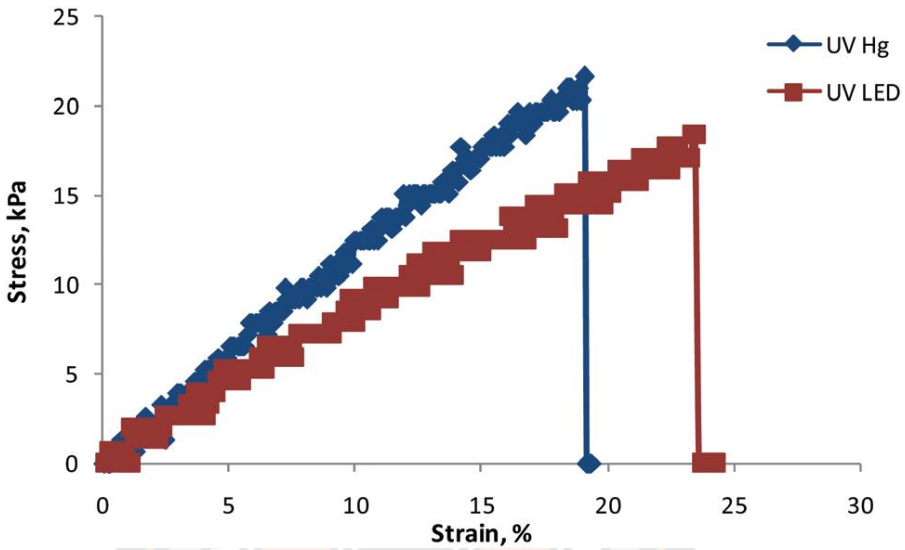


Figure 6 Stress–strain curve of PAAm hydrogels using WSPI via UV Mercury and UV LED system [82].

Nevertheless, PAAm hydrogels via a UV Hg have slightly higher tensile strength compared to PAAm hydrogels via a UV LED system and a strain at break for PAAm hydrogels via a UV Hg system was lower by 30% compared to UV LED cured PAAm hydrogels. So, from this graft, the UV mercury system were stiffer and more brittle than UV LED system.

In another work, Nur Farizah Ayub. Et al. [83]. A UV LED was used, the energy generated from its light source triggered photopolymerization to directly convert acrylamide and N-isopropylacrylamide monomers to polyacrylamide and poly(Nisopropylacrylamide) Hydrogels. Recent trends show that the UV LED light source can greatly reduce environmental effect without compromising performance as compared to conventional UV mercury-based lamp (UV Hg) system. Indeed, UV LED technology can be used to polymerize PAAm hydrogels with comparably high conversion to conventional UV Hg system [84].

In 2013, Ed Kiyoi et al. [85] have been interested to study the State of UV-LED Curing. They studied the benefits of UV-LED as compared to traditional mercury-arc UV lamps. Table 1 presents a comparison of the key characteristics of UV-LEDs and traditional mercury-arc UV lamps. Most UV-LEDs are specified to last

for 10,000 hours or more, compared to an arc lamp's lifespan of 500-2,000 hours. Additionally, UV-LED output only drops about 5% over their lifetime, while arc lamps can lose up to 50% of their original output. In a production environment, UV-LEDs require significantly less space, monitoring, maintenance, and downtime, resulting in higher productivity rates, less waste, and better end products. Retrofitting or replacing existing UV arc lamps with UV-LEDs can yield paybacks in as little as 12 months.

Table 1 A comparison of key characteristics of UV-LEDs versus traditional mercury-arc UV lamps [85]

Comparison of UV-LED to mercury-arc UV lamps

	UV-LED	Mercury Arc
Life	20,000+ hrs	500-2,000 hrs
On/Off	Instant	10 Minutes
Output Consistency	Very Good. 95%+	Drops up to 50%
Heat Generated	60°C	~350°C
Energy Efficiency	Saves 50-75%	
Environmental	Mercury Free, Ozone Free	Mercury Waste, Generates Ozone
Footprint	30-50% less	

Previous studies showed that UV LED system is a green technology and highly efficient as compared to UV Mercury (UV Hg) system. In 2015, Nur Farizah Ayub et al. [86]. Have been developed the UV LED curing formulations of polyacrylamide (PAAm) hydrogels. The formulations consisted of acrylamide (AAm) as a main monomer, N, N'-methylenebisacrylamide as a crosslinker and photo-initiator. UV LED emits monochromatic light sources only (365 nm or 385 nm). Thus, in order to developed formulation of UV LED curable hydrogels, a suitable water soluble photo-initiator ($\lambda \sim 365$ nm) has to be employed. A commercially available photo-initiator Oligo [2-hydroxy-2-methyl-1-[4-(1-methylvinyl) phenyl]

propanone] under the trade name Chivacure 300 (λ ~ 330 nm) was chosen in the first formulations. They also synthesized a photo-initiator based on 2,2-dimethoxy-2-phenyl acetophenone (DMPA) and methylated-β-cyclodextrin (MβCD) to be used in the second formulation. The complexation of DMPA and MβCD resulted in transparent and water-soluble supramolecular-structured photo-initiator (SSPI) (λ ~ 330 nm). Both formulations were irradiated using UV LED system (Hoenle AG, Germany, 365 nm) for 15 min. Synthesis of PAAm hydrogels with both photo-initiators has yielded almost complete conversion of hydrogels (> 80 %). Clearly, this study has revealed that enhanced formulation of UV LED curable hydrogels are due to appropriate choice of excellent water-solubility photo-initiators (Chivacure 300 and modified DMPA). They concluded that UV LED is an important tool for curing hydrogel formulations of various acrylate water-based monomers.

Poly(N-vinylformamide)

N-vinylformamide (NVF), as a precursor to high molecular weight amide and amine functional polymers, has shown attractive high reactivities in polymerization, copolymerization, and hydrolysis [87]. It was first synthesized more than three decades ago. Different synthetic routes to NVF have since been reported [88]. A recently developed production technology used two simple chemicals, acetaldehyde and formamide, to synthesize the precursor of NVF, ethylideneformamide. The latter was then heated to produce NVF via cracking with the help of a catalyst. This synthetic route has become commercially attractive and renewed industrial and academic research interest in NVF and its polymers.

N-Vinylformamide was first synthesized in1964 more by accident, it was originally tried to form 2-formylamino-propionitrile by pyrolysis. However, the desired product reacted on with elimination of HCN to N-Vinylformamide. In the BASF developed large-scale synthesis of N-Vinylformamide is from acetaldehyde and formamide prepared (see figure 7). First acetaldehyde (1) is reacted with hydrocyanic acid to lactonitrile (2). At this stage formamide is added and condensed, so that 2-formylamino-propionitrile (3) arises. This cleaves at elevated temperatures

hydrocyanic acid, so that eventually arises N-Vinylformamide (4). The cyanide going back, the reaction cycle.

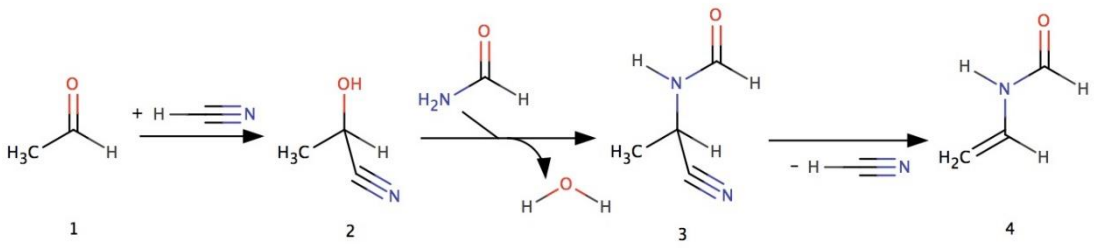


Figure 7 Synthesis of N-vinylformamide

NVF is an isomer of acrylamide (Aam) (see figure 8), but possesses some favorable properties over Aam such as; a lower toxicity [52], is more hydrophilic, more reactive [86] and is liquid at room temperature so has easier usage. The physical properties of poly(N-vinylformamide) (PNVF) gels are quite similar to the technologically important hydrogel polyacrylamide (PAAm). Polyacrylamide (PAAm) hydrogels are used in electrophoresis [30], chromatography [30], cosmetics [89], biomedical implants [32], superabsorbent products [33], and soil conditioners [90] among numerous other applications.

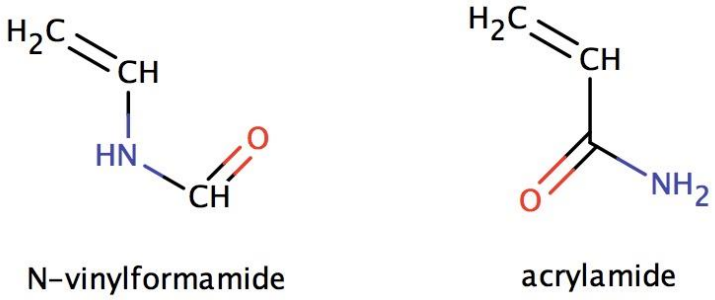


Figure 8 Chemical structures the isomer pair of N-vinylformamide and acrylamide

PNVF gels are also chemically related to poly(N-vinyl pyrrolidone) (PNVP) gels, another important biomedical hydrogel, widely used in contact lenses [34], drug delivery systems, and wound dressings. NVF monomer has been produced on the

commercial scale by several major chemical companies in the US, Europe, and Japan, though the market is still developing.

Poly(vinylamine); PVAm

PNVF is readily produced by conventional free radical polymerization methods. PNVF and its hydrolysis product polyvinylamine (PVAm) have been evaluated for use in areas such as the papermaking, water treatment, rad cure, and oil recovery [91]. PVAm is of significant technological interest because it is a high charge density cationic polymer and PNVF is a good precursor polymer for PVAm as the hydrolysis reaction proceeds quickly under mild hydrolysis conditions.

However, PVAm cannot be produced directly from monomer since vinylamine is not stable in its free state, and thus polyvinylamine (PVAm) must be synthesized through indirect methods, typically hydrolysis of polymers such as PNVF, poly(N-vinylcarbamate) or poly(N-vinylacetamide) [92]. PNVF is a good precursor polymer for PVAm as the hydrolysis reaction proceeds quickly under mild hydrolysis conditions.

In other work, L. Gu, S. Zhu et al. [93] studied the Acidic and Basic Hydrolysis of Poly(N-vinylformamide). N-Vinylformamide (NVF) was developed as a precursor for simple and economical production of PVAm. PNVF can be easily converted into PVAm by hydrolysis in either acidic or basic aqueous solution. Acid hydrolysis produces cationic polymers, whereas base hydrolysis yields polymers with free amine func- tional groups. Base hydrolysis is usually a more effective approach, with almost 100% completion. In contrast, acid hydrolysis conducted in the mixture of methanol and aque- ous solvent produces methylformate, which can be easily stripped as a light component. An alternative approach recently developed was a catalyzed thermal hydrolysis.12 The synthesis routes of PVAm via base or acid hydrolysis of PNVF are shown in Figure 9.

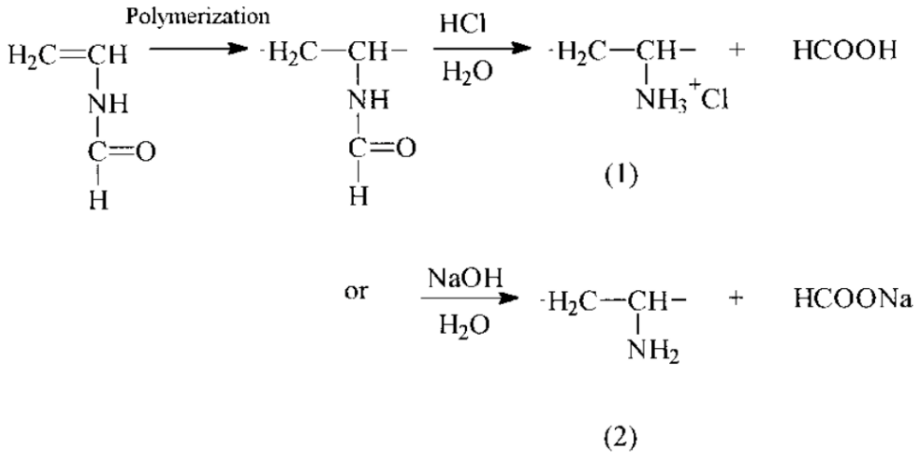


Figure 9 Synthesis route of polyvinylamine (PVAm) via (1) acidic and (2) basic hydrolysis of poly(N-vinylform- amide) (PNVF) [93]

In 2012, Joseph M. Scalet et al. [94] studied the novel synthesis of the polyelectrolyte IPN hydrogel derived from simultaneous hydrolysis two neutral networks is capable of ionic complexation. These hydrogels were made by hydrolysis of a neutral interpenetrating network (IPN) of poly (N-vinylformamide) PNVF and polyacrylamide (PAAm) networks to form an IPN of polyvinylamine (PVAm) and poly (acrylic acid) (PAAc) capable of intermolecular ionic complexation. Figure 10 shows a schematic of the types of hydrogels synthesized in this study and the The half of the hydrolyzed IPNs of PVAm/PAAc and PAAc/PVAm which were not treatments performed.

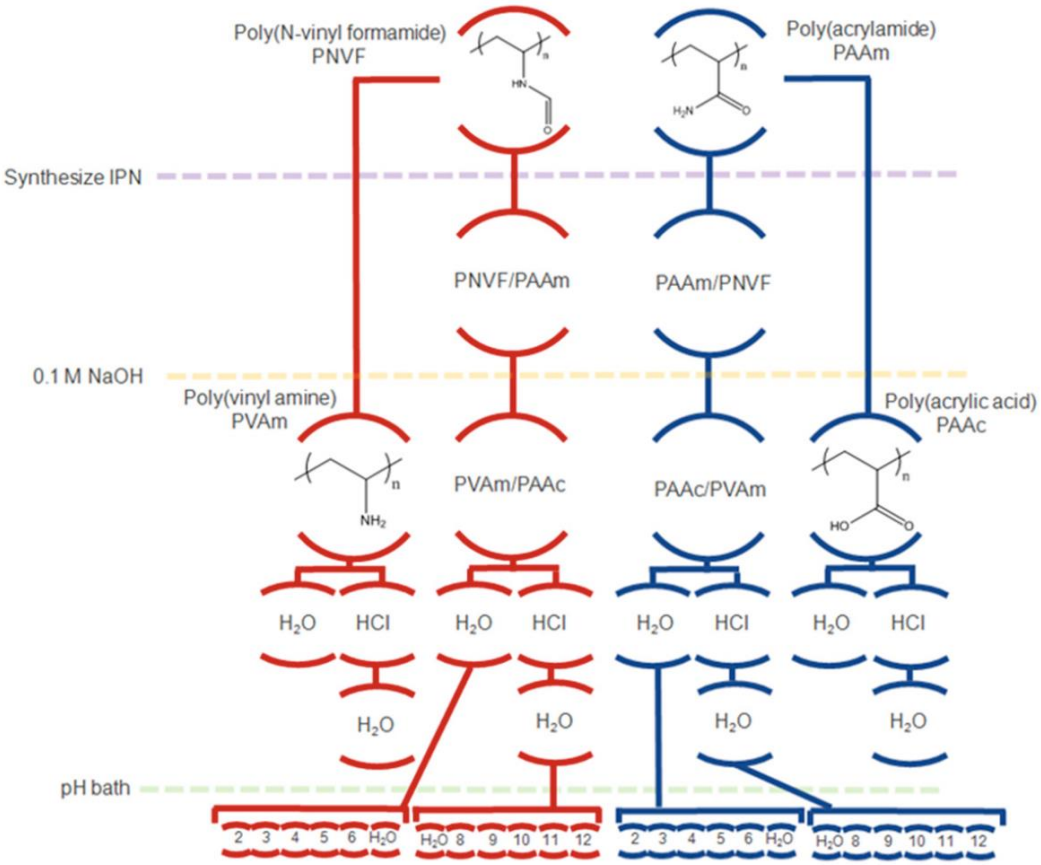


Figure 10 Schematic of formulations and the treatments [94]

Starting with two neutral monomers that are isomers of each other and exhibit similar χ parameters as with NVF and AAm, an IPN hydrogel formed was optically clear, which would be expected when there is a high degree of intermolecular mixing at a sub-micron scale and limited phase separation. It was shown that the order of the two networks in the sequential IPN, whether P(NVF)/PAAm, PAAm/P(NVF) or the hydrolyzed PVAm/PAAc, PAAc/PVAm did not matter, as the amounts of the two networks was found to be approximately equal which was confirmed by mass analysis, and the swelling and mechanical properties were statistically similar. However, after hydrolysis, physical properties changed significantly. At the higher pH values, the gels tended to be more brittle and have a higher swelling degree when compared to the unhydrolyzed IPN hydrogels, as shown in Figures 11 and 12.

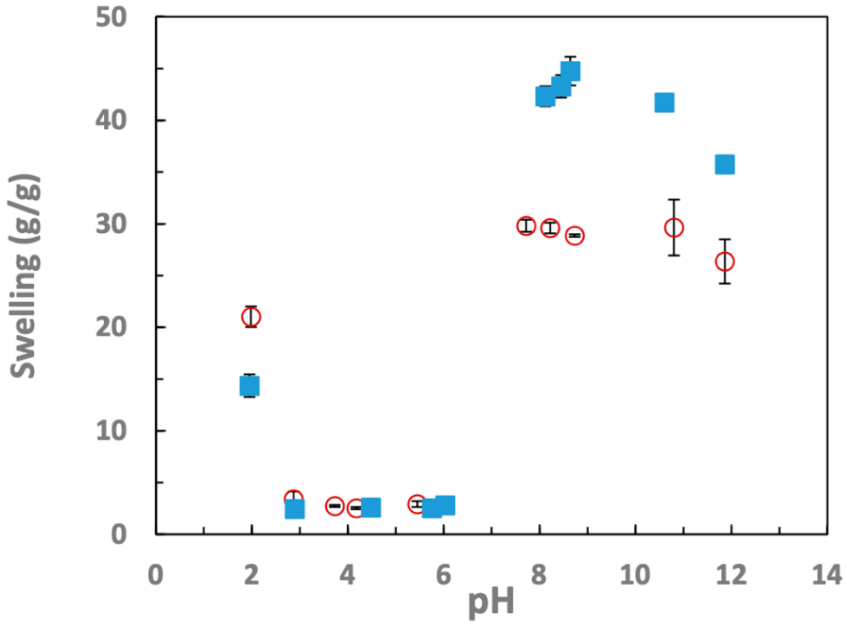


Figure 11 IPN swelling degree as a function of solution pH. The shape decrease in swelling between pH3 and 6 is consistent with charge complexation of the two networks. IPNs of PAAm/PNVF and PNVF/PAAm were partially hydrolyzes to PVAm/PAAc (blue closed symbols) and PAAc/PVAm (red open symbols)

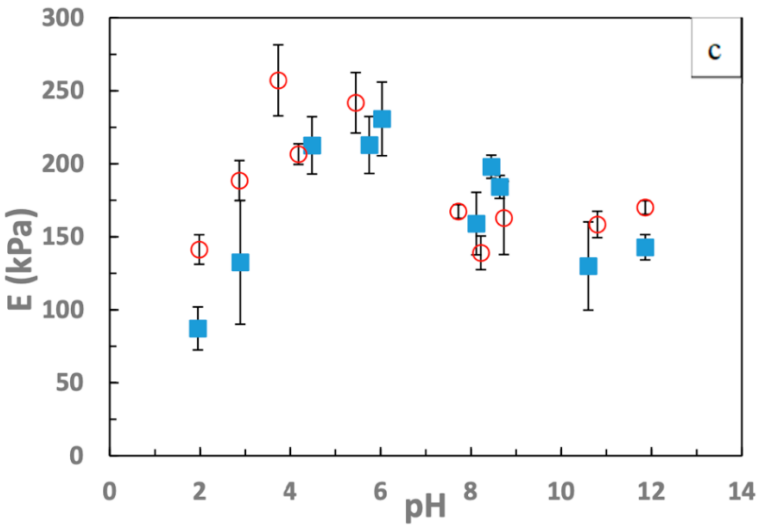


Figure 12 Changes in mechanical properties with solution pH. IPNs of PAAm/PNVF and PNVF/PAAm were partially hydrolyzes to PVAm/PAAc (blue closed symbols) and PAAc/PVAm (red open symbols)

The increase in swelling in basic solution is due to the poly(vinylamine) groups being in the free base form of R-NH_2 while the poly(acrylic acid) are in the charged sodium salt form R-COO-Na^+. This creates a difference in mobile ion concentration between the inside the gel and the solution, leading to a large increase in the osmotic swelling pressure, thus causing a highly swollen gel [49-51].

Yukie Takemoto, et al. [98], they were prepared a surface-modified hydrogel with a PIC (surface-PIC gel: sPIC gel), which comprised two segments: a stable swollen segment composed of poly-(NVA-co-NVF) and PAAc for drug release, and a stimuli responsive layer composed of PIC for controlled drug release. These polymers are hydrophilic and nonionic, and their amide groups are easily hydrolyzed with acidic or basic aqueous solutions to produce cationic PVAm, which can form a PIC with anionic PAAc.

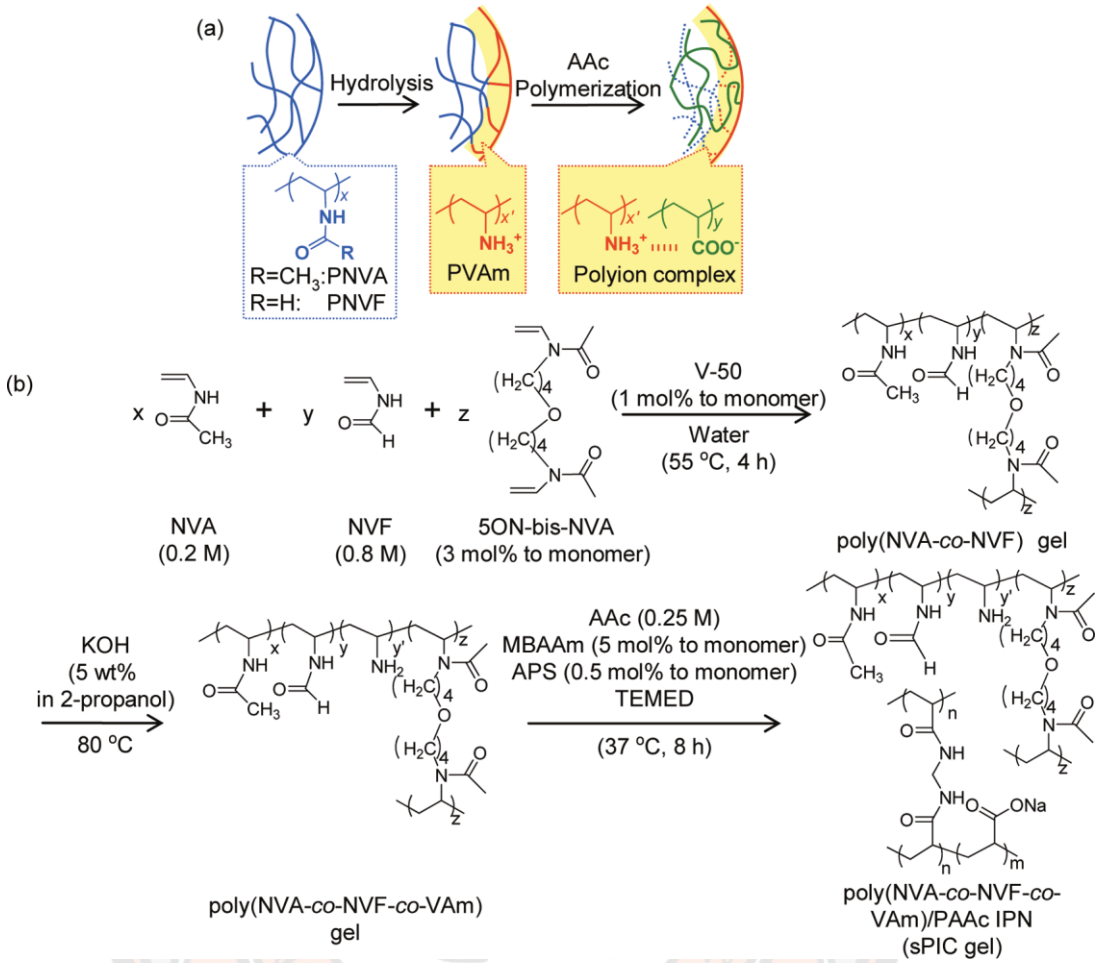


Figure 13 Preparation of a surface polyion complex hydrogel (sPIC gel). (a) Schematic illustration of preparing an sPICgel in three steps. (1) Preparation of a poly(NVA-co-NVF) hydrogel. (2) Hydrolysis of the amino group in poly(NVA-co-NVF) (3) Aac polymerization to obtain an sPIC gel. (b) Scheme of preparation of sPIC gel [98]

The sPIC gel, a surface-modified hydrogel with a PIC layer, was successfully prepared via hydrolysis of the surface of poly(NVA-co-NVF) gel, the sPIC gel has a continuous polymer network, and possesses different functions between the surface and the interior of the hydrogel; the surface segment is designed for the controlled release of drug molecules, and the interior of the gel is designed for stable drug retention.

Cosmetic hydrogels

Although the main use of hydrogels in the cosmetic industry is in the contact lens market, the use of hydrogels as wound dressings and their potential for delivery of active agents has lend to use in other cosmetic applications. Cosmetic industry is a market constantly increasing in dimension and products offered. One of the reasons of this behaviour is related to the long path to approval necessary for medical devices, medical procedures, drugs or biomolecules, in order to make allowed to be sold on the market. As the cosmetic market requires less time and money for the approval of a product, it is common to test new inventions and devices in cosmetics before and in more risky clinical applications.

For a product to be approved in cosmetics, the most important parameter to be assessed is Primary Irritation Index (PII). This index is simple to obtained and exist both for skin and eyes, indeed for each level of PII corresponds a determinate effect [99]. Considering that the majority of hydrogels used in this field are suitable for cells culture and for other biomedical applications (e.g. wound dressings), is not surprising that their irritation index is among the lowest. Thus, with a relatively small investment, companies are able to launch on the market new cosmetic products based on hydrogels, such as “beauty masks”.

Figure 14 Example of the cosmetic products based on hydrogels

Usually made with engineered collagen (MasqueologyTM by SEPHORA USA Inc., BioCollagen Cosmeceuticals by NOVOSTRATA UK Ltd.), hyaluronic

acid (SEPHORA USA Inc.), or polyvinylpyrrolidone (Pecogel®), these masks claim to hydrate the skin, restore its elasticity and promote anti-aging actions [100]. Pecogel by Phoenix Chemicals Inc., has a wide selection of hydrogels, based on polyvinylpyrrolidone, with differences in composition and/or crosslinking method. Furthermore, in some of the commercially available compounds such as Hydro Gel Face Masks by Fruit & Passion Boutiques Inc., the moisturizing action of these organic polymeric gels is coupled with more complex drug-delivery systems developed to release of biomolecules like vitamin C or B3.

The cosmetic industry is on the cutting edge of hydrogels; indeed, a pH-Sensitive material P(MAA-co-EGMA) has been developed for release of cosmetics drugs like arbutin, adenosine, and niacinamide, well knowing molecules for wrinkle treatment and for skin-whitening [101]. This hydrogels change is permeability responding to the pH changes: At pH 4.0 it holds the pharmaceuticals inside the matrix, when in contact with skin, at pH 6 and above, the permeability rises, and the drugs will be delivered.

For cosmetic hydrogels, the approval of a product it is common to test new inventions and devices in cosmetics before and in more risky clinical applications. For example, in the part of physical properties testing, S. Silvia et al. [102] formulated the hydrogel mask with the ethanolic extract of noni fruit, sodium alginate, and xanthan gum. This formulation produces a hydrogel mask that has good physical stability and consistency. Physical stability was evaluated using organoleptic observation, consistency rates, swelling index measurements, and mechanical strength.

The development of the transdermal patch based on a hydrogel matrix has recently received considerable attention. The physicochemical properties including mechanical strength, water holding capacity, bio-adhesion ability, and the rate-controlling characteristic of the developed patch can be modified by varying types and concentration ratios of the polymer blends. A range of polymers, whether natural or synthetic, has been employed in hydrogel system [103].

J. Viyoch et al. [103] developed a new matrix system of the bio-adhesive hydrogel patch by blending chitosan with various types of starch, coupled with the presence of crosslinking agent in the polymeric matrix for controlling the release of hydrophilic compounds including tamarind's AHAs. The results showed, the variations in the type of polymer used, the polymer ratio and the amount of crosslinking agent affected the physicochemical properties of the hydrogel patch, thereby influencing the physical characteristics, mechanical resistance, bio-adhesion property, surface morphology and finally the release profile of the active compound, tartaric acid.

To determine the safety of the developed hydrogel patch, a preliminary study assessing the irritation effects of the product after single application was designed and conducted. J. Viyoch et al. [104] focus on the development of the hydrogel patch for personal care purposes by using natural origin polymers like chitosan from various sources. For determination of skin irritation after application of the hydrogel patch, the protocol of the clinical study was approved by Human Ethical Committee, Naresuan University, Phitsanulok, Thailand.

pH sensitive hydrogels for delivery of active ingredients

Many academic papers about possible applications of hydrogels in drug delivery have been published, however, only a few have resulted in commercial products [53]. The drug can be loaded into a hydrogel and then its release may proceed through several mechanisms: diffusion controlled, swelling controlled, chemically controlled and environmentally responsive release.

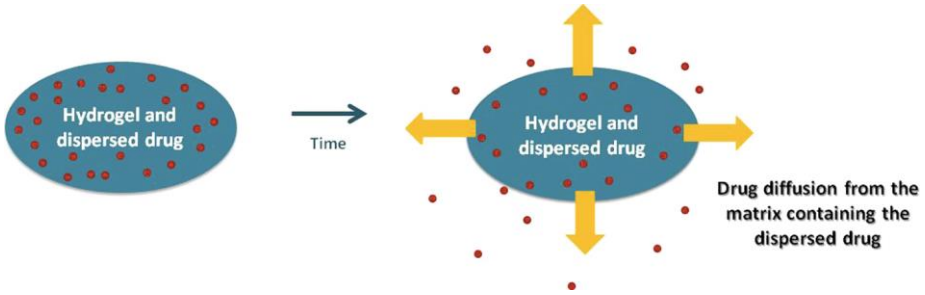


Figure 15 Scheme of drug release from matrix systems [53]

From figure 15, in matrix systems the drug is dispersed or dissolved uniformly throughout the three-dimensional structure of the hydrogel. Drug release is achieved through the macromolecular mesh or the pores, and the initial release rate in this case is proportional to the square root of time, rather than being constant and time independent as happens in reservoir systems [105]. In addition, controlled release based on a large volume change would become a problem when the hydrogel is embedded in a small space such as the defective tissues or in the soil for water retention, because it produces a physical gap between the hydrogel and the exterior [98].

Lately, much effort has been focused on the development of stimuli-responsive systems adapted to the dermatology and cosmetology fields. The delivery of active ingredients to the skin is a true challenge for researchers trying to combine efficacy with this local and non-traumatic route of administration. Smart drug delivery systems is a promising research path since skin conditions or dermatoses can lead to imbalances in the skin physiological parameters like pH, temperature or redox potential [60-64]. To facilitate drug delivery by topical application, smart drug delivery systems (SDDS) can be used as an alternative or complement to the above-mentioned skin delivery technologies. In 2012, Louise Van Gheluwe et al. [111] provides an overview of polymer-based smart drug delivery systems (SDDS) that have been developed to trigger the release of active ingredients for treating various skin conditions and pathologies. The article also discusses the methods used to test the stimuli-responsiveness of these systems, both in vitro, ex vivo, and in vivo. The preparation of these smart gels is obtained by using stimuli-responsive polymers. Figure 16 compiles the different stimuli and their mode of action used in the skin area.

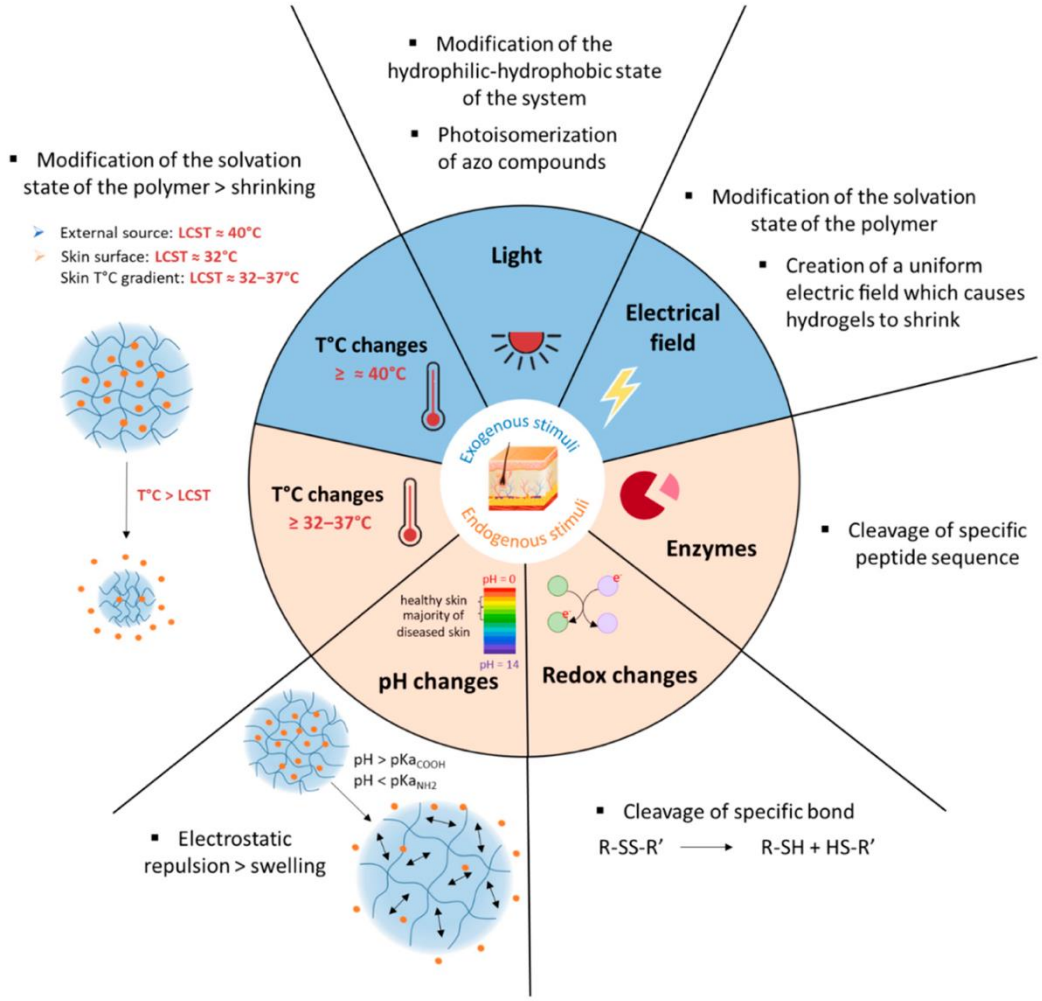


Figure 16 Schematic overview of stimuli and their mode of action, applied to the design of smart drug delivery systems for skin applications [111]

One strategy for designing smart drug delivery systems (SDDS) based on cationic polymers (which contain amine groups) is to prevent the initial burst release of active ingredients into the dosage form at neutral pH. Once in contact with skin that is more acidic (with a pKa below that of the amine groups), the drug release is triggered [66-67]. In a pH-responsive system, the release of the drug is often associated with the swelling it stimulates. However, in one pH-responsive system study, the release of the drug was associated with the shrinking of the cationic system.

For another work, Zhu et al. [112] studied pH-sensitive methacrylated chitosan (MAC) hydrogels, and significant decreases in the swelling ratios were observed

when the hydrogel was exposed to increasingly alkaline environments (pH 3, 5, 7.4 and 9). Indeed, the amino groups of MAC (pKa = 6.5) will be deprotonated when the environmental pH is higher than the functional group's pKa. Therefore, the number of positive charges in the network of chitosan-based hydrogel decreases as the pH increases, which results in the shrinking of the system (because there are less electrostatic repulsions). Results showed that MAC hydrogels have a potential application in responding to specific wound healing stages by pH dependence and accelerate wound healing by the liberation of antibiotics and anti-inflammatory drugs.

From the another studied, in 2012 Muhammad Rizwan et al. [113] studied the drug Release Mechanism of pH Sensitive Hydrogels. Drug release due to the reactions of hydrogels (hydrolytic or enzymatic degradation of polymer chains) is said to follow a chemically controlled mechanism. The general mechanism of pH dependent swelling as well as drug release is shown in figure 17.

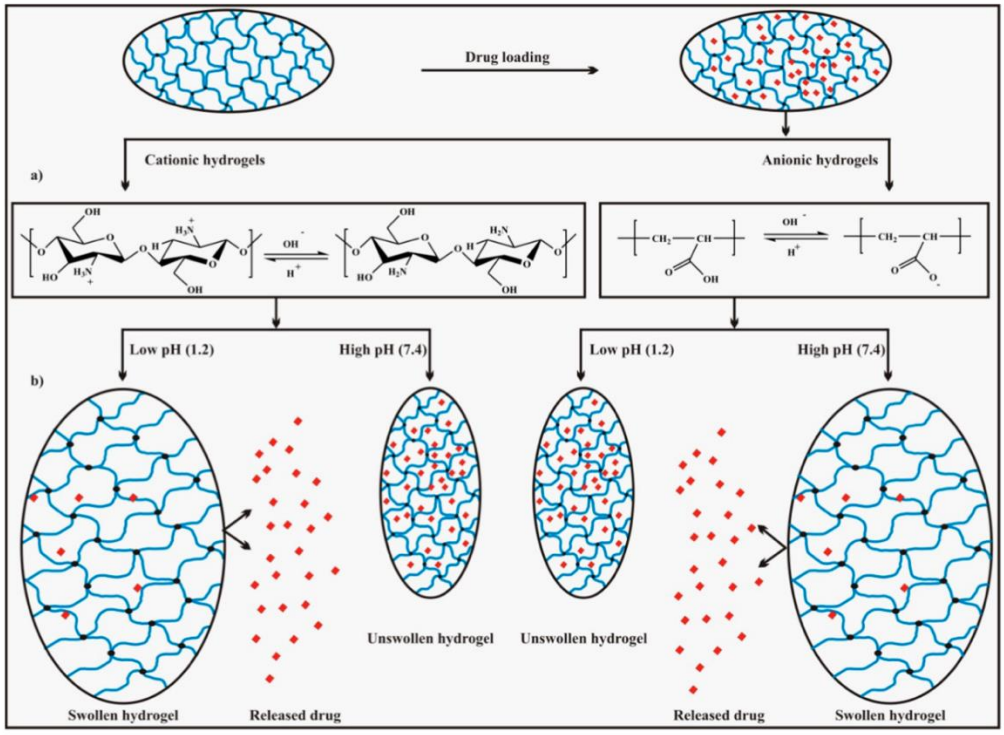


Figure 17 (a) pH dependent ionization of specific acidic or basic functional groups on hydrogel chains responsible for swelling, (b) pH dependent swelling and drug release mechanism [113]

Eunmi Lee and Bumsang Kim [114] were synthesized the potential to be used as a smart carrier for cosmetic ingredients triggered by an external pH change for cosmetic applications. The pH-responsive copolymer of MAA and PEGMA, designated P(MAA-co-EGMA), hydrogel microparticles were prepared and the feasibility of the P(MAA-co-EGMA) hydrogel microparticles as delivery carriers was evaluated with these cosmetic ingredients. Therefore, the pH-responsive swelling and release behavior of the hydrogel microparticles, the loading efficiency of the cosmetic ingredients in the hydrogel microparticles, and the skin permeability of the cosmetic ingredients as a function of pH were investigated. The cumulative percentage of cosmetic ingredients released from the particles as a function of time is shown in Figure 18.

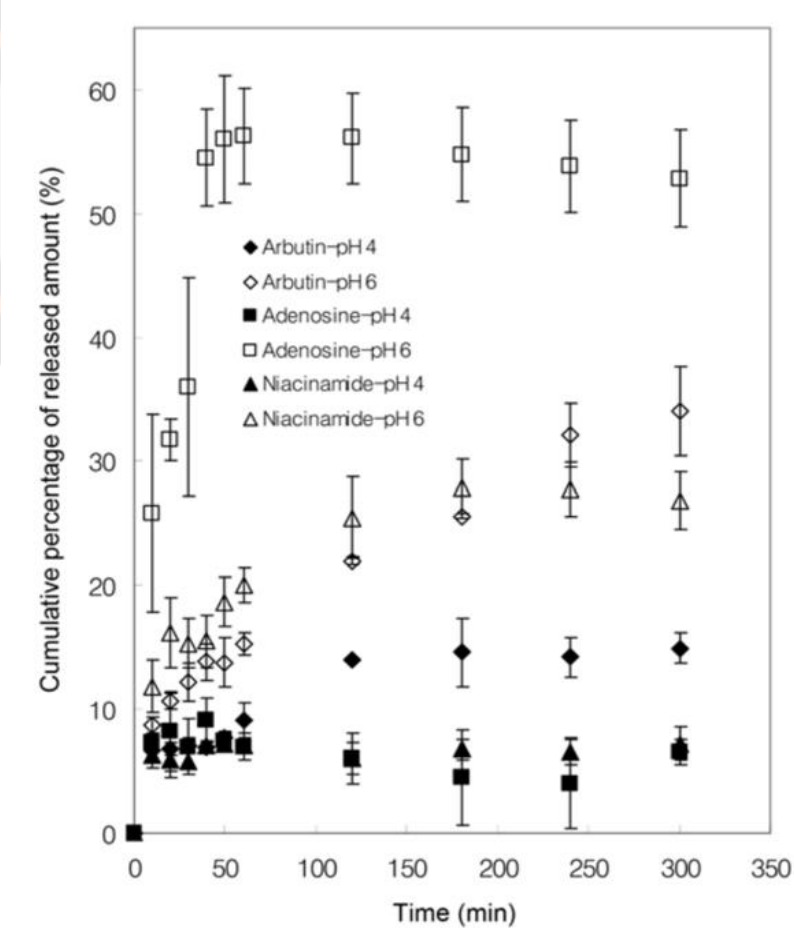


Figure 18 Cumulative percentage of cosmetic ingredients released from P(MAA-co-EGMA) hydrogel microparticles with MAA:EG = 0.6 at pH 4 and 6

Therefore, researchers have been concluded that the loading efficiency of the cosmetic ingredients was affected by the electrostatic interaction between the hydrogel and the cosmetic ingredients. The P(MAA-co-EGMA) hydrogel microparticles showed a pH-sensitive release behaviour. At low pH (pH 4) small amounts of cosmetic ingredients were released from the particles, while at high pH (pH 6) relatively large amounts of cosmetic ingredients were released from the particles. Thus, at pH 4 almost none of the cosmetic ingredients except adenosine permeated through the skin, while at pH 6 relatively high skin permeability was obtained. These results indicate that the P(MAA-co-EGMA) hydrogel microparticles synthesized in this study have the potential to be used as a smart carrier for a cosmetic ingredient such as arbutin, adenosine, or niacinamide, triggered by an external pH change for cosmetic applications.

Water states in hydrogels

The water present in hydrogels is often defined as a percentage at equilibrium, known as the equilibrium water content (%EWC). This value is very important because it is directly related to the behavior of hydrogels in the desired application. However, understanding how water interacts with polymer chains or how water structuring affects the behavior of gels is also crucial.

Water mobility plays a crucial role in determining transport properties of small molecules in polymer matrices. In drug delivery systems, water state affects the pharmacokinetics, especially drug absorption, diffusion and release. Also in controlled drug delivery systems, the adsorbed water state affects the pharmacokinetics, influencing drug solubility, absorption/desorption, diffusion and matrix interaction with the body tissue [115]. The study of water in different surfaces has been driven by the interest in investigating the effects of water on the functionality of these surfaces. As mentioned above, it includes hydrophobic/hydrophilic surfaces, metals, polymers, among others. Because of the large number of studies on this matter, a summary of the different denominations that water receives on surfaces is presented [116].

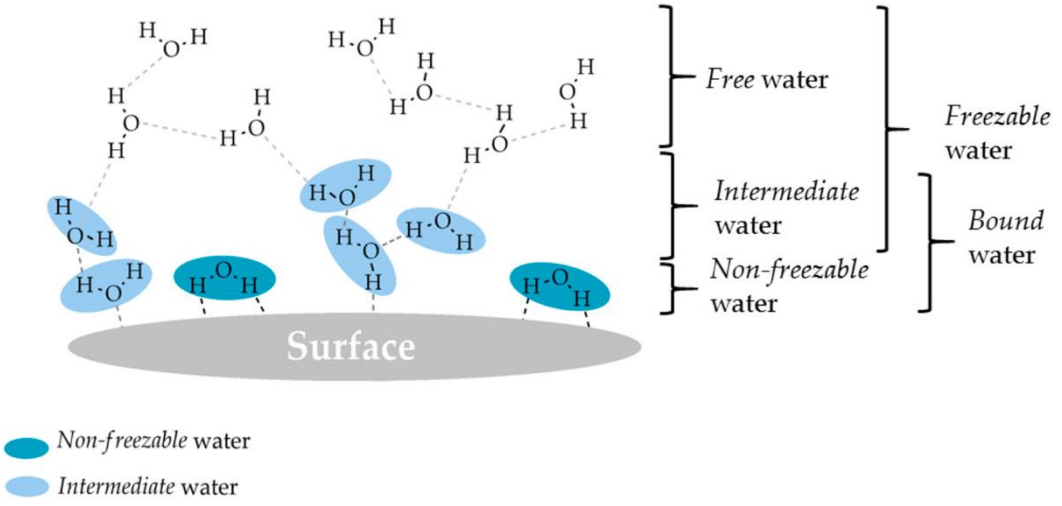


Figure 19 Types of water on surfaces with the denominations, their structure and interactions on surfaces [116]

They refer to the different types of water as: non-freezable water, intermediate water and free water, according to the freezing temperature criterion. Non-freezable and intermediate water are bound water, while intermediate and free water are freezable water (Figure 19). In terms of structure and interaction with surfaces, the three water types are characterized by the following [72-73]:

- Non-freezable water is tightly bound to the surface and the water-surface interactions are very strong, while water–water interactions are very weak.
- Intermediate water interacts moderately with the surface (stronger than free but weaker than non-freezable water), involving both water-surface and water–water interactions.
- Free water hardly interacts with the surface and there is mainly water–water interaction.

Differential Scanning Calorimetry [74-76]

The identification of the states of water on a surface and their quantities can be analyzed using DSC. Figure 20 shows a DSC thermogram of a hydrated surface containing the three types of water: non-freezable, intermediate, and free water. The

exothermic peak below 0◦C corresponds to the cold-crystallization of water and indicates the presence of intermediate water.

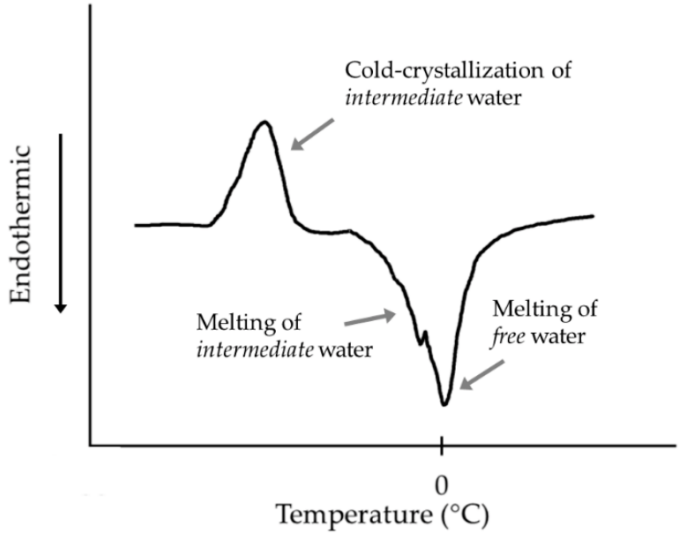


Figure 20 Schematic of a DSC of a hydrated surface with three stages of water. Non-freezable water not visible in the thermograms. Modified from [119]

The thermogram gives useful information to calculate the amount of the types of water. The mass of *intermediate* (W_{int}) and *free* water (W_f) can be calculated according to the following equations:

$$W_{int} = \frac{Q_{cc}}{\Delta H_{cc}} \quad (1)$$

$$W_f = \frac{Q_m}{\Delta H_m} - W_{int} \quad (2)$$

where ΔH_{cc} and ΔH_m are enthalpy changes in the cold-crystallization and the melting of ice, respectively; and, Q_{cc} and Q_m are the heat absorbed during the cold-crystallization process and the melting process, which are obtained from the area of the respective peaks in the thermogram. The enthalpy changes (ΔH_{cc} and ΔH_m) are assumed to be the same as that of bulk water (334 $J \cdot g^{-1}$).

The mass of *non-freezable* water (W_{nf}) is calculated as follows:

$$W_{nf} = \text{EWC (wt\%)} - W_{int} - W_f \quad (3)$$

where EWC is the equilibrium water content of the sample.

The number of *non-freezable* (N_{wnf}) and *freezable* (N_{wf}) water molecules per polymer repeating unit can be calculated using the DSC information, as previously described. The weight ratio ($WR_{nonfreezable}$) of *non-freezable* water/polymer, and the weight ratio ($WR_{freezable}$) of *freezable* water/polymer c be calculated using the following equations:

$$WR_{nonfreezable} = \frac{W_{nonfreezable}}{W_{polymer}} = \frac{(\text{EWC} - W_{freezable})}{W_{polymer}} \quad (4)$$

$$WR_{freezable} = \frac{W_{freezable}}{W_{polymer}} \quad (5)$$

where $w_{nonfreezable}$, $w_{freezable}$, and $w_{polymer}$, are the weight percentages of *non-freezable* water, *freezable* water, and *polymer*, respectively. $w_{freezable}$ is obtained from the DSC thermograms and calculated as follows:

$$WR_{freezable} = \frac{Q_m}{\Delta H_m} \text{ x } 100\% \quad (6)$$

Finally, N_{wnf} and N_{wf} are obtained using the equations:

$$N_{wnf} = \frac{M_p}{M_{water}} \text{ x } WR_{nonfreezable} \quad (7)$$

$$N_{wf} = \frac{M_p}{M_{water}} \text{ x } WR_{freezable} \quad (8)$$

where M_p is the molecular weight per polymer repeating unit, and M_{water} is the molecular weight of water.

Therefore, in this research work, for fabrication of the novel cosmetic hydrogels patch, glycerin was used as a humectant agent for enhance the skin moisturizers and also enhances the gel flexibility in term of tensile properties. In 2022, H. J. Chen et al. [122] were to analyze the moisture-retention capacity of glycerin, a common ingredient in cosmetic products. Specifically, this study applied gravimetric analysis, transepidermal water loss (TEWL) analysis, and differential scanning calorimetry (DSC) to examine the evaporation of glycerin solutions of different concentrations. DSC thermograms further confirmed the consistent results and identified two hydrated water microstructures (nonfreezable water and free water) in the glycerin solutions, which explained why the measured evaporation rate decreased with the glycerin concentration. These findings can be applied to prove the moisture-retention capacity of a humectant in cosmetic products by different measuring methods.

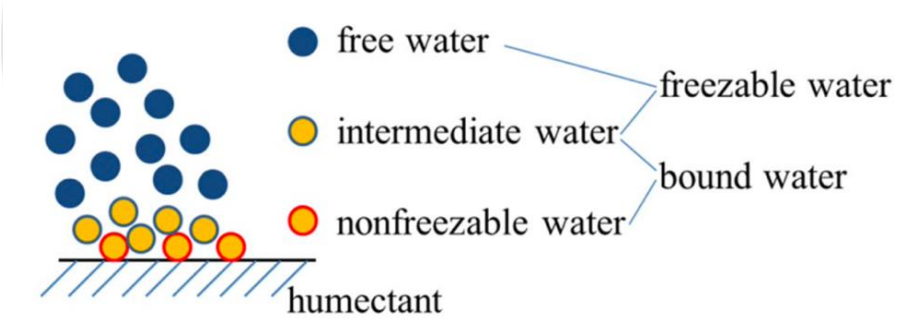


Figure 21 Three hydrated water types in a humectant

Therefore, DSC is a powerful tool for exploring the microstructure and thermal behavior of a liquid sample [123] also be applied for evaluating the moisture retention of a humectant [124]. According to the freezing temperature criterion, the microstructure of water in a humectant can be categorized into three types: nonfreezable water, intermediate water, and free water [72-75], as shown in Figure 21 for three hydrated water types. Nonfreezable water and intermediate water can easily bind to a humectant through hydrogen bonding and are thus called bound water. Intermediate water and free water can exhibit phase transitions and are thus called

freezable water [128]. Non- freezable water tightly binds to the hydrophilic sites of a humectant and has low mobility because of the strong water–humectant interactions. Specifically, nonfreezable water involves very weak free water–water interactions. Intermediate water is oriented around nonfreezable water and the humectant as a hydration shell, forming cage- like structures through which the maximum number of hydrogen bonds is achieved in the available space [129].

CHAPTER III

RESEARCH METHODOLOGY

This chapter is concerned with information related to materials, instruments and the research methodology used in this research work. This chapter is divided into three parts. The first part is fabrication and preparation hydrogels, which describes poly(N-vinylformamide) based materials via photopolymerization and then the synthesis of the NVF-copolymer hydrogels including the cationic poly(vinylamine) by partially hydrolyzing the amide group to form PVAm under acid hydrolysis conditions. The second part is the investigation and characterization of hydrogels, which characterized the base gel properties and applied gel properties to assess the properties of this novel hydrogel system of poly(N-vinylformamide) based materials. The third part is specific testing of application properties, these include tests that are directly related to the future cosmetic application, such as *in vivo* testing as well as the a small clinical trial.

Materials

Table 2 The information of materials used in this work

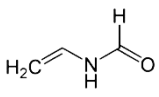
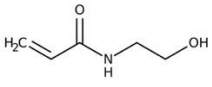
Name	Abbre-viation	Chemical structures	Sources/ Companies	Used As
N-vinylformamide	NVF		Sigma-Aldrich Co. Inc, Singapore	Monomer
N-Hydroxyethyl acrylamide	HEA		Sigma-Aldrich Co. Inc, Singapore	Monomer

Table 2 (cont.)

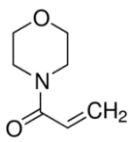
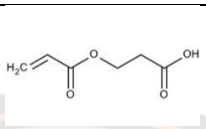
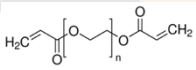
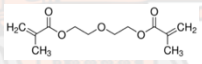
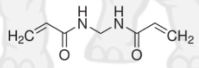
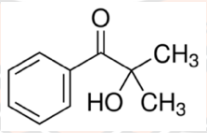
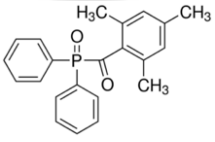
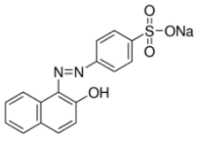
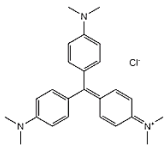
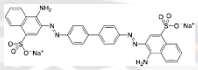
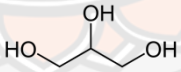
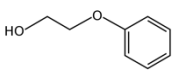
Name	Abbre-viation	Chemical structures	Sources/ Companies	Used As
4-Acryloyl morpholine	AMO		Sigma-Aldrich Co. Inc, Singapore	Monomer
2-Carboxyethyl acrylate	CEA		Sigma-Aldrich Co. Inc, Singapore	Monomer
Poly(ethylene glycol) diacrylate (MW 575)	PEGDA		Sigma-Aldrich Co. Inc, Singapore	Cross-linker
Di(ethylene glycol) diacrylate	DEGDA		Sigma-Aldrich Co. Inc, Singapore	Cross-linker
N,N-methylenebis-acrylamide	MBAAm		Sigma-Aldrich Co. Inc, USA	Cross-linker
2-Hydroxy-2-methylpropio-phenone	HMPPN		Sigma-Aldrich Co. Inc, Singapore	Photo-initiator of UVA system
Diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide	TPO		Sigma-Aldrich Co. Inc, Germany	Photo-initiator of UVLEDs system
Orange II sodium salt dye	O2S		Sigma-Aldrich Co. Inc, Singapore	For dye release

Table 2 (cont.)

Name	Abbre-viation	Chemical structures	Sources/ Companies	Used As
Crystal violet dye	CV		Sigma-Aldrich Co. Inc, Singapore	For dye release
Congo red dye	CR		Sigma-Aldrich Co. Inc, Singapore	For dye release
Phosphate buffered Saline	PBS	-	Sigma-Aldrich Co. Inc., Singapore	For pH buffer and dye release
Sodium hydroxide	NaOH	-	Ajax Finechem, Australia.	For pH buffer
Hydrochloric acid	HCl	-	Ajax Finechem, Australia.	For hydrolyze hydrogels
Glycerin	-		Sigma-Aldrich Co. Inc, Singapore	For forming hydrogels; skin moisturizer
Phenoxyethanol	-		Chanjao Longevity Co., Ltd	For forming hydrogels; preservative
Lactobionic acid	-	-	Chanjao Longevity Co., Ltd	Cosmetic ingredients

Instruments

1. Contact angle (CA, Dataphysics Model OCA20)
2. Differential Scanning Calorimetry (DSC, Mettler Model DAC1)
3. Microplate reader (Biotek, Model Synergy H1 Hybrid Reader)
4. Universal testing machine (INSTRON CALIBRATION LAB model 5965)
5. Fourier Transform Infrared Spectroscope (FT-IR, Perkin Elmer Spectrum GX, 4000-400 cm^{-1})
6. Mexameter® Model MX 18; Courage and Khazaka Electronic GmbH
7. Tewameter® Model TM210; Courage and Khazaka electronic GmbH

Methodology

Part 1 : Preparation and fabrication of hydrogels

To produce novel cosmetic hydrogels of poly(N-vinylformamide); PNVF based materials via photopolymerization, the initiation process was first to studied. Then, the detail of fabrication of PNVF-copolymer hydrogels were described with varies the composition for the formulation of the hydrogels and also synthesized the hydrogels containing the cationic poly(vinylamine).

Non-hydrolysed hydrogels: Fabrication of poly(N-vinylformamide) based materials via photopolymerization for a novel cosmetic hydrogels

To produce poly(N-vinylformamide) and copolymer hydrogels were prepared by UVLEDs photopolymerization. The method of preparation of PNVF-copolymer containing the vinyl group and undergo free radical addition polymerization is schematically in figure 22. N-vinylformamide (NVF) was used as a monomer and N-Hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl Acrylate (CEA) were used as the copolymer and using crosslinking and photo-initiator for reaction with different the effect of composition of hydrogels.

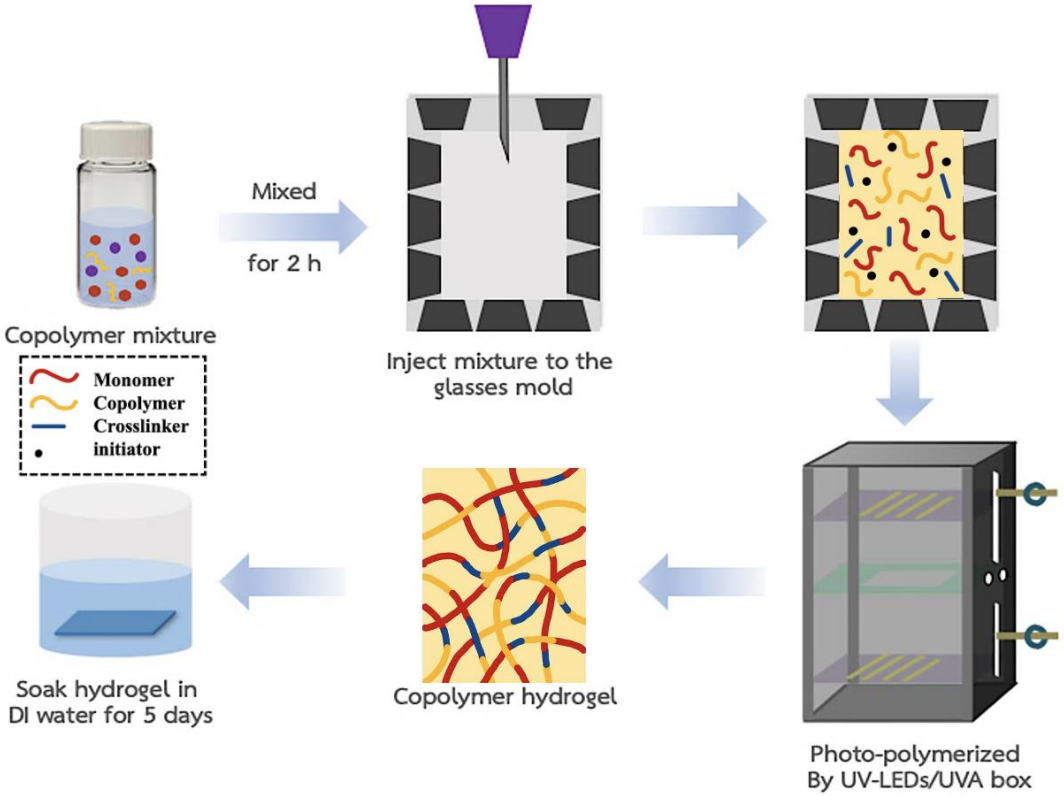


Figure 22 Schematic of preparation of PNVF-copolymer hydrogels

N-vinylformamide (NVF) and its copolymer are all added together and mixed at 300 rpm for 2 h thoroughly. This solution is then injected into a mould between plastic gaskets of a known size (controls the dimensions of the sheet hydrogel) and two glass plates covered with a PET sheet to prevent adhesion. The mould is then placed under UVLEDs lamps box for 2 minutes to cure the comonomer mixture into a cross-linked polymer network, and then washed with de-ionized water to remove the unreacted monomer before further use. The swollen hydrogel was soak by de-ionized water for 5 days (the water was changed every day).

Hydrolysed hydrogels: Synthesis of the PNVF-copolymer hydrogels containing the cationic poly(vinylamine); PVAm

First, a PNVF and copolymer hydrogels was prepared. The cross-link network was achieved by using 0.5% of N,N-methylenebisacrylamide (MBAAm) as a crosslinker, 1% of PI and hydroxyethyl acrylamide (HEA) were used as copolymer.

For synthetic of hydrogels containing the cationic poly(vinylamine), poly(NVF-co-HEA) (75:25) hydrogels was first prepared by via UVLEDs photopolymerization. Then, the poly(NVF-co-VAm-co-HEA) hydrogels were prepared by partially hydrolyzed amide group of poly(NVF-co-HEA), is a good precursor polymer for PVAm under acid hydrolysis conditions. For hydrolysis hydrogels, the 5% of 2M hydrochloric acid was added to 250 mL deionized water into a three-neck round bottom flask, after that the hydrogel samples were placed into three neck round bottom flask and refluxed at a temperature of 80 ^{o}C for 8 hours to produce PVAm is schematically in figure 23.

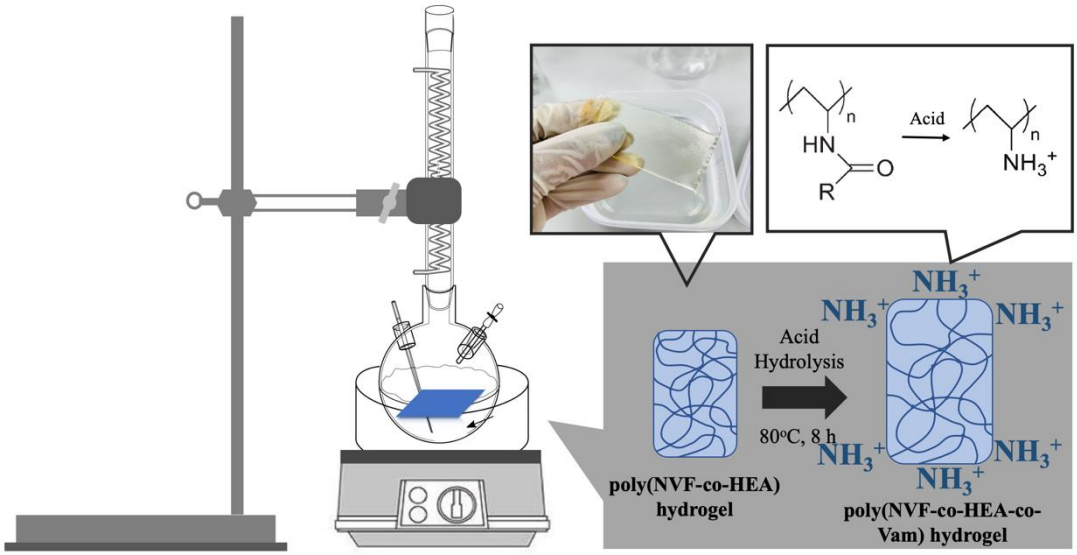


Figure 23 Schematic picture of synthesis of poly(NVF-co-VAm-co-HEA) hydrogels

After hydrolysis, the poly(NVF-co-VAm-co-HEA) hydrogels were rinsed with phosphate buffered Saline solution (pH 7.4) to remove the unreacted acid before

further use and then each hydrogel was immersed into de-ionized water for 5 days (the water was changed every day) to remove reaction residues.

Part 2: Investigation and characterization of hydrogels

The characterization of the novel cosmetic hydrogels of poly(N-vinylformamide) based materials, was to assess the base gel properties as a guideline for the most suitable for cosmetic hydrogels to take further into the self-perception questionnaire. Then, a more detailed characterization of hydrogels was undertaken over various compositions in order to formulate the hydrogels and also find the idea composition for hydrogels containing the cationic poly(vinylamine).

Base gel properties

Water content / Equilibrium water content (EWC%)

Equilibrium water content is one of the most basic but also most important measurement for any hydrogel, in summary it measures the natural amount of water the gel will uptake if left to reach equilibrium.

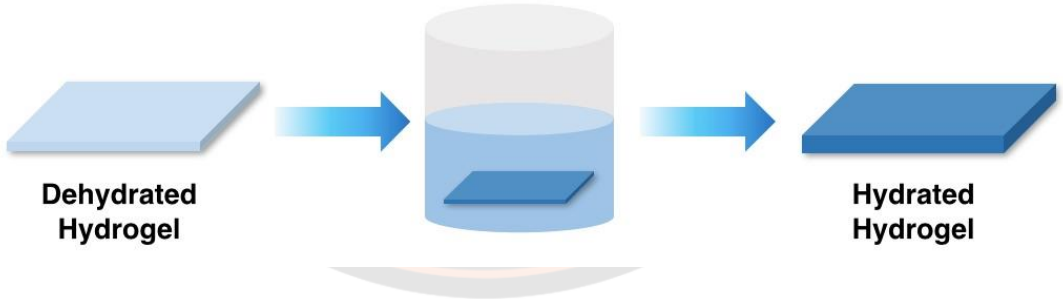


Figure 24 Schematic picture of hydrating hydrogels

The EWC% is measured by placing the gel into an abundant amount of distilled water for one week to allow the gel to reach full equilibrium, the water is changed for the first 5 days to remove any unreacted monomer. The gel is then weighed and placed into a microwave and weighed each time until a constant weight is recorded. The EWC% is then calculated using the following equation.

$$\text{EWC\%} = \frac{\text{Hydrated hydrogel (g)} - \text{Dehydrated hydrogel (g)}}{\text{hydrated hydrogel (g)}} \times 100 \qquad (1)$$

State of water

Differential scanning calorimetry (DSC) was used to understand the state of water in the hydrogels. How the water is structured in the gels governs the majority of the final materials properties. EWC gives us the amount of water present but as seen in the figure below, the water is present in two different states; freezing (free) and non-freezing (bound) water, the non-freezing water is normally split into two more categories (tightly and loosely bound water, which depend on the strength of the interactions between the polymer network and the water molecules.

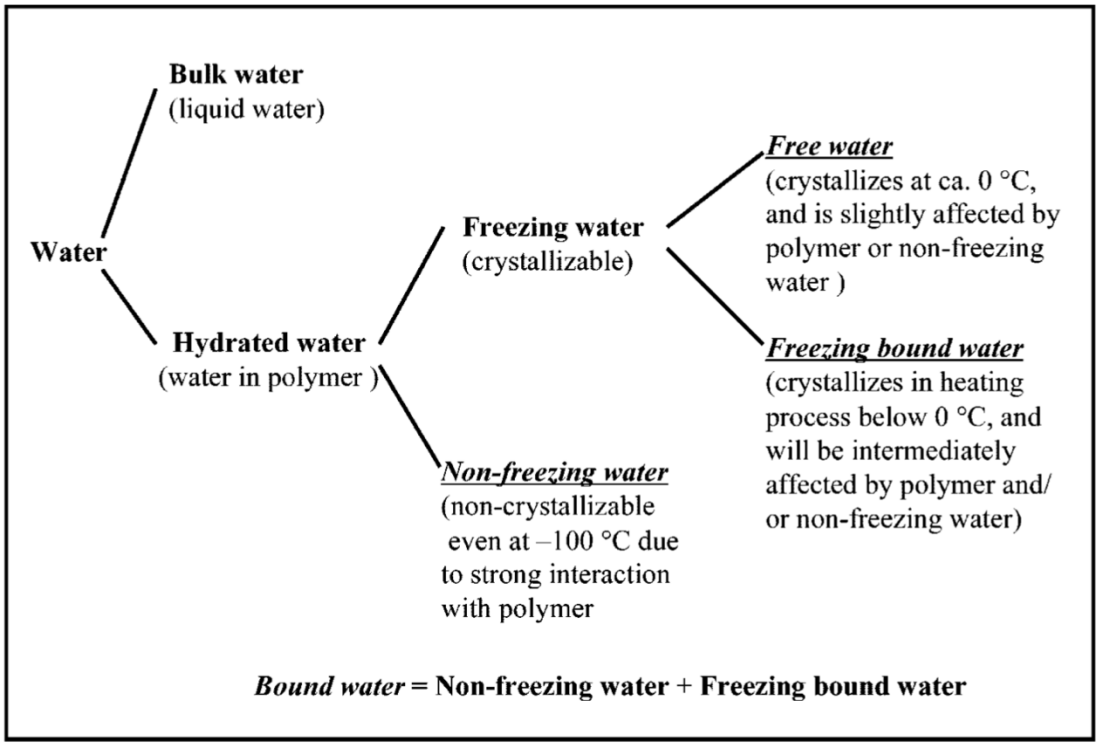


Figure 25 Classification of water structure in a hydrated polymer based on DSC analysis

Differential scanning calorimetry (DSC) will be used to calculate the percentage of freezing water in the hydrogels. Thermograms as seen in Figure 26 is

obtained using a Mettler Toledo Series DSC1 star, connected to a liquid nitrogen cooling unit.

In this wok, the hydrated hydrogels were cut and weighed about 10 mg and excess water carefully blotted with damp filter paper (to prevent absorption from the sample). The disc is then weighed and sealed in an aluminum pan to prevent water evaporation. The pan is then placed in the sample holder of the thermal analyzer.

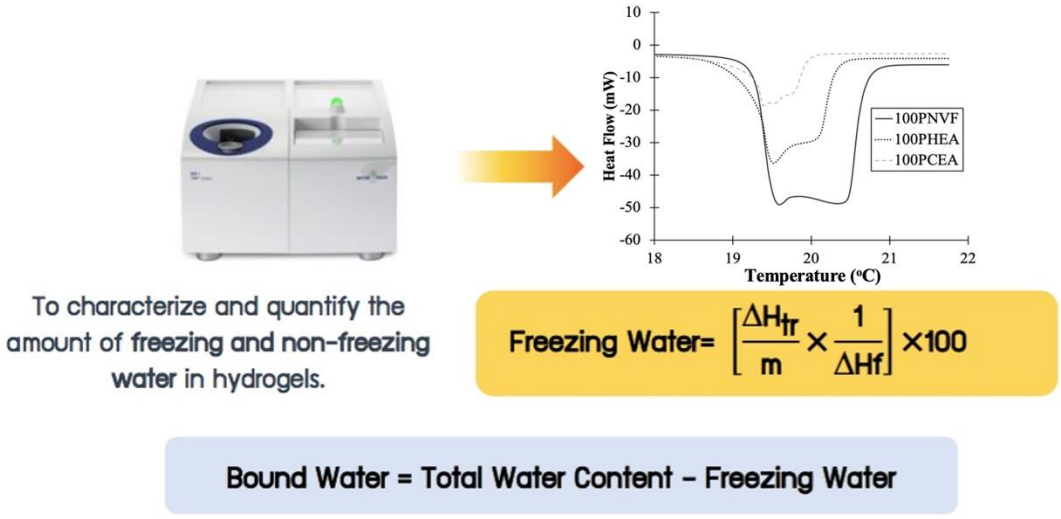


Figure 26 Schematic picture of information on using DSC for determining hydrogel water structure

The area under the endothermic peak(s) corresponds to the energy required to melt the freezing water in the sample. The weight of the sample is known, as is the energy required to melt 1g of pure water is known (333.5 Jg-1) and so using Equation **(2)** below, we can calculate the percentage of freezable (free) water in the sample, whereby:

$$\text{Free water (\%)} = [\Delta H_{Tr} / (m \times \Delta H_f)] \times 100 \quad \textbf{(2)}$$

Where: ΔH_f = 333.5 J/g

ΔH_{Tr} = heat of transition

m = sample weight (mg)

The amount of bound water was obtained by subtracting the amount of free water the total percent water content, whereby:

Bound water content (%) = EWC% – Free water content (%) **(3)**

Wettability

The wettability of the hydrated hydrogels surface was measured by contact angle from CA, Dataphysics Model OCA20 at room temperature. From figure 27, the hydrated hydrogel samples were cut in 2x6 cm^2 and adhered on the cover slide. A volume of 10.0 μL deionized water was dropped onto the surface sample with a 0.50 μL/s dispensing rate by micrometric syringe. The images of water droplets was taken by HD camera and measured the contact angle automatically (n=10).

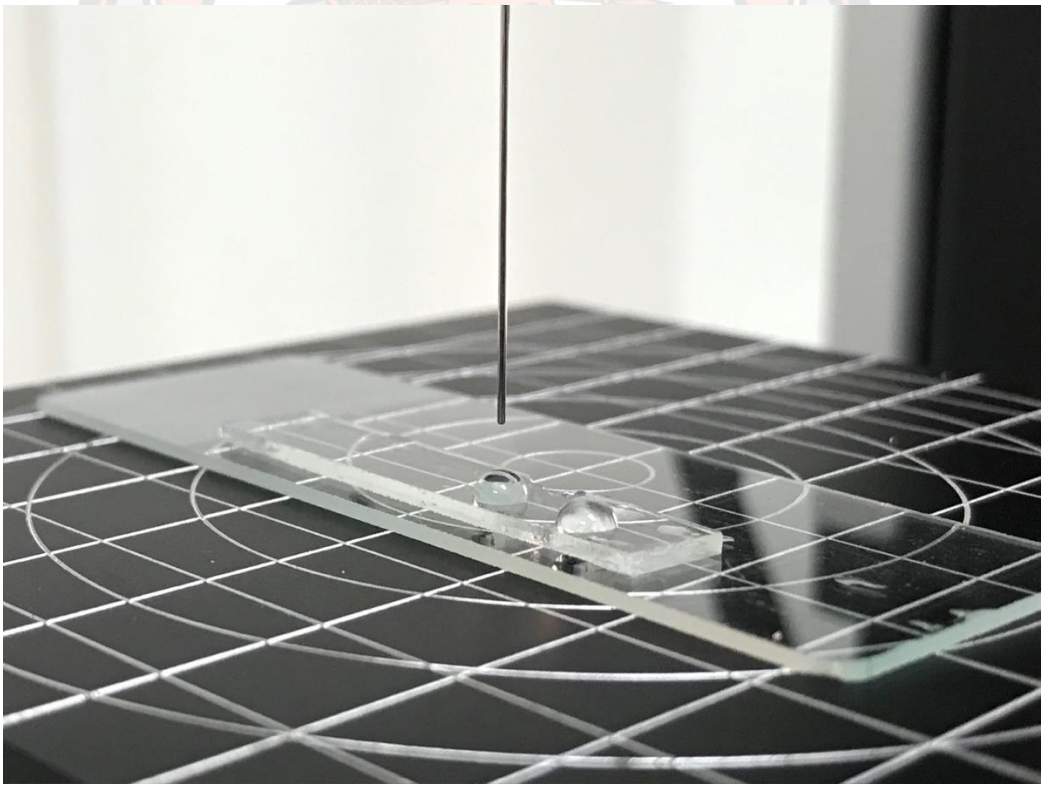


Figure 27 Electronic photograph of water dropped onto the surface hydrogels with micrometric syringe

Mechanical properties

Mechanical properties were determined by testing the hydrated hydrogels using Universal Testing Machine (INSTRON® CALIBRATION LAB model 5965) for measuring tensile properties of the hydrogel samples.

The tested hydrogel was cut out in rectangular shape into 20 mm x 60 mm (3 pieces per sample). The thickness of individual patch was the average value of three separate measurements taken along the middle 20 mm section of such hydrogel using micrometer. The hydrogel was clamped between two grips provided with a 100-N load cell before being pulled at the rate of 20 mm/min, measurement was continued until the samples were ruptured. In this experiment, three specimens were subjected to the test for one hydrogel sample formulation. The tensile strength at break, modulus and the percentage of elongation at break of hydrogels were reported.

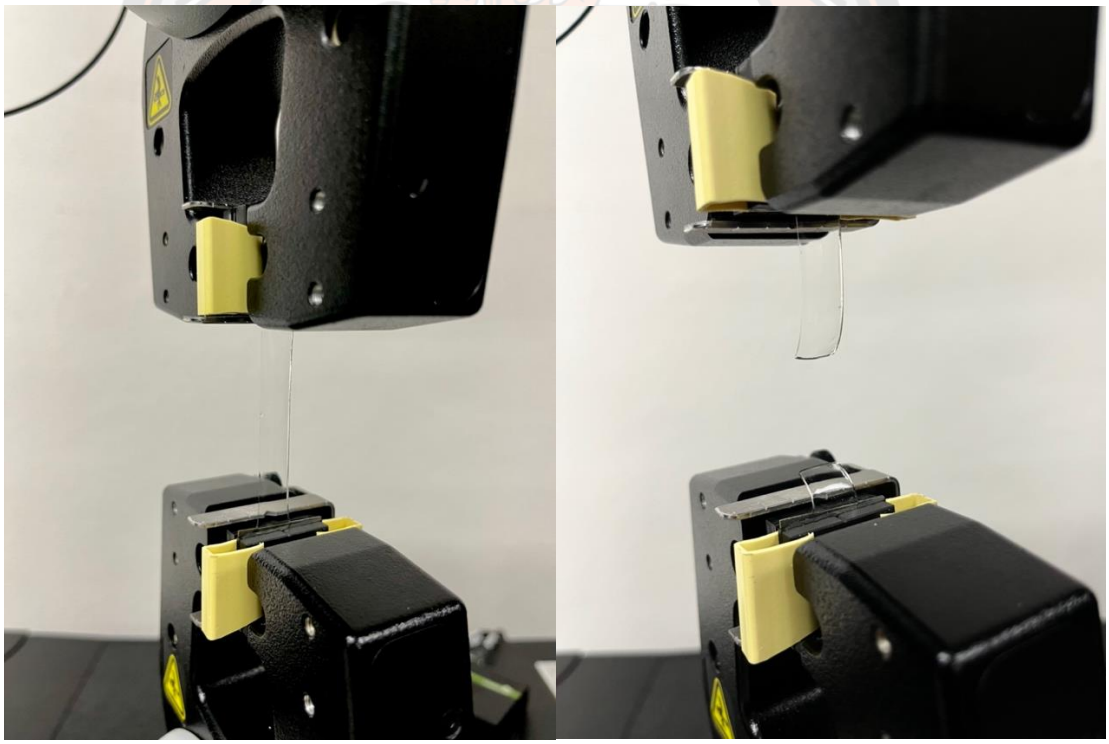


Figure 28 Electronic photograph of tensile testing of hydrogels before (left) and after (right) samples were ruptured

Applied gel properties

Network stability of hydrogel over time in water and ethanol

The stability of hydrogel over time in water and ethanol was measured according to conventional gravimetric methods. First cycle, the hydration hydrogels at full equilibrium were weighed (W_0) and immersed in 70% ethanol solution at room temperature for 24 hours and then weight (W_t) is recorded, following equation **(4)**. This studied determined time intervals and tested for 10 cycles observation is schematically in figure 29.

$$\text{Swelling ratio} = [(W_0 - W_t) / W_0] \times 100 \qquad \textbf{(4)}$$

Where: W_0 is the weight of the completely hydrated hydrogel and W_t is the weight of hydrogel after 24 hours.

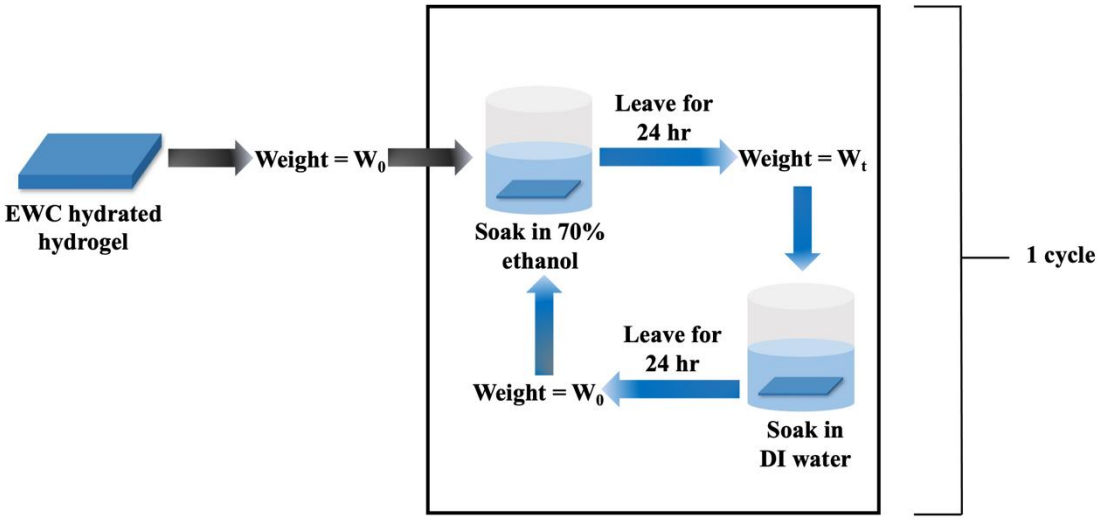


Figure 29 Schematic picture of stability of hydrogel over time in water and ethanol for 1 cycle observation

The swelling behavior as a function of pH

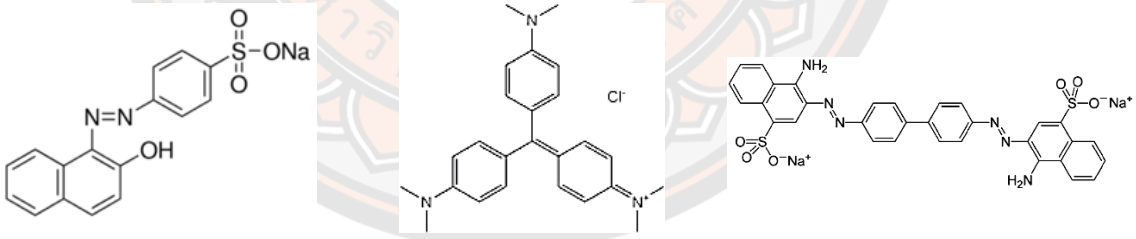
The swelling behavior as a function of pH of the hydrogel was observed at pH 2, 4 ,7, 10 and 12 at room temperature. First, three hydration hydrogels at full

equilibrium from each composition were weighed (W_0) were taken out at pre-determined time. Then, the hydrated hydrogel samples remained in pH solution for the time to equilibrium from 24 to 36 hours and then weight (W_t) is recorded, following equation **(4).**

In vitro study of dye release from the hydrogels

The objective of this project is to produce cosmetic hydrogels based on N-vinylformamide, the main reason for these gels is the uptake of active agents. A simple but effective test for this is to use organic dyes, we will use dyes with different structures, meaning dyes of different size and that contain different functional groups this will enable us to predict the amount of active released from the hydrogels and incorporating of model cosmetic ingredients-loaded hydrogel was studied.

For this work, three organic dyes were selected all with different characteristics: Orange II sodium salt azo dye (anionic - pH dependent), Crystal violet azo dye (cationic - pH dependent) and Congo red azo dye (Zwitterion - pH dependent). The details information of the study on release profile is described in the following sections.



Orange II sodium salt Crystal violet Congo red

Figure 30 Structure of dyes

1. **Preparation of dye stock solution for standard calibration curve**

Organic dyes and some active agents have very similar structures; therefore, this method always is also a good method for understanding active delivery. For this work, the first step for this method is to create standards for each organic dye, this is done by dissolving a known concentration of dye ($1x10^{-2}$ to $1x10^{-6}$ M) in deionized

water and then diluting to generate a series of solutions with known concentrations this are then scanned at the optimum UV wavelength. The optimum UV wavelength is determined from doing a wavelength sweep and detecting at which wavelength. The Microplate reader (Biotek, Model Synergy H1 Hybrid Reader), which is a UV/Visible plate reader, was used to observe the absorbance of dye solution. The results are plotted of absorbance versus concentration to produce a calibration curve for each dye. Once the calibration standard for each dye has been made uptake and release of the hydrogels can be assessed.

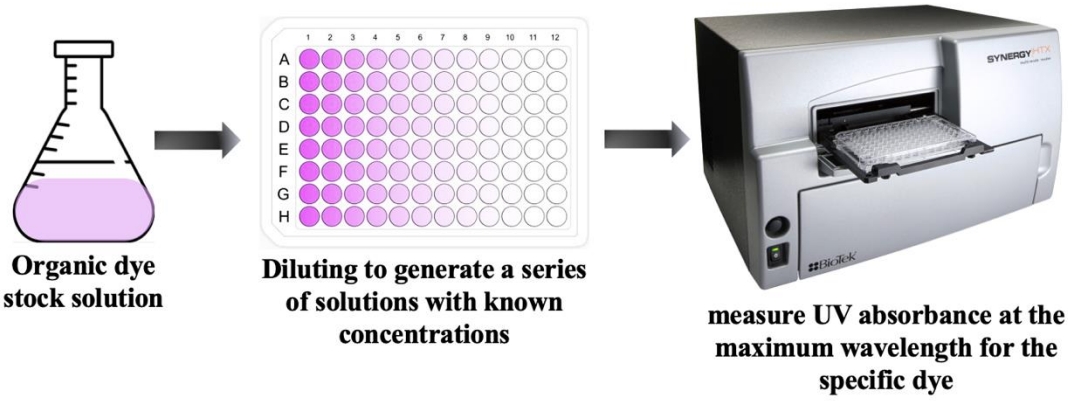


Figure 31 Schematic picture of preparation of dye stock solution for standard calibration curve

1. **Uptake and release studies**

For uptake and release studies three organic dyes were used: Orange II sodium salt dye, Crystal violet dye and Congo red dye. Dye uptake was carried out by immersing hydrated hydrogel samples into a solution containing a known concentration of dye. Each hydrogel was weighed and then soaked in 1.0 ml of dye solution with a 0.0001 M concentration of dye for 96 hours in order to give time for the system to reach equilibrium before testing the dye release.

For the release study, each sample used the following method. After the uptake period (4 days) the hydrogels with the absorbed dye were placed into a new clean vial, which contained 1 ml of DI water. After transferring the hydrogel, the vial was vortexed using a Vortex Mixer GENIE 2 (Model G560E) at 5,000 rpm for 15

seconds. The gel was then left in the solution for a set time. The time periods were six, 60-minute periods, which combined to make a total release time of 6 hrs. After each time period the solution was removed and transferred to a 96 well-plate, and fresh media (1 ml DI water) was placed into the vial for the next time period. The amount of dye released was determine by the UV absorbance of the solution measured using a multiplate reader (Synergy H1 Multi-Mode Reader, Detection mode UV-visible absorbance). When using Orange II sodium salt a wavelength of 480 nm was used, Crystal Violet a wavelength of 590 nm, and Congo red a wavelength 495 nm. The absorbance value at the specific wavelength for the given dye was then converted into μg/ml using calibration curves of the appropriate dye.

Part 3 : In vivo clinic / Specific testing of Applied gel properties

In this *in vivo* clinic test section, therefore, a total of thirty healthy female (aged 20–55 years) volunteers with normal skin properties was included in this study, their skin characteristics around the forearm area were evaluated for a single application of the hydrogels tested, followed by the skin adhesion, skin irritation and Self-perception Questionnaire (SPQ) after application of hydrogel patch. The detail information of the techniques is described in the following sections.

1. Skin adhesion ability after application of hydrogel patch

Each square patch of 5x5 cm^2, which had been controlled to have similarities in thickness and size, was manually attached to a volunteer's forearm. The ability of patch adhesion was evaluated from the duration for the adhesion of the patch to the forearm of the volunteer for 30 minutes. During the studies, all volunteers were allowed for normal activity. Figure 32 shows the electronic photograph of patch adhered to the volunteer's forearm.

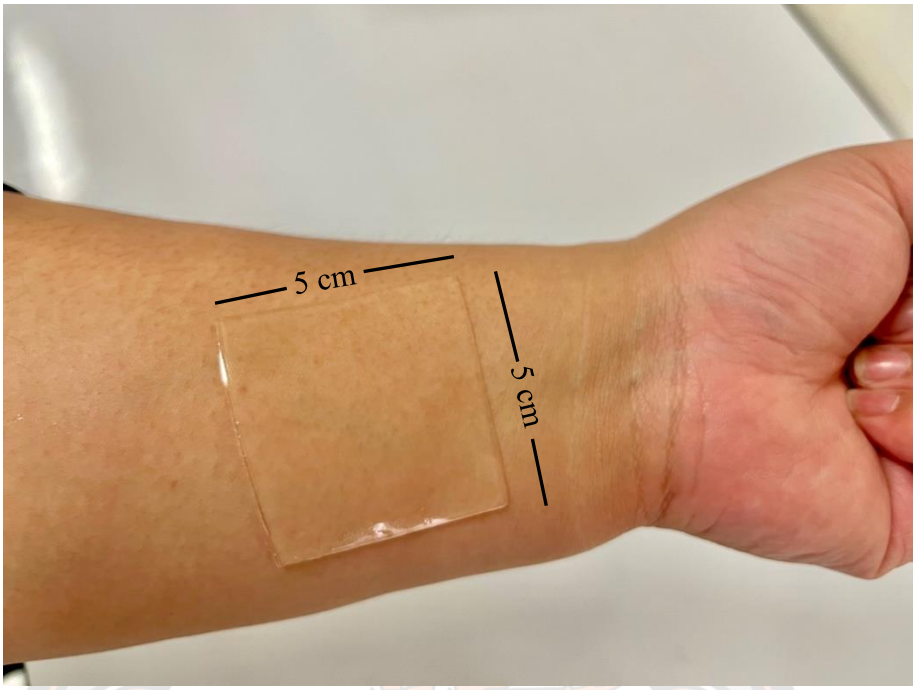


Figure 32 Electronic photograph of patch adhered to the volunteer's forearm on skin adhesion ability testing

2. skin irritation after application of hydrogel patch

After observed for sensitivity by bio-adhesion test, the next step will determine the safety of the novel cosmetics hydrogel patch, all volunteers were determined the safety of the developed hydrogel patch, a preliminary study assessing the irritation effects of the product after single application was designed and conducted.

Each square patch of 5x5 cm^2 was slightly pressed to a volunteer's forearm. The duration of application was designed for 30 min. The skin properties were evaluated by biophysical instruments and a dermatologist at 0 (T0), 30 (T30), 60 (T60), 120 (T120) and 240 (T240) minute after removal of the patch, their skin characteristics around the testing area were taken a photo.

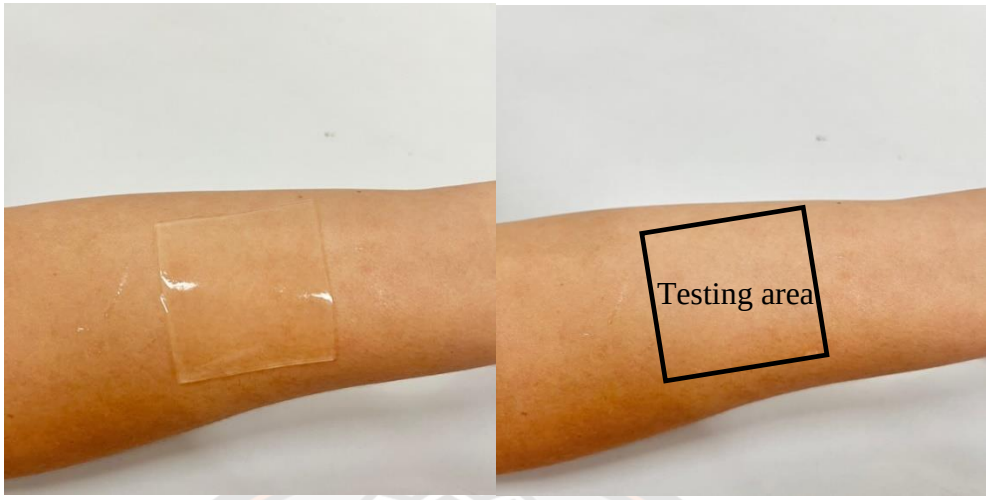


Figure 33 Electronic photograph of the volunteer's forearm for evaluation of skin irritation procedure before (left) and after (right) one application

3. Self-perception Questionnaire after application of hydrogel patch

The final part of the study, the subjects were assessed for their skin appearance and were asked to complete a self-perception Questionnaire (SPQ) at beginning of the trial (T_0) and after the 30 minutes test product application (T_1) of hydrogel patch test. The detail of the Self-Perception Questionnaire form was reported which, more relaxed, more cooled, more hydrated, softer and adhesive hydrogels after 30 minutes applied the hydrogel patches. Combining the level of score (1 is strongly disagree, 2 is Disagree, 3 is neutral, 4 is Agree and 5 is Strongly agree).

Questionnaire for apply product

Number.....Name ..... Phone .....

Code .....

Sexual

☐ male ☐ female

Age (years)

☐ below 20 ☐ 20-30 ☐ 31-40 ☐ above 40

Have you ever used mask hydrogels product?

☐ Yes ☐ No

How do/did you choose your product?

☐ Friend / relative recommendation ☐ Brand

☐ Dermatologist advice ☐ Pricing

☐ Product's ingredients

Details	Level of score				
	5	4	3	2	1
1. More Relaxed					
2. More Cooled					
3. More hydrated					
4. Softer					
5. More adhesive					

Note:

1 is strongly disagree

2 is Disagree

3 is neutral

4 is Agree

5 is Strongly agree

Figure 34 Example of the Self-Perception Questionnaire form of the study subjects after one application (T30)

CHAPTER IV

RESULTS AND DISCUSSION

This chapter presents the results and discussion of this research, which is divided into two approaches. The first approach involves the fabrication of poly(N-vinylformamide), PNVF-based materials through photopolymerization to create novel cosmetic hydrogel patches. The second approach is the synthesis of poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine) by partially hydrolyzing the amide group of poly(NVF-co-HEA) under acidic conditions.

For the first approach, the aim was to fabricate novel cosmetic hydrogel patches by first characterizing commercial cosmetic products that had been classified based on gel matrix components for their base gel properties. This information was then used to guide the selection of the most suitable properties needed for cosmetic hydrogels. The study evaluated the impact of varying the ratio of ingredients in the formula for producing PNVF materials and NVF-copolymer hydrogels. The base gel properties such as equilibrium water content (EWC), state of water, wettability, and mechanical properties of the hydrogels were observed to determine the optimal composition for the novel cosmetic hydrogel patches. The applied properties, such as skin adhesion and skin irritation, were also studied, along with self-perception questionnaires to determine the suitability of the cosmetic hydrogel patches.

For the second approach, the study compares the effect of poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine), PVAm, after performing acid hydrolysis on poly(NVF-co-HEA) hydrogels. The analysis covers equilibrium water content (EWC), state of water, wettability, swelling behavior as a function of pH and mechanical properties of the hydrogels. The results of pH-responsive release provide an understanding of the active delivery properties and help identify the best conductive cosmetic active ingredients.

Approach 1: Fabrication of poly(N-vinylformamide; PNVF)-based materials for novel cosmetic hydrogel patches

The first approach involves the fabrication of poly(N-vinylformamide; PNVF)-based materials through photopolymerization to create novel cosmetic hydrogel patches. This approach presents the results and discussion, which will be divided into the two parts:

1. Study and evaluation of the commercial cosmetic products
2. Poly(N-vinylformamide) based novel cosmetic hydrogel patches

Part 1: Study and evaluation of the commercial cosmetic products

The commercial cosmetic products considered in this study are: Faith in face, Medius heart and Beauty formulas gold eye gel patches. The commercial cosmetic products have been specifically formulated with water soluble bio matrix and are classified based on gel matrix and all products are formulated as shown in table 3. The gel matrix contained in the hydrogel eye patches mask tested in this study (Dimethicone ceratonia siliqua gum, cellulose gum, Ceratonia siliqua Gum and Cetearyl Alcohol) were all selected for their gelling agents. These formulations produce a hydrogel mask that has good physical stability and consistency. One point of interest is that all of the commercial cosmetic products do not contain a separate chemical crosslinking agent, and therefore when immersed in water all dissolve.

Table 3 Commercial cosmetics hydrogel eye patches - list of ingredients

Products	Ingredients	Used
Faith in face eye am not tired 	Water	Solution
	Butylene glycol	Humectant
	Glycerin	Humectant
	Diethoxyethyl succinate	Emollient ester
	Citrus paradisi (grapefruit) fruit extract	Active Ingredient

	Dimethicone ceratonia siliqua gum	**Gelling agent**
	Chondrus crispus (carrageenan)	Thickening agent
	Cetyl ethylhexanoate	Moisturizing agent
	Linum usitatissmum (Linseed) seed extract	Active Ingredient
	Euphrasia officinalis extract	Active Ingredient
	Brassica oleracea italica (broccoii) extract	Active Ingredient
	Juglans regla (walnut) seed extract	Active Ingredient
	Glyceryl stearate	Thickening agent
	Cellulose gum	**Gelling agent**
	Polyacrylate-13 polyisobutene	Thickening agent
	Polysorbate 20	Surfactant
	Disodium EDTA	Preservative
	Phenoxyethanol	Preservative
	Fragrance	Fragrance
	CI 17200 (Lavanya Hebe)	Colour
The Medius heart Ppyoung 	Water	Water
	Dipropylene Glycol	Texturizer
	Glycerin	Humectant
	caprylic/capric	Solvent
	Niacinamide	Active ingredient
	Dimethicone	Humectant
	Ceratonia siliqua Gum	**Gelling agent**
	Chondrus crispus powder	Active ingredient
	Trehalose	Active ingredient
	Betaine	Surfactant
	Polysorbate60	Surfactant
	Butylene glycol	Humectant
	Cellulose gum	**Gelling agent**

Paeonia suffruticosa root extract	Active ingredient
Centella asiatica extract	Active ingredient
1,2-hexanediol	Solvent
Potassium chloride	
Sodium polyacrylate	Thickening agent
Chamomille recutita (matricaria) flower extract	Active ingredient
Hydrogenated polydecene	Emollient ester
Glyceryl caprylate	Solvent
Glyceryl stearate	Surfactant
PEG-11 stearate	Emulsifier
Panthenol	Humectant
Sucrose	Emulsifier
Adenosine	Active ingredient
Ethylhexylglycerin	Texture agent
CI77491	Colour
Red Iron Oxide/Lavanya Laal	
Trideceth-6	Emulsifier
Hydrolyzed collagen	Humectant
Theobroma cacao (cocoa) extract	Active ingredient
Sodium hyaluronate	Active ingredient
Saccharum officinarum (sugarcane) extract	Active ingredient
Octyldodecanol	Texture agent
Pantolactone	Humectant
Ascorbic acid	Active ingredient
Citrus limon (Lemon) fruit extract	Active ingredient
Hydrogenated lecithin	Active ingredient
Isopropyl titanium triisostearate	Texture agent
Pearl extract	Active ingredient

	Ceramide NP	Active ingredient
	Caprytyl glycol	Preservative
	Disodium EDTA	Preservative
	Phenoxyethanol	Preservative
	Fragrance	fragrance
Beauty formulas reviving gold eye gel patches 	Aqua	Water
	Glycerin	Humectant
	Dimethicone	Humectant
	Paraffinum Liquidum	Solvent
	Cetearyl Alcohol	**Gelling agent**
	Isopropyl Palmitate	Texture agent
	Propylene Glycol	Moisturizing agent
	Sorbitan Stearate	Emulsifier
	Hydrolyzed Collagen	Humectant
	Phenoxyethanol	Preservative
	Lavandula Angustifolia (Lavender) Extract	Active ingredient
	Mica	Texture agent
	CI 77891 (Titanium dioxide)	Colour
	CI 77491	Colour

Characterizations of the commercial cosmetic products on base gel properties

The commercial cosmetic products were applied onto the forearm following the manufacturer's instructions, for a maximum of 30 min before being removed and then the commercial cosmetic products were determined visually for physical properties. The water properties of hydrogels on the structure are very important to study because of the water in hydrogels is related how the hydrogels behave in many applications. The water in the gel was measured as water content (%WC).

Table 4 The water content of the commercial cosmetic products

Test Product	% water content
Faith in face	86.59±4.3
Medius heart	84.97±4.2
Beauty formulas	94.89±4.7

Values are mean ± SD (n=3)

The water content (%WC) of each commercial cosmetic products were determined and shown in table 4. The results showed that %WC of the following products, Faith in face, Medius heart and Beauty formulas had %WC of 86.59%, 84.97% and 94.89%, respectively. The highest %WC was 94.89% when using the Cetearyl Alcohol and Isopropyl Palmitate as the copolymer which allowed of largest amount of water the hydrogels. However, all the products can be claimed to be high %WC (> 84%).

Table 5 The wettability of the commercial cosmetic products

Products	Contact angle (A^O)
Faith in face	5.1±0.2
Medius heart	2.4±0.1
Beauty formulas	1.0±0.1

Values are mean ± SD (n=10)

To further compare the surface of the hydrated hydrogels, the hydrophilicity of each sample was assessed using contact angle measurements. Photos were taken to measure the contact angle, and the results are presented in Table 5. The Faith in face, Medius heart and Beauty formulas is 5.1, 2.4 and 1.0, respectively. It can be observed that all of the commercial cosmetic products have very low contact angles when a liquid is placed on their solid surface. Importantly, all of these values are below 45 degrees, meaning that all of the hydrogel samples are considered very hydrophilic in terms of their surface properties.

Good physical characteristics are needed for ease of use in term of applications. All of the commercial cosmetic products were characterized for their tensile properties and reported in terms of percent elongation at break (%), tensile strength (KPa) and modulus (KPa), as shown in table 6. This table contains the data of commercial cosmetic products, tensile strength, percent elongation at break and modulus.

Table 6 Tensile properties of commercial cosmetic products

Product	Tensile strength (KPa)	Percent elongation at break (%)	Modulus (KPa)
Faith in face	914.85±45.7	83.90±4.5	1126.20±5.8
Medius heart	174.49±8.7	232.70±11.6	97.68±4.9
Beauty formulas	184.17±9.2	252.64±12.6	72.90±3.6

Values are mean ± SD (n=3)

From table 6, tensile strength at break (KPa) and percent elongation at break (%) of all commercial cosmetic products were not significantly different. Therefore, the Faith in face gave the highest of tensile strength and modulus value are 914 KPa and 1126.20 KPa, respectively, due to these commercial cosmetic products having the fiber blended into their composition.

Self-Perception Questionnaires

The product testing was not only physical testing but also included a Self-perception Questionnaire after application. The subjects were assessed via their skin after application onto the forearm and were asked to complete a Self-perception Questionnaire (SPQ) after 30 minutes single application. The detail of the Self-Perception Questionnaire form was reported as, more relaxed, more cooled, more hydrated, softer and adhesive hydrogels after 30 minutes application of the hydrogel patch. Combining the level of score (1 is strongly disagree, 2 is Disagree, 3 is neutral, 4 is Agree and 5 is Strongly agree).

Questionnaire for apply product

Number.....Name ..... Phone .....

Code .....

Sexual

☐ male ☐ female

Age (years)

☐ below 20 ☐ 20-30 ☐ 31-40 ☐ above 40

Have you ever used mask hydrogels product?

☐ Yes ☐ No

How do/did you choose your product?

☐ Friend / relative recommendation ☐ Brand

☐ Dermatologist advice ☐ Pricing

☐ Product's ingredients

Details	Level of score				
	5	4	3	2	1
1. More Relaxed					
2. More Cooled					
3. More hydrated					
4. Softer					
5. More adhesive					

Note:

1 is strongly disagree

2 is Disagree

3 is neutral

4 is Agree

5 is Strongly agree

Figure 35 The Self-Perception Questionnaire form of the study subjects after one application (T30)

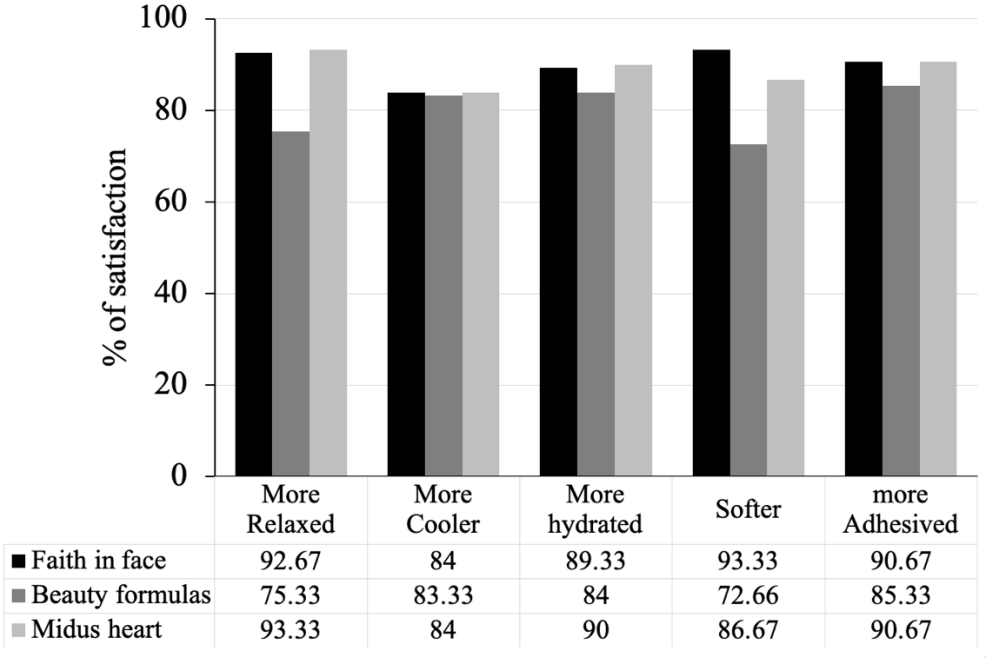
Thirty female healthy volunteers, aged between 20 and 40 years old were enrolled and completed the study, with variable skin characteristics of skin type. Most of the volunteers presented a mixed hydration skin type (Table 7).

Table 7 Analysis of the skin type of the study subjects, based on the subjects' self-assessment of skin hydration

Skin type		**%**
Hydration	Oily	5.1
	Normal	31.2
	Dye	10.9
	Mixed	52.8

The results were confirmed by the subjects' perception as reported in the Self-perception Questionnaire (SPQ). Combining the level of score (1 is strongly disagree, 2 is Disagree, 3 is neutral, 4 is Agree and 5 is Strongly agree). Figure 36 shows Self-Perception Questionnaire responses of the study subjects after one application of the commercial cosmetic products.

(a)



	More Relaxed	More Cooler	More hydrated	Softer	more Adhesived
Faith in face	92.67	84	89.33	93.33	90.67
Beauty formulas	75.33	83.33	84	72.66	85.33
Midus heart	93.33	84	90	86.67	90.67

(b)

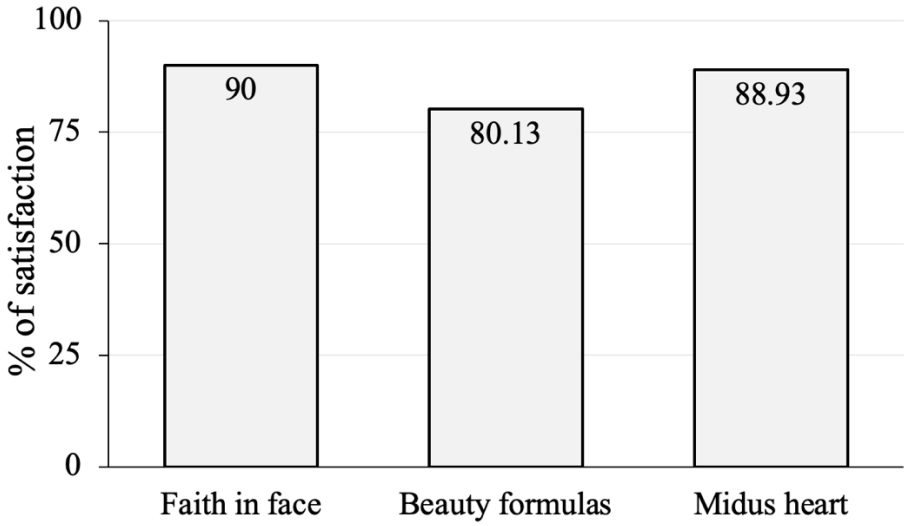


Figure 36 Self-Perception Questionnaire responses of the study subjects after one application of the commercial cosmetic products; (a) each detail and (b) the overall of satisfaction

From figure 36(b), Faith in face gave the highest score of overall of %satisfaction from the study subjects after one application (T30) at 90%. The study

subjects felt their skin was more relaxed after treatment with a score of 92.67%. They also felt their skin was more cooled (84%), more hydrated (89.33%), softer (93.33%) and the gel had good adhesion (90.67%). Moreover, Beauty formulas gave the lowest score of overall of %satisfaction of 80.13. The study subjects who felt their skin was more relaxed after treatment was 75.33% and also felt their skin was more cooled (83.33%), more hydrated (84%), softer (72.66%) and the gel had good adhesion (85.33%). To maintain a healthy skin, it is important to keep the skin hydrated, for this test it was only a single treatment; however, prolonged use of the commercial cosmetic products promotes should result in long term improvements.

Part 2: Poly(N-vinylformamide) based novel cosmetic hydrogel patches

In order to obtain cosmetic hydrogels that had the best properties for the desired application was studied the effect of varying the composition on the production of PNVF-based materials and its copolymer hydrogels.

In general, the preparation of hydrogels depends on several factor such as initiator system, monomer, crosslinker type and crosslink density to control the hydrogel polymerization and the final hydrogels properties. In this work, the fundamental hydrogel properties or "base gel" properties such as equilibrium water content (EWC), state of water, wettability and mechanical properties of hydrogels were first assessed to find the best composition for novel cosmetic hydrogels. Additional characterization also included the uptake and release ability of active agents and *in vivo* clinic; skin adhesion and skin irritation and Self-perception Questionnaire (SPQ) after application of the hydrogel patches. These characterization methods are more closely related to the application use and are referred to as the applied properties. This part is presented in three sections:

1. **Preparation of poly(NVF)-copolymer hydrogels**
2. **Development of hydrogels for novel cosmetic hydrogel patches application**
3. **Comparison of the novel cosmetic hydrogel patches with the commercial cosmetic products**

Preparation of poly(NVF)-copolymer hydrogels

1. **Effect of initiation system**

The majority of synthetic hydrogels are produced via free-radical polymerization. This means they require some initiation process; in industry this is normally a thermal process. However, other initiation methods are available, this work investigates and compares thermal and photo-initiation. The photo-initiation is divided further depending on the light source (Mercury Arc UV and UV-LEDs). UV-LEDs can be acquired that have a wavelength of ~395nm with a very narrow band (+/- 20mn) this is a significant difference when compared to mercury Arc UV light sources which cover a board range of the electromagnetic spectrum.

For this purpose, hydrogels were synthesized using Poly(ethylene glycol) diacrylate; PEGDA as a crosslinker with hydrogels of 2-hydroxyethylmethacrylate (HEMA)-co-methacrylic acid (MAA) via free-radical polymerization via three systems; (1) ammonium persulfate was used as a thermal-initiator for thermal-initiation system, (2) 2-Hydroxy-2-methylpropiophenone and (3) Diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide was used as a photo-initiator for mercury Arc UV (UVA) and UV-LEDs initiation system, respectively. So, three different hydrogel samples were prepared and characterized.

Table 8 The composition of PHEMA/MAA hydrogels via free-radical polymerization for three different initial processes (Thermal, UVA and UV-LEDs)

Methods	**Monomer (%w/w)**		**Cross-linker (%w/w)**	**Initiator (%w/w)**	**Time (min)**
	HEMA	**MAA**			
Thermal-initiation	98	2	3	1	1,440
UVA-initiation	98	2	3	1	15
UV-LEDs initiation	98	2	3	1	3

PHEMA/MAA hydrogels were successful formed via free-radical polymerization depending on the initiation process. From Table 8, the biggest initial difference between thermal- and photopolymerization was reaction time, UVA and UV-LEDs initiation was polymerized within 15 and 3 minutes, respectively. Whereas, Thermal-initiation was synthesized by overnight polymerization (24 hours).

In order to evaluate the effect of the initiation system on water content, wettability, and mechanical properties, the synthesized hydrogels were investigated. The water content of hydrogels shows the amount of water in the hydrogels after allowing the gels enough time to reach equilibrium. In this work, PHEMA/MAA hydrogels were used, which are two hydrophilic polymers.

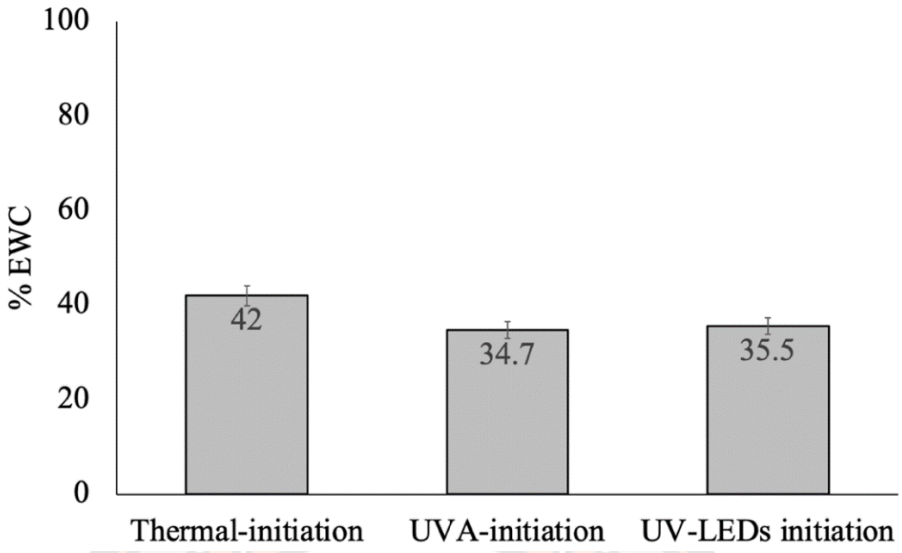
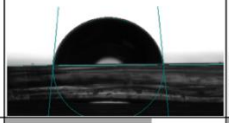
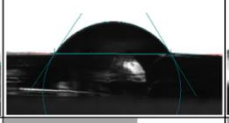
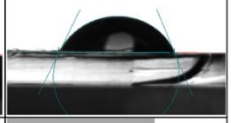
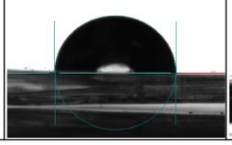
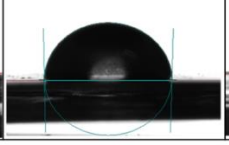
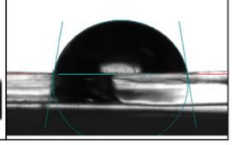


Figure 37 The %EWC of PHEMA/MAA hydrogels with different initiation processes

From figure 37, Thermal-initiation processing has the highest water content of the hydrogels studied, although the difference is not significant. Thermal initiation is also a longer process which allows more time for cross-linking and therefore, a slightly difference 3D network when compared to photo-initiation. There also will be differences in polymer chain length and molecular weight distribution.

Table 9 Contact angles of hydrated and dehydrated PHEMA/MAA hydrogels with different initiation processes

	Thermal-initiation	**UVA-initiation**	**UV-LEDs-initiation**
Hydrated hydrogels	84.7°± 4.09 	55.2°± 7.61 	65.8°± 3.83 
Dehydrated hydrogels	90.2°± 2.84 	91.4°± 8.0 	80.8°± 4.47 

The contact angle of hydrated and dehydrated PHEMA/MAA hydrogels was observed of the three different initiation processes. From Table 9, the dehydrated hydrogels show more similar contact angles than the hydrated hydrogels, with Thermal processing giving the highest contact angle. For this result the differences are significant with the UV-LEDs hydrogels presenting considerably lower contact angles for both hydrated and dehydrated gels.

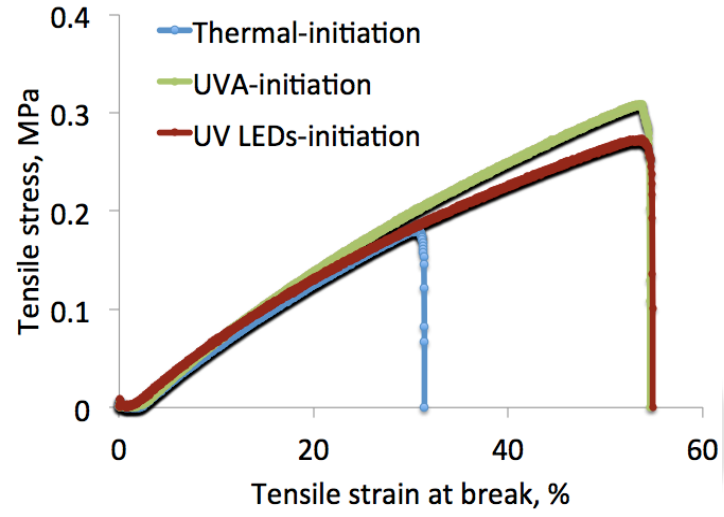


Figure 38 The stress-strain curves of PHEMA/MAA hydrogels with different initiation processes

Tensile properties of PHEMA/MAA hydrogels were compared, and the stress-strain curves plotted in figure 38. The results show tensile stress at break for hydrogels, thermally initiated samples gave 50% lower tensile stress than hydrogels UVA and UV-LEDs initiated. The Young's modulus of all three initiation systems gave very similar values. The polymerization time can affect the network structure, resulting in varying polymer chain lengths and crosslinker density. The elongating chain are randomly crosslinked by crosslinker that is influenced by the time of polymerization and initiation process used.

Time of polymerization for each process significantly effects the electric cost and also the energy used (watts). Therefore, the final part of this section calculated the energy usage of the three initiator systems (thermal, UVA and UV-LEDs). Figure 39

shows the comparison of the systems in terms of power usage (watts) and electric cost (baht) per 100 hydrogels.

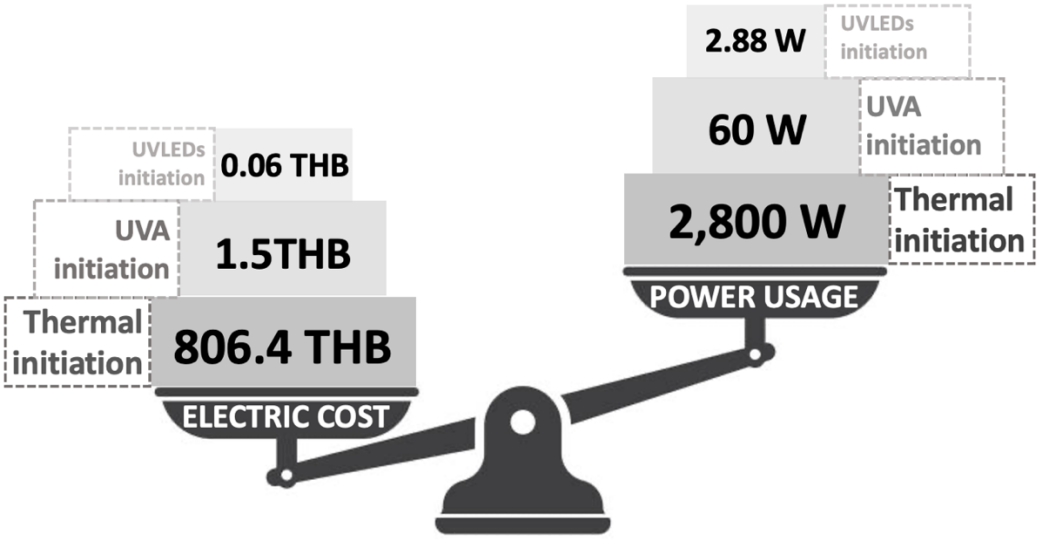


Figure 39 The comparison of initiation process in term of power usage (watts) and electric cost (baht) per 100 hydrogels

Figure 39 compares the initiation process of hydrogels in terms of electric cost and power usage. The electric cost of 100 hydrogels showed that thermal initiation, UV-A initiation and UV-LEDs initiation is 806.4, 1.5 and 0.06 Baht, respectively. The highest electric cost of thermal initiation was 806.4 Baht when using 2,800 watts of power usage due to the longer time of polymerization in these hydrogels. Whereas UV-LEDs initiation process gave the best results by far in terms of power usage (watts) and electric cost (Baht).

Table 10 Wettability, tensile strain at break, electric cost, processing time and environmentally friendliness of the three initiation processes

	Thermal-initiation system	**UVA-initiation system**	**UVLEDs-initiation system**
Good wettability	yes	yes	yes
Tensile strain at break	poor	good	good
Electric Cost	High → Low		
Processing Time	High → Low		
Environmentally friendly	LOW	LOW-MID	HIGH

The aim of this study was to compare the effect of the initiation system on physical properties of the hydrogels. Table 10 shows the summary of the physical properties of PHEMA/MAA hydrogels prepared using three different initiation systems. Both photo-initiation methods gave very similar results for the EWC, mechanical strength and contact angle. There were some differences with thermally cure hydrogels due to the length of time of polymerization that is required for this process. Interesting in terms of cost and environmental concerns UV-LEDs gave the best results by far in terms of energy usage and use of toxic materials. This is the newest method of initiation and can be used to produce hydrogels at a fraction of the cost to both the environment and industry compared to thermally initiated hydrogels.

2. Effect of copolymer structure on hydrogels synthesis

Poly(N-vinylformamide) (PNVF) gels are chemically related to poly(N-vinyl pyrrolidone) (PNVP) gels, another important biomedical hydrogel, widely used in contact lenses, drug delivery systems, wound dressings and cosmetics. This makes NVF based hydrogels of interest, however, to even greater control the final hydrogels performance, NVF was copolymerized with two other co-monomers, N-hydroxyethyl acrylamide (HEA) and 2-carboxyethyl acrylate (CEA).

This work was concerned with the investigation of a series of Poly(N-vinylformamide) (PNVF) and two hydrophilic comonomers. Therefore, to enhance the properties, we become interested in N-hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl acrylate (CEA). They are two differences between HEA and CEA, one is the end group, which is -OH and -COOH, respectively, and also HEA contains an amide group, as shown in Figure 40. However, CEA is less polar than HEA due to the presence of two highly electronegative oxygen atoms covalently bonded to the same carbon atom in the carboxylic acid group. As a result, the dipole moment of each C-O bond is oriented towards the oxygen group. In addition, they are typically more hydrophilic than nonfunctional monomers like methyl methacrylate (MAA). As higher molecular weight vinyl acid monomers, they contain an ester group and can be covalently crosslinked to optimize the mechanical properties of NVF-based hydrogel performance.

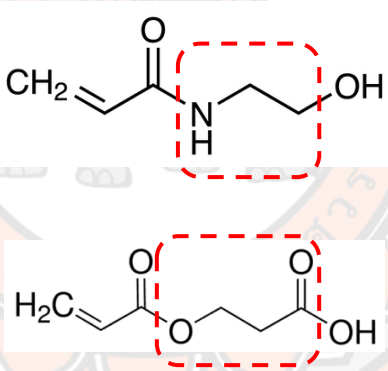


Figure 40 Comparison of the molecular structure of N-hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl acrylate (CEA)

To evaluate the effect of copolymer structure on the produced hydrogels, the base gel properties which determined including water content, water state, wettability, and dye uptake and release ability of the hydrogels. Table 11 shows the compositions of PNVF-copolymer hydrogels investigated, the method used to produce the gels was photo-polymerization using a UVLED light source, which has high throughput and short processing time. This is the major advantage of this method with the ability to

simply and effective scale the fabrication of the hydrogels from batch to continuous production on an industrial scale easily achieved.

Table 11 Formulations of a series of PolyN-vinylformamide (PNVF) and N-hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl Acrylate (CEA) hydrogels

Sample code	Monomer (%w/w)			Cross-linker (%w/w of monomer)	Photo-initiator (%w/w)
	NVF	HEA	CEA		
Homopolymer					
100PNVF	100	-	-	3	1
100PHEA	-	100	-	3	1
100PCEA	-	-	100	3	1
Copolymer					
75PNVF/25PHEA	75	25	-	3	1
50PNVF/50PHEA	50	50	-	3	1
75PNVF/25PCEA	75	-	25	3	1
50PNVF/50PHEA	50	-	50	3	1

For this work, UVLEDs photopolymerization was used to produce PNVF and copolymer hydrogels. N-vinylformamide (NVF) was used as the main monomer, N-Hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl Acrylate (CEA) were used as the co-monomers, Poly(ethylene glycol) diacrylate (PEGDA575) and Diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide was used as a cross-linker and photo-initiator, respectively. The physical appearance of poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogels, in both dehydrated and hydrated forms are shown in figure 41.

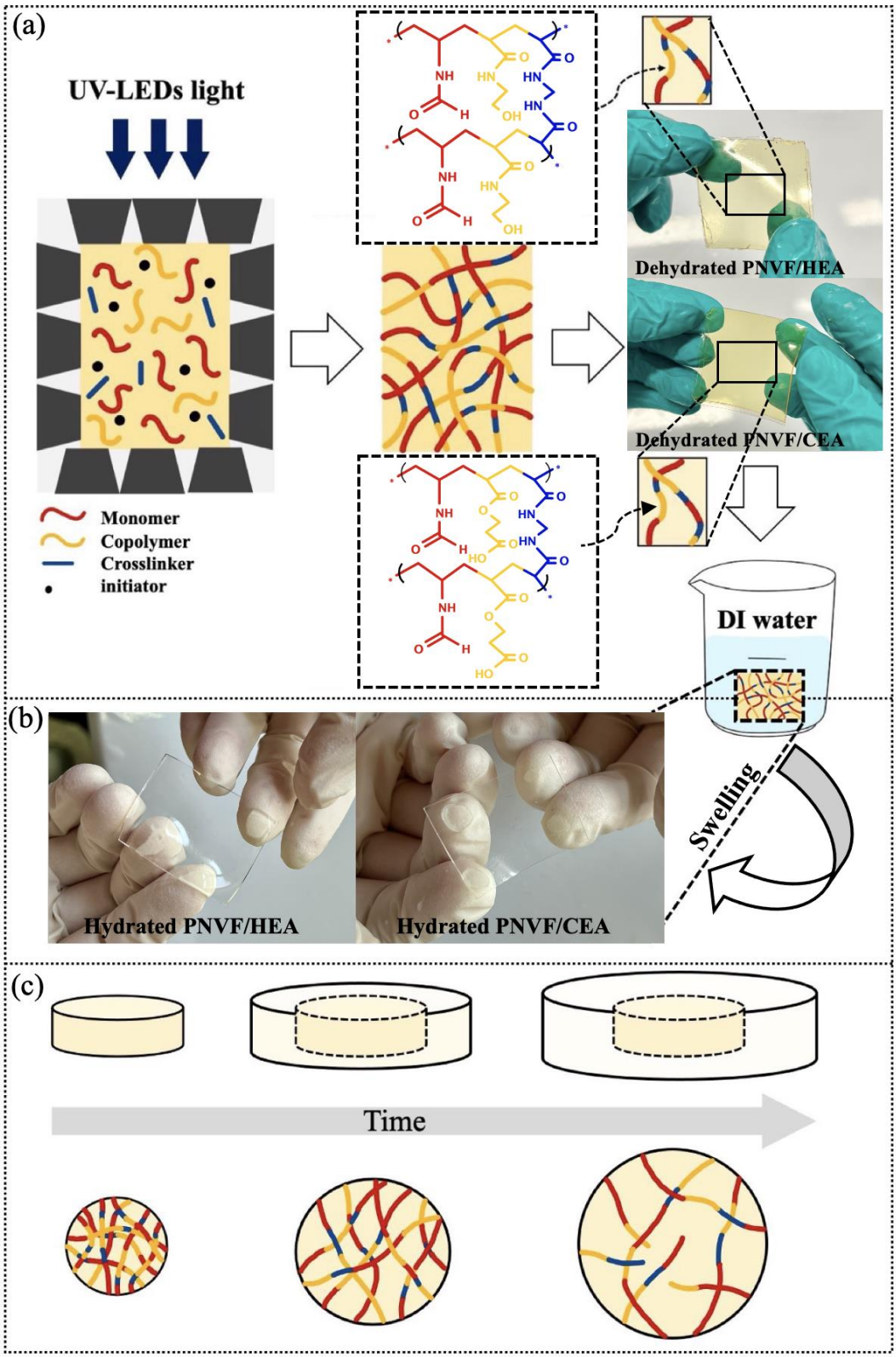


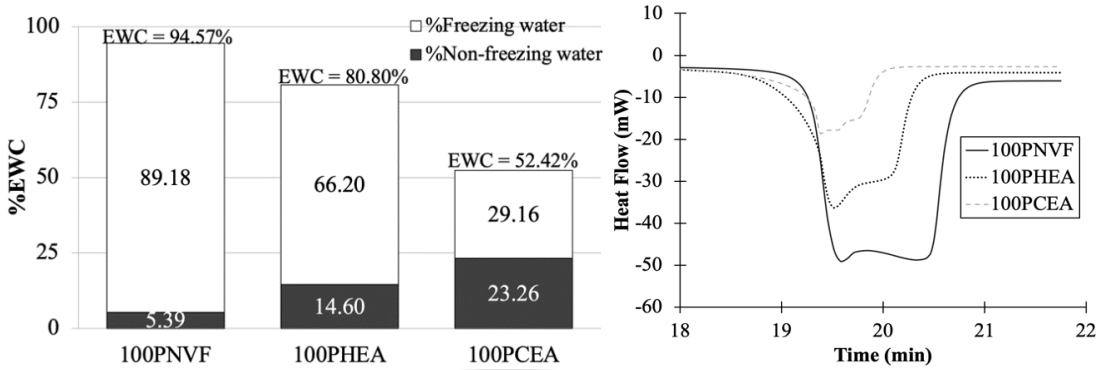
Figure 41 Schematic of preparation of hydrogels; (a) dehydrated poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogel, (b) hydrated poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogel and (c) swelling ability of polymer network of the hydrogels

In order to synthesis of poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogels, chemical crosslinks are formed by the reaction of the PEGDA acrylate end-groups, follows the typical free radical polymerization. Figure 41a shows the reaction between monomer, comonomer and crosslinker to form the polymer network in the hydrogel. After polymerization of the hydrogels, the thickness of the dehydrated hydrogels was measured and was approximately 0.1 cm. From figure 41a, it can be seen that both poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogels without water are stiff and brittle materials but after soaking in water for 5 days, they become soft and flexible gels. However, their thickness and softness depended on the copolymer type and ratio used, as for HEA in the hydrogels promoted higher flexibility and swelling of the hydrogels than that of CEA (figure 41b).

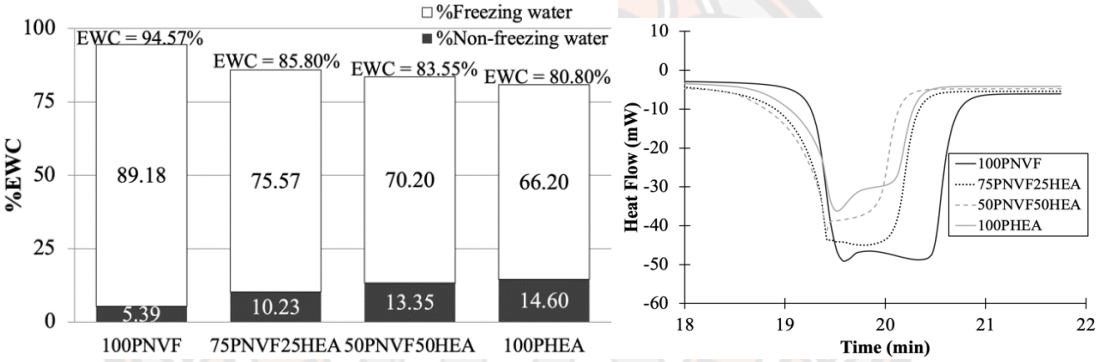
Equilibrium Water Content (%EWC) and water structure of the hydrogels

Hydrophilic polymers can absorb different amounts of water depending the density of the hydrophilic groups present on the polymer. The different loadings of copolymer from 0 to 50%wt of total weight of monomer was used to prepare hydrogels and to study the effect of copolymer loading on PNVF based hydrogels. The water properties of the hydrogel are very important to study because of the water in hydrogels is related how the hydrogels behave in many applications. To investigate the water, bulk water content is measured by equilibrium water content (%EWC). The state of water molecules in hydrogels can be further investigated by Differential Scanning Calorimetry (DSC) as water in hydrogels can exist in two separate types: freezing water (free water) and non-freezing water (bound water).

(a) Homopolymer



(b) PNVF and HEA hydrogels series



(c) PNVF and CEA hydrogels series

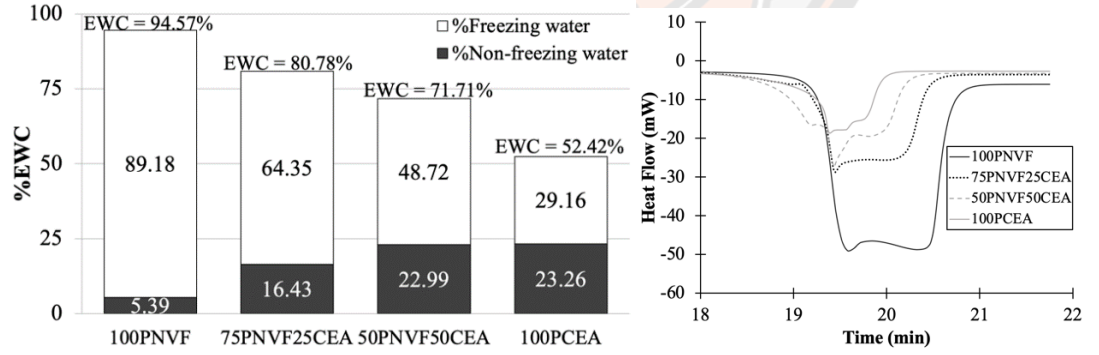


Figure 42 %EWC (right) and freezing and non-freezing water (left) in (a) homopolymer, (b) PNVF and HEA hydrogels series and (c) PNVF and CEA hydrogels series

The water content (%EWC) of each sample were observed and shown in figure 42 (left). The %EWC of homopolymer hydrogels showed that 100NVF, 100HEA and 100CEA were 94.57%, 80.80% and 52.42%, respectively. When the copolymers of NVF/HEA and NVF/CEA were measured, the %EWC presented a similar trend with a close to linear relationship between %EWC and content of the copolymer (trending towards the lower homopolymers %EWC). In both examples, 100PHEA and 100PCEA have a lower %EWC than 100PNVF, therefore the trend is for a decrease in the %EWC in the copolymer hydrogel samples.

Figure 42 (right) shows the amount of freezing water in the hydrogel that is calculated from the area under each peak. The figures show that the intensity of freezing water peak increase from 100NVF, 100HEA and 100CEA due to the water molecules of hydrogel have the different behavior in the hydrogel depending on the composition of monomer and interaction between water molecules and hydrogel structure. Whereas increasing copolymer loading in NVF, the DSC curve are increase sharp peak and also decrease freezing water may be due to the hydrogels have more H-bonding interaction of water and hydrogels. This result corresponds to the results from %EWC, when the water content increasing the freezing water is also increase for all polymer. In summary, it was found that all of NVF copolymer decrease water content when increase other monomer to copolymerization and the amount of freezing water follows the same trend as %EWC.

Contact angle (CAA) of hydrogels

In order to compare the surface of the hydrated hydrogels further, the hydrophilicity of the series was assessed using contact angle measurement and the photograph was taken that shows the contact angle automatically. For ease of understanding, when there is a low contact angle of a liquid on a solid surface, the surface is said to have good wettability.

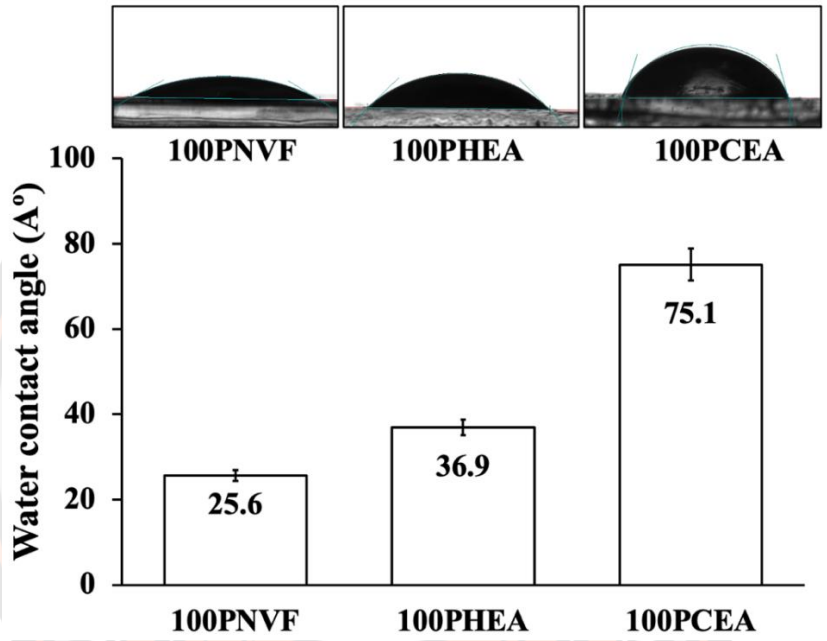
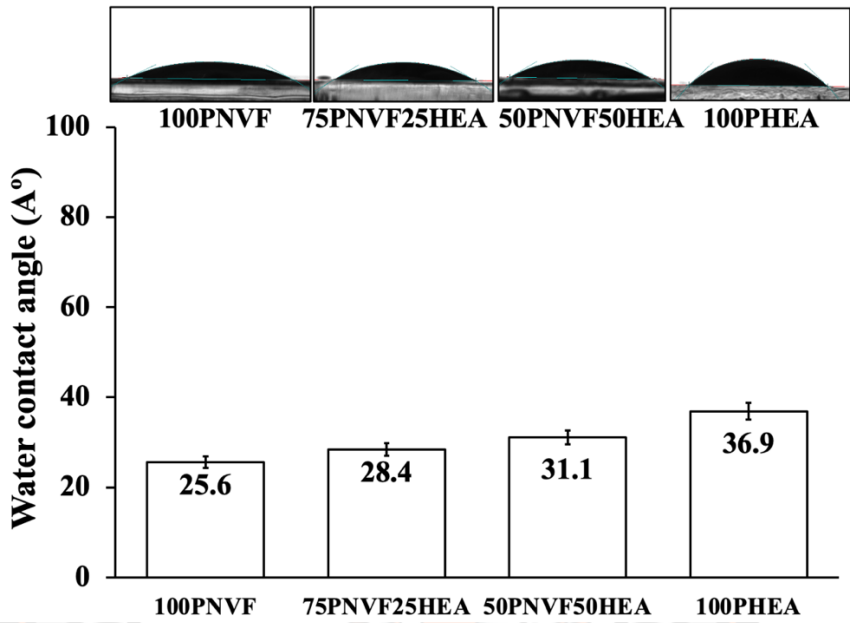


Figure 43 The water contact angle of homopolymer of hydrogels

Figure 43 presents the contact angle of each sample from table 11. The contact angle of the homopolymer hydrogels showed that 100PNVF, 100PHEA and 100PCEA were 25.6º, 36.9º and 75.1º, respectively. Importantly, all of these values are under 90º and therefore all the hydrogel samples are defined as hydrophilic.

(a)



(b)

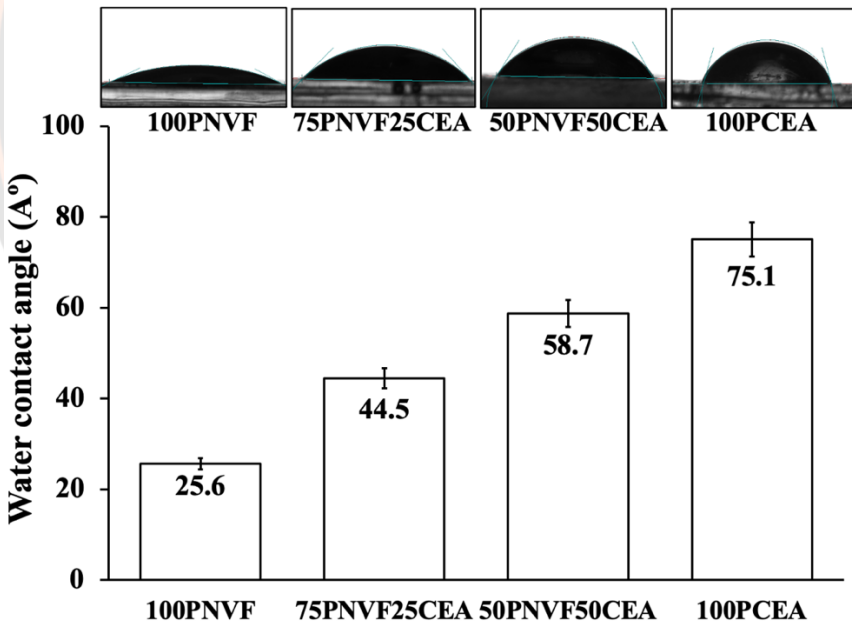


Figure 44 The water contact angle of PNVF-copolymer; (a) HEA, and (b) CEA hydrogels series

Figure 44 presents the PNVF/PHEA and PNVF/PCEA hydrogel samples contact angles at different ratios of copolymer. This followed a linear relationship between the two monomers used. For example, as PNVF has the lowest angle of the

three polymers, when the ratio of either HEA or CEA is increased the contact angle of the gel trends towards the higher angle of the homopolymer. This is typical behaviour for materials of this type as the surface properties are directly linked to the functional groups present at the surface of the hydrogel in the hydrated state.

Dye uptake and release

For controlled drug release studies, each hydrogel was soaked in a dye solution of know concentration (0.0001M) for 96 hours, three surrogate dyes were used (Orange II sodium salt, crystal violet and Congo red). Table 12 shows information about the organic dyes that we used in this work.

Table 12 Information of three surrogate dyes used

Name	Dye type	MW	pKa
Orange II sodium salt	Anionic azo dye	350.32	8.26, 11.4
Crystal violet	Cationic dye	407.99	9.4
Congo red	*neutral-ionic azo dye*	696.68	4

All dyes are classified as hydrophilic dyes and in order to calculate the amount of dye released a standard calibration curve for each dye was used to measure the release in μg/ml. This work involved dissolving a known concentration of a standard stock solution in deionized water. Solutions with various concentrations ranging from $1x10^{-2}$ to $1x10^{-6}$ M were prepared using a serial dilution technique. UV spectrophotometric scanning was performed in the range of 300-600 nm to determine the optimum absorbance under the experimental conditions.

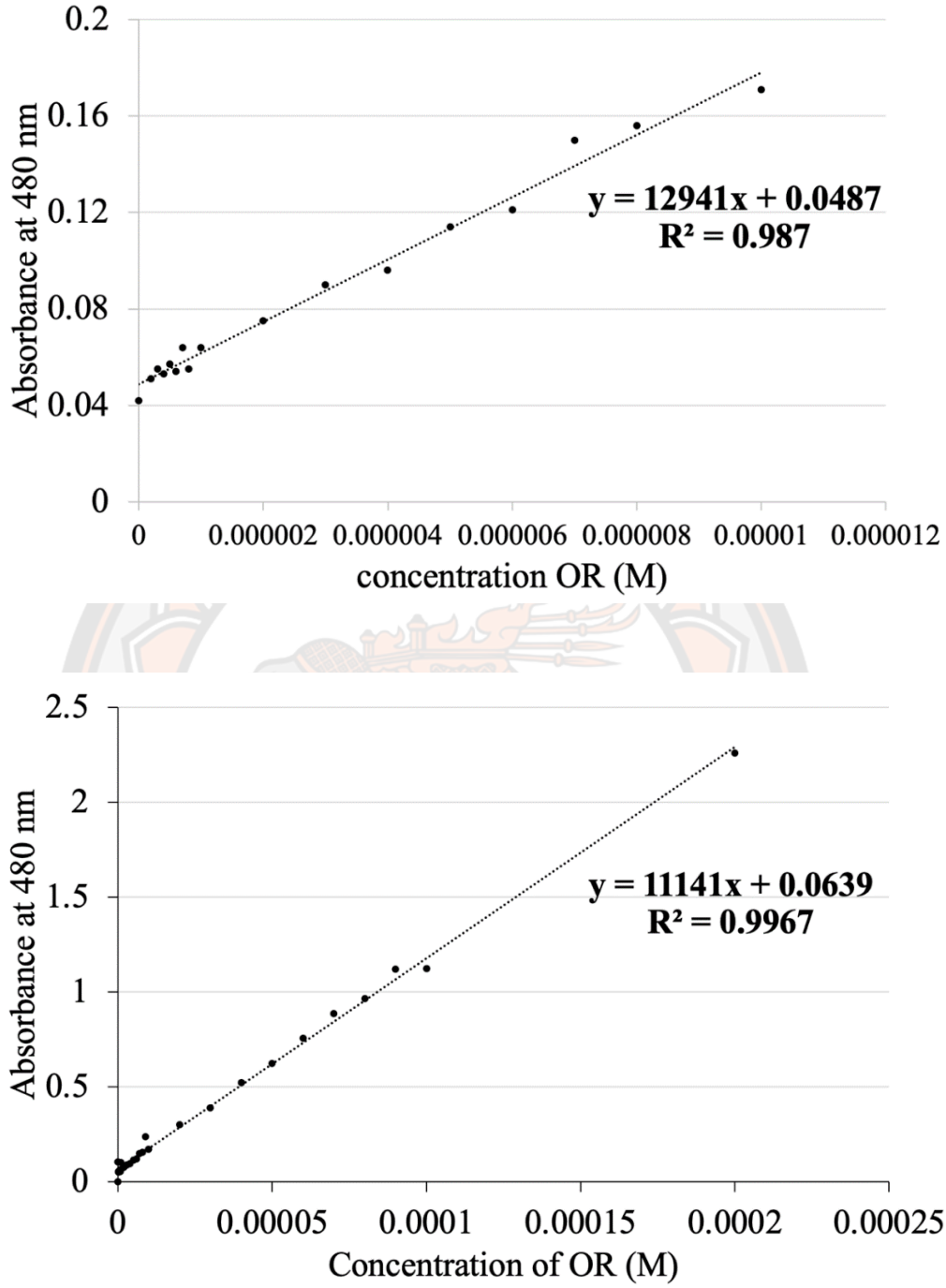


Figure 45 Standard calibration curve of orange II sodium dye solution at pH 7 in at 480 nm depended on the absorbance measurement; (left) below 0.2 and (right) upper 0.2

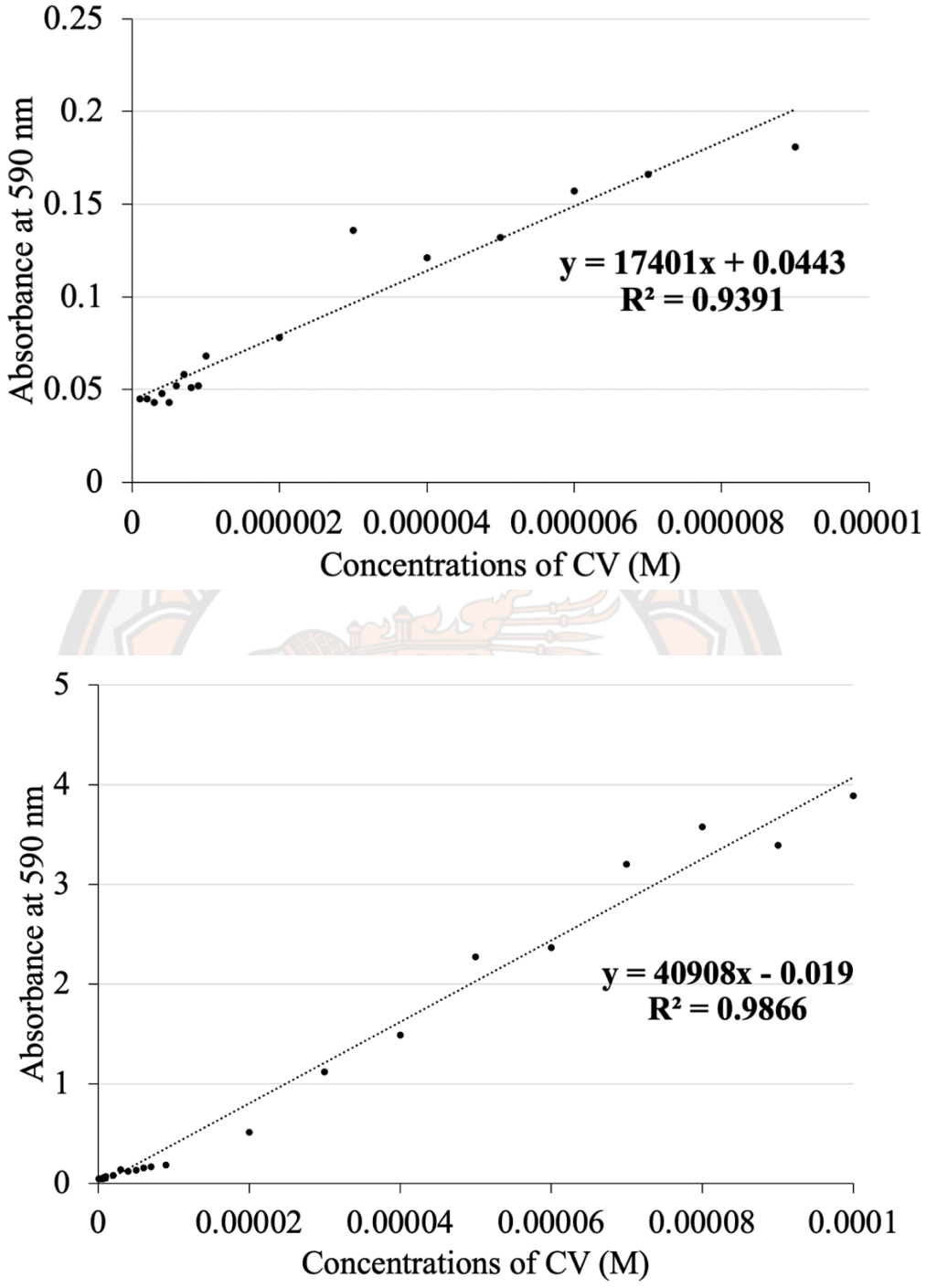


Figure 46 Standard calibration curve of crystal violet dye solution at pH 7 in at 590 nm depended on the absorbance measurement; (left) below 0.2 and (right) upper 0.2

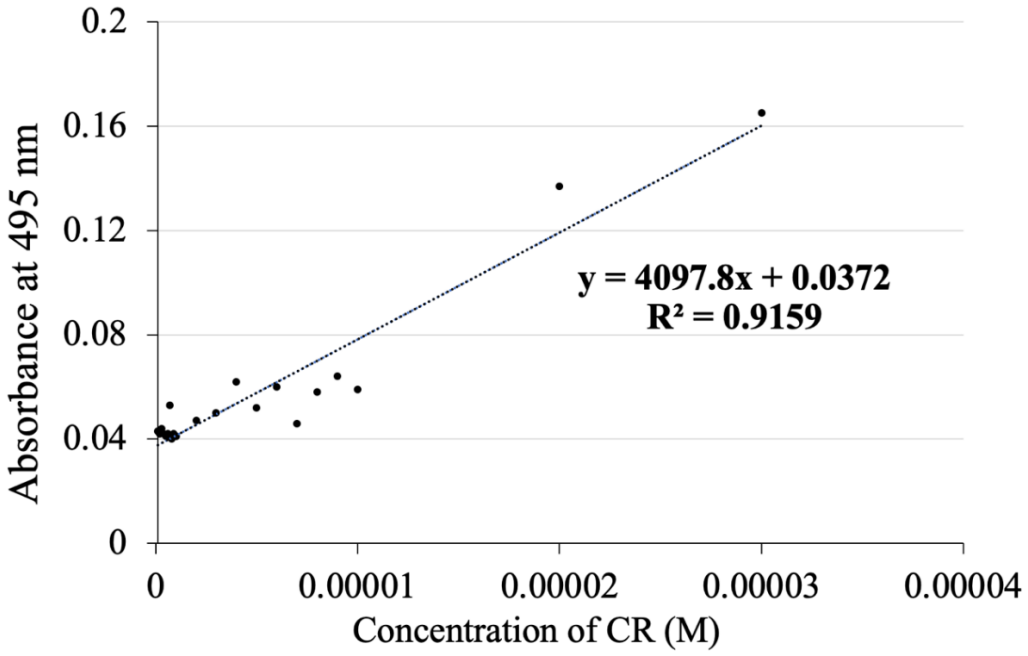


Figure 47 Standard calibration curve of Congo red dye solution at pH 3 in at 495 nm

A calibration curve was obtained from figure 45, 46 and 47 by plotting the concentration against absorbance, which was measured using a UV spectrophotometer. The optimum UV absorbance was determined by conducting a wavelength sweep and detecting the wavelength with the highest absorbance. These were: for orange II sodium salt 480nm, for crystal violet 590 nm and for Congo red 495nm.

Each graph plotted absorbance versus concentration to produce a calibration curve for each dye. With the calibration standards established, the uptake and release of the hydrogels could then be assessed.

1. Homopolymer hydrogels

Homopolymer hydrogels were chosen that all process difference functional group on the polymer structure. This is to interact with the functional group of dye molecules and the amounts of dye can uptake and release through hydrogels depends on the interactions between the dye and the hydrogel.

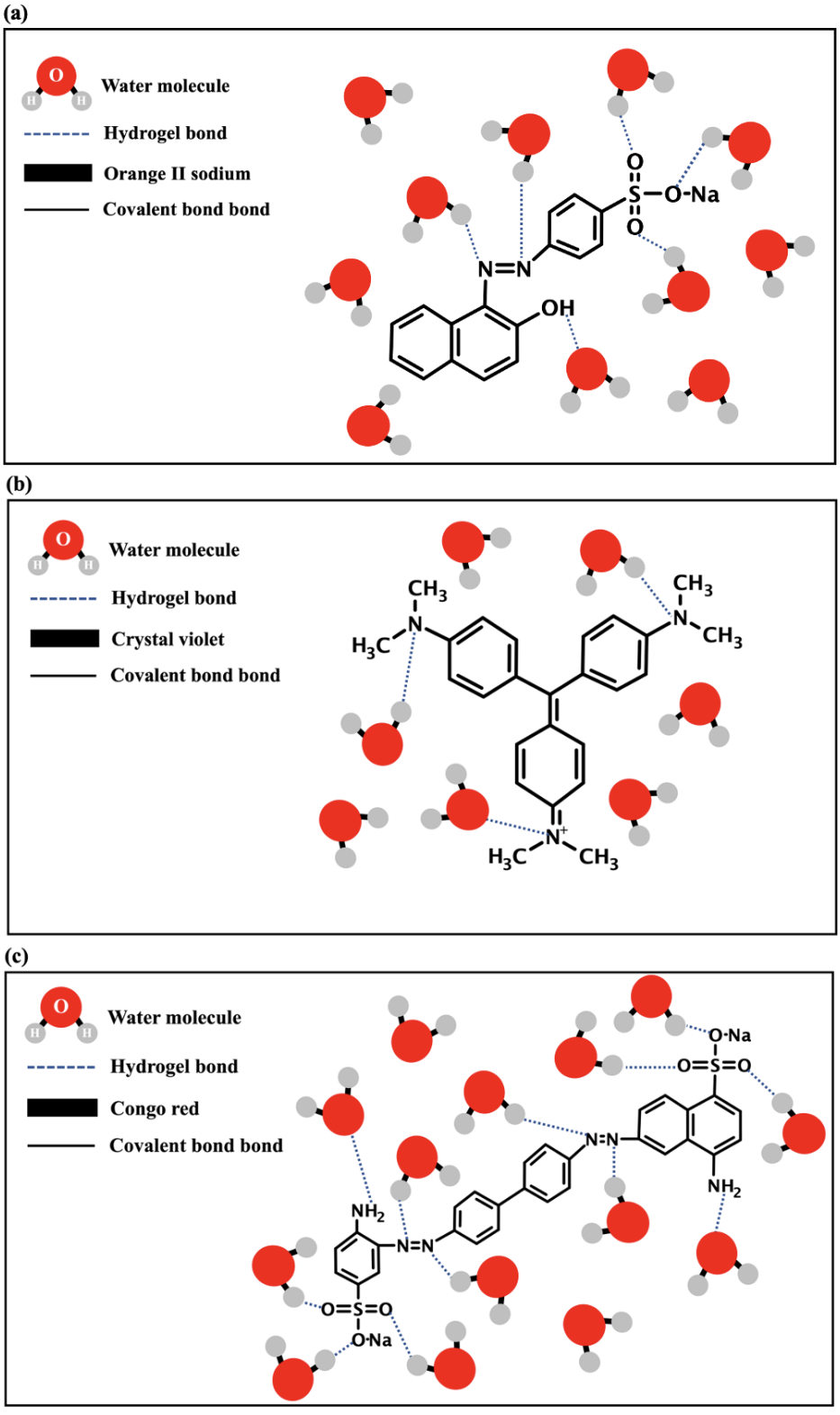


Figure 48 Graphic illustration of three surrogate dyes interaction: (a) O2S (b) CV and (c) CR interaction with water molecules at pH 7

Figure 48 shows the chemical structures of three dye molecules. Three dyes contain several electronegative nitrogen and oxygen atoms, and electropositive hydrogen atoms. The polar hydrogen, nitrogen, and oxygen atoms can form hydrogen bonds with free water molecules. Both types can interact and bind water molecules via hydrogen bonding. Therefore, the three surrogate dyes can potentially recruit water molecules to participate in strong ion–dipole bonding for ion–water interactions.

In order to investigate the release of dye molecules the colour parameter of the homopolymer hydrogels were measured before and after dye release and are presented along with the dye release profiles. Figure 49 shows the 3-dimensional CIELAB colour space of colour parameter measurements.

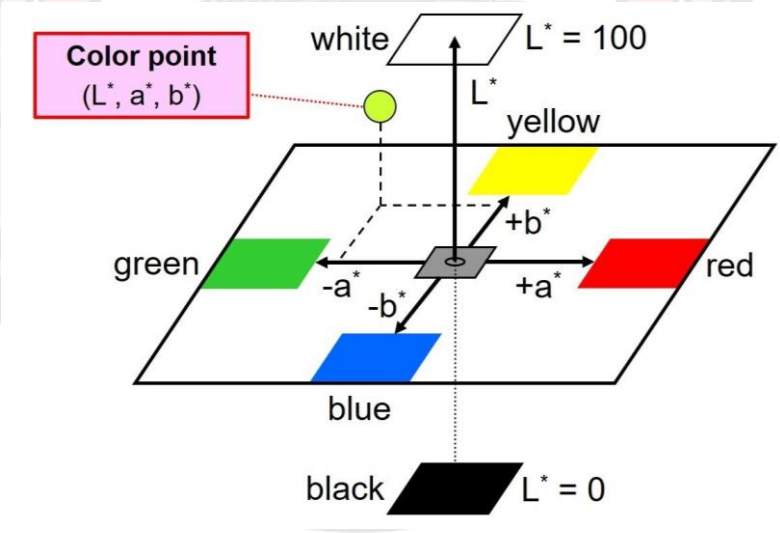


Figure 49 The 3-dimensional CIELAB colour space

For the homopolymer hydrogels, the samples were 100PNVF, 100PHEA and 100PCEA, with the results of the colour parameter presented in table 13, which consist of L*, a* and b*.

Table 13 Colour parameters and the total colour difference parameter of homopolymer hydrogels before and after dye releasing

samples	Colour parameters			ΔE
	L* (lightness)	a* (green-red)	b* (blue-yellow)	
Reference				
100PNVF	94.9±0.00	-0.2±0.00	3.9±0.05	Ref.
100PHEA	94.2±0.09	-0.2±0.00	3.6±0.10	Ref.
100PCEA	95.0±0.05	-0.2±0.00	5.0±0.00	Ref.
Before release				
OrangeIIsodium salt				
100PNVF	68.5±0.14	27.3±0.27	40.0±0.66	52.5
100PHEA	72.2±0.25	18.5±0.60	21.6±1.04	34.0
100PCEA	67.9±0.23	27.8±0.46	38.5±1.12	51.4
Crystal violet				
100PNVF	35.3±0.08	33.8±0.11	-65.5±0.09	97.6
100PHEA	21.5±0.04	33.8±0.08	-50.4±0.07	96.7
100PCEA	39.5±0.23	10.5±0.11	-28.3±0.23	65.6
Congo red				
100PNVF	47.9±1.18	44.6±2.50	20.9±2.22	67.1
100PHEA	57.8±0.96	30.4±2.01	12.9±1.45	48.5
100PCEA	63.0±0.09	-8.0±0.11	-11.5±0.12	36.8
after release				
OrangeII sodium salt				
100PNVF	80.6±0.08	-0.2±0.04	-3.4±0.09	16.1
100PHEA	80.7±0.23	-0.5±0.04	-4.7±0.00	15.9
100PCEA	77.7±0.44	9.0±0.63	7.8±0.90	19.8
Crystal violet				
100PNVF	78.8±0.30	0.4±0.05	-16.3±0.08	28.5
100PHEA	24.7±0.60	34.5±0.70	-52.8±0.59	95.9
100PCEA	44.3±1.57	5.5±0.47	-18.0±0.90	55.9

Congo red				
100PNVF	82.3±0.10	14.8±0.21	2.6±0.08	19.6
100PHEA	72.9±0.00	28.2±0.00	12.6±0.00	36.6
100PCEA	68.0±0.00	-17.5±0.04	-8.9±0.04	34.9

Values are mean ± SD (n=5)

The results show that after releasing of dye solution, the ΔE parameter of homopolymer hydrogels (100PNVF, 100PHEA and 100PCEA) change to lower values with 100PNVF showing the highest change. Moreover, when uptake (before releasing) the three dyes into the hydrogels the colour of each gel changes considerably and are presented in figure 50. The results from the colour parameter, show that 100PNVF hydrogels release the largest amounts of the dye solution.

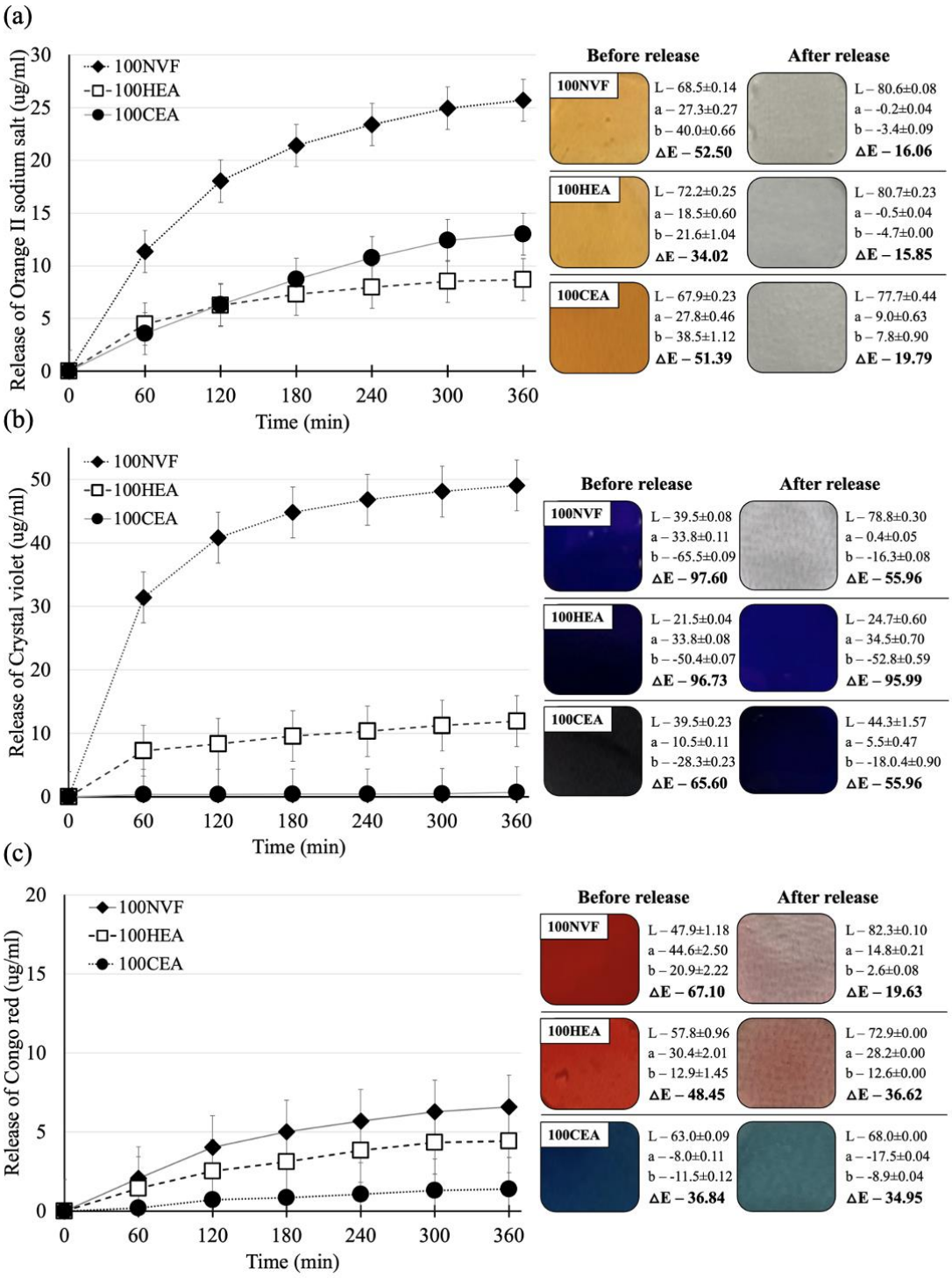


Figure 50 Release profiles of homopolymer; 100PNVF, 100PHEA and 100PCEA in difference dye (left) and the optical photographs and the colour parameters (right) before and after dye releasing; (a) O2S, (b) CV and (c) CR

The cumulative amount of dye released from the homopolymer hydrogels in DI water are shown in figure 50 (left). The release profiles of the homopolymers show that 100NVF releases a higher amount of dye that the other polymers for each of the three dyes. This could possibly due to 100PNVF processing the highest water content as well as high freezing water content (free water). Thus, has the potentially has the largest reservoir of dye and the dye that is incorporated into the gel should also be fully solubilized by the free water and therefore is available for release. In direct contrast to this 100PCEA gives the lowest release across the range of dyes. Although, CEA also gives the most differences in both the release profiles and also the colour of gels before and after release. This may be due to CEA being an anionic hydrogel, thus the dye can be strongly held in the hydrogel structure but interacting with the polymer structure.

The optical photographs and the colour parameters in figure 50 (right), visually allow for the amount of dye released to be tracked. The change in colour parameter of the homopolymer with the different dyes, shows that the value of each parameter varies both before and after releasing. The 100PNVF had the largest change in the total colour difference parameter (ΔE). Therefore, the colour change for 100PNVF hydrogels follows the same trend as the cumulative amount of dye released. Again, we see that the P100CEA gel shows considerable differences from the other gels in terms of the physical colour of the gel and also in the colour parameter. This is because the CEA polymer structure contains the pendent carboxyl group and lowers the pH of the hydrogel, by taking a litmus paper strip and placing on the surface of the hydrogel gives a pH of 4.5.

2. Release profiles of copolymer hydrogels

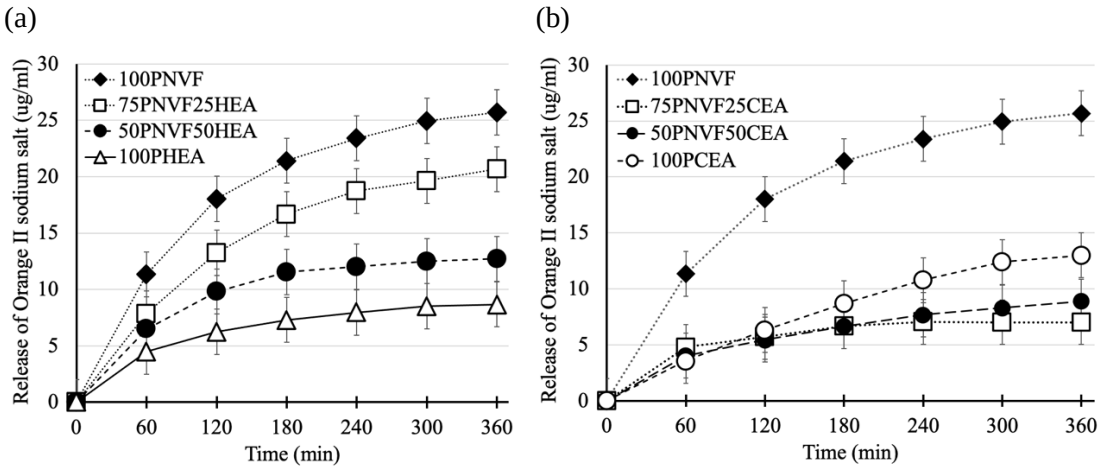


Figure 51 Release profiles of (a) PNVF and PHEA hydrogels series and (b) PNVF and PCEA hydrogels series with 0.0001M Orange II sodium salt dye solution

Figures 51-53 present the release profiles of poly(NVF-co-HEA) and poly(NVF-co-CEA)copolymer hydrogels with the three selected dyes (OR, CV and CR). Figure 51 shows the release profiles of the copolymer hydrogels after the uptake of orange II sodium salt (OR) dye. The results show that as the amount of either HEA or CEA is increased (25 or 50%wt) the amount of OR released decreases. A noteworthy observation is that 100PCEA hydrogels gives a higher value that the copolymers. The values are similar for the copolymer gels and because CEA is acidic that alters the colour of the gel meaning that errors associated with this pair are higher than the majority of the other samples. For the poly(NVF-co-HEA) samples we find a reduction in the release profile curves as the composition of the copolymer changes.

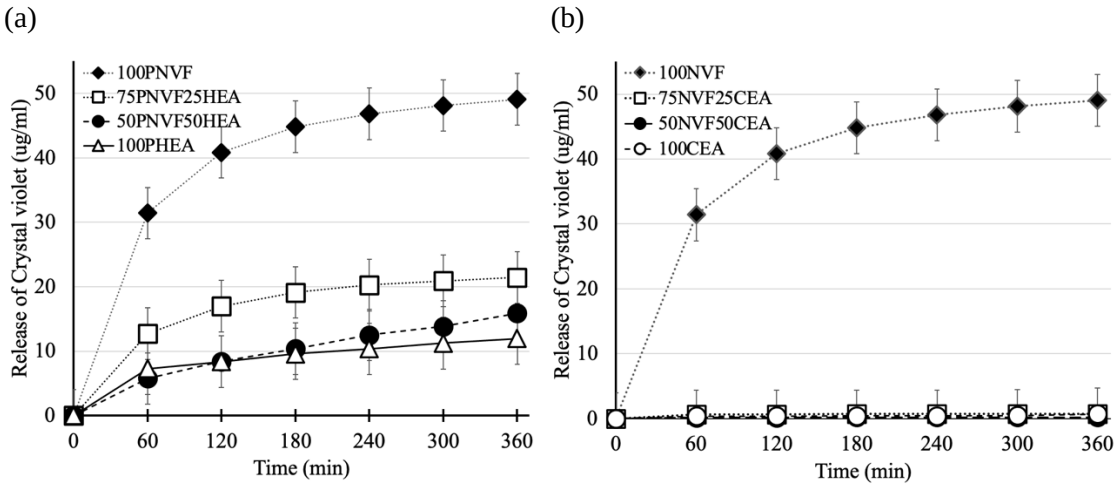


Figure 52 Release profiles of (a) PNVF and PHEA hydrogels series and (b) PNVF and PCEA hydrogels series with 0.0001M Crystal violet solution

Figure 52 shows the release profiles of the copolymer hydrogels after the uptake of crystal violet (CV) dye. The differences in the release between poly(NVF-co-HEA) and poly(NVF-co-CEA)hydrogels are the most pronounce of all the dyes tested. The main reason for this is that CEA contains a carboxylate group which can strongly interact with crystal violet structure. Whereas, HEA contains hydroxyl group which can interact with the amide groups of CV but not as intensely. Thus, from the colour pictures in figure 50 (b) we can see that both PHEA and PCEA hydrogels present a dark blue / purple colour but with CEA being darker after uptake and also remaining almost the same colour after release, where HEA colour is less intense.

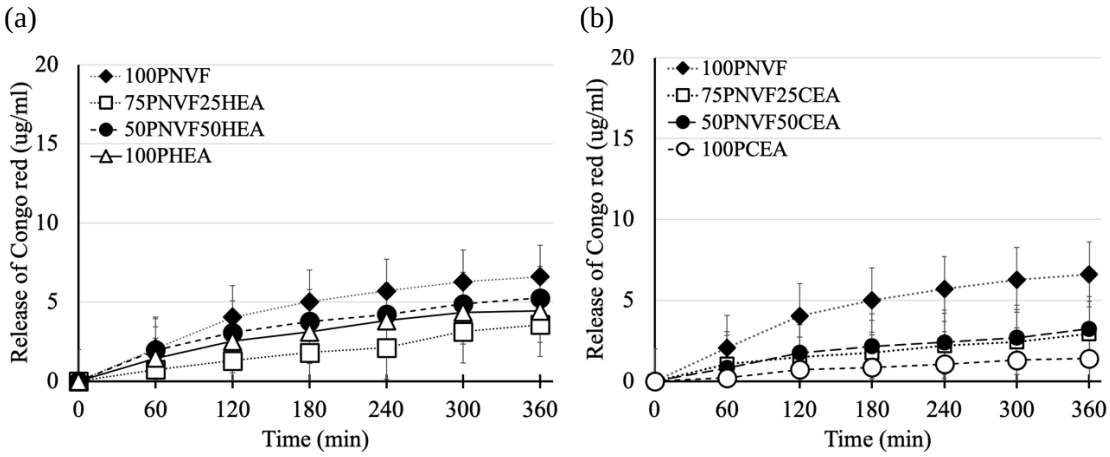


Figure 53 Release profiles of (a) PNVF and PHEA hydrogels series and (b) PNVF and PCEA hydrogels series with 0.0001M Congo red

Figure 53 shows the release profiles of the copolymer hydrogels after the uptake of Congo red. The results show that Congo red gives the lowest release of all the tested dyes. From figure 50(b) we see that all the homopolymers did not fully release Congo red during the 6 hours release period. The release profiles although low are also the most linear. Both of these findings are the result of the inherent properties of Congo red, which contains both sulfonic acid and amine groups, thus has the ability to interact with all three of the polymers present in the different gels.

Development of hydrogels for novel cosmetic hydrogel patches

For this section, after we studied the effect of initiation system and various copolymer ratios for producing the PNVF based materials and its copolymer hydrogels. 75PNVF25HEA hydrogels via UVLED photopolymerization gave overall the best results in term of physical gel properties. In general, to development of hydrogels depends on several factor such as crosslinker and Photo-initiator which control the final hydrogels properties. Therefore, the most suitable for cosmetic applications of hydrogel patches are important to study. The fabrication and characterization will be presented into three parts: **(1) Effect of cross-linker on hydrogel patches, (2) Effect of Photo-initiator on hydrogel patches (3) Effect of glycerin and preservative on hydrogel patches.**

1. Effect of cross-linker on hydrogel patches

In this part, novel cosmetic hydrogel patches of 75PNVF25HEA were prepared by UVLED photopolymerization. Hydrogels are chemically synthesized by adding crosslinking agent for this section two sets of experimental preparation of hydrogels were studied. The first experiment studied the effect of the type of crosslinker and the second experiment studied the effect of the concentration of the crosslinker onto the properties of the fabricated hydrogels.

The results will be discussed and compared between the three crosslinkers at different concentrations from 1.0 to 4.0 %w/w to produce hydrogels, the different crosslinkers were; Di(ethylene glycol) diacrylate (DEGDA), Poly(ethylene glycol) diacrylate (PEGDA), and N,N-methylenebisacrylamide (MBAAm). Figure 54 shows the molecule structure of three crosslinker which DEGDA, PEGDA and MBAAm, their molecular weight is 214.22, 575 and 154.17, respectively.

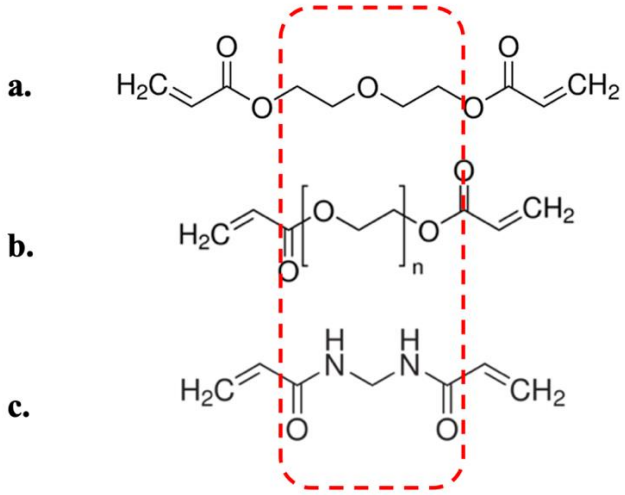


Figure 54 Comparison of the molecular structure of (a) DEGDA, (b) PEGDA and (c) MBAAm

These three crosslinker have been employed in the crosslinking of polymers, they are used as a prepolymer solution. Both Diethylene glycol diacrylate (DEGDA) and Poly(ethylene glycol) diacrylate (PEGDA) are a hydrophilic crosslinkers derivative of ethylene glycol that can be used for a variety of drug delivery and tissue engineering based applications.

The formulations of 75PNVF25HEA hydrogels with various concentration of crosslinker from 0 to 4.0 %w/w, are shown in table 14. All hydrogel sample were successfully fabricated, however, only some were suitable for the desired application as cosmetic patches. The reason for the gels being not suitable were one of the following: brittle, weak gel and non-adhesive to the skin.

Table 14 Formulations of a series of hydrogels at different composition

Sample code	Crosslinker (%w/w of monomer)			Photo-initiator (%w/w)	Gel formation for applied gel properties
	DEGDA	PEGDA	MBAAm		
1	4.0	-	-	1	X
2	3.0	-	-	1	X
3	2.0	-	-	1	X
4	1.0	-	-	1	X
5	-	4.0	-	1	X
6	**-**	**3.0**	**-**	**1**	✓
7	**-**	**2.0**	**-**	**1**	✓
8	**-**	**1.0**	**-**	**1**	✓
9	-	-	4.0	1	X
10	-	-	3.0	1	X
11	-	-	2.0	1	X
12	**-**	**-**	**1.0**	**1**	✓

Note: The symbol of ✓ means the successful fabrication, while X means the failure to produce hydrogels with suitable properties for cosmetic applications

In this part, three different crosslinkers were used to form the hydrogels that are classed as chemical crosslinking agents. From Table 14, in case of using DEGDA as a crosslinker from 1.0 to 4.0 %w/w were successful fabrication into form to hydrogels but shows the failure of the fabrication into the form in term of applied gel properties for the cosmetic application of hydrogel patches. On the other hand, in the case of using PEGDA as a crosslinker from 1.0 to 3.0 %w/w were successful of the fabrication into the form applied gels. For the MBAAm of crosslinker, at 1.0%w/w were successful fabrication into the form in term of applied gel properties for cosmetic application of hydrogel patches. However, it can be seen that at the high

concentration of crosslinker than 1.0%w/w was not suitable for cosmetic application of hydrogel patches.

To evaluate and compare the effects of these two parameters on the %EWC and wettability of the hydrogels, two different types of crosslinkers at various concentrations were studied. However, the 1% MBAAm 75PNVF25HEA hydrogel showed a similar texture to the 3% PEGDA 75PNVF25HEA hydrogel, so low concentrations of MBAAm at 0.5% w/w and 0.1% w/w were used to fabricate the hydrogels instead.

In this part, five different samples were investigated, which are 3%PEGDA 75PNVF25HEA, 1%PEGDA 75PNVF25HEA, 1%MBAAm 75PNVF25HEA, 0.5%MBAAm 75PNVF25HEA and 0.1%MBAAm 75PNVF25HEA. Following fabrication these gels were characterized for equilibrium water content and wettability. Table 15 shows the compositions, %EWC and wettability of a series of hydrogels with various crosslink densities.

Table 15 Formulations, %EWC and wettability of a series of hydrogels comparing crosslink density

Sample code	Feature of hydrogels	% EWC	Contact angle (A^O)
3%PEGDA 75PNVF25HEA	Firm gel and surface moisture gel	81.36	17.7
1%PEGDA 75PNVF25HEA	More hydrated gel and softer gel	88.95	15.8
1%MBAAm 75PNVF25HEA	Good adhesion to the skin and more surface moisture	86.59	22.3
0.5%MBAAm 75PNVF25HEA	Flexible gel, good adhesion to the skin and surface moisture	88.92	26.2
0.1%MBAAm 75PNVF25HEA	More flexible, good adhesion to the skin and more surface moisture	93.02	10.7

The equilibrium water content (%EWC) of each sample was observed and shown in Table 15. The %EWC showed that, 3%PEGDA 75PNVF25HEA, 1%PEGDA 75PNVF25HEA, 1%MBAAm 75PNVF25HEA, 0.5%MBAAm 75PNVF25HEA and 0.1%MBAAm 75PNVF25HEA had %EWCs of 81.36%, 88.95%, 86.59%, 88.92 and 93.02, respectively. The results show that, when decreasing the concentration of crosslinker into the polymerization system the %EWC of 1%PEGDA 75PNVF25HEA is the slightly lower than the rest. It can be seen that, the crosslink density effects the %EWC of these hydrogels. The highest %EWC was 93.02% when using MBAAm as the crosslinker at 0.1% of concentration, this can be explained due to hydrophilic character of MBAAm along with the “looser” nature of the network.

However, the %EWC of 1%PEGDA 75PNVF25HEA and 0.5%MBAAm 75PNVF25HEA gave the similar %EWCs of 88.95 and 88.92, respectively. However, both samples gave a similar texture of hydrogel but in term of industry processing concern, 0.5%MBAAm 75PNVF25HEA gave the best results in terms of cost of processing, which can be used to produce hydrogels at a fraction of the cost to both the environment and industry compared to high concentration of crosslinker.

From Table 15, the contact angle (Aº) of all the sample hydrogels showed that, 3%PEGDA 75PNVF25HEA, 1%PEGDA 75PNVF25HEA, 1%MBAAm 75PNVF25HEA, 0.5%MBAAm 75PNVF25HEA and 0.1%MBAAm 75PNVF25HEA were 17.7º, 15.8º, 22.3º, 26.2º and 10.7º, respectively. Importantly, all these values are under 45 and therefore all the hydrogel samples are defined as super hydrophilic. This is typical behaviour for materials of PNVF-co-HEA gels as the surface properties are directly linked to the functional groups present at the surface of the hydrogel in the hydrated state. In terms of the differences with contact angle an error of between 5-10º is often associated with the technique.

2. Effect of Photo-initiator on hydrogel patches

In this part, the novel cosmetic hydrogel patches of 1%PEGDA 75PNVF25HEA, 0.5%MBAAm 75PNVF25HEA and 0.1%MBAAm 75PNVF25HEA hydrogels were investigated with different concentrations of photo-initiator. The results will be discussed and compared of Diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide (TPO); TPO as a photo-initiator at different concentrations, which were 0.5%w/w and 1.0 %w/w to produce the hydrogels. %EWC and wettability were observed to find the best composition that can be most suitable in term of fabrication and application in the field of cosmetic hydrogel patches. Table 16 shows the formulations, %EWC and wettability of a series of hydrogels with various amounts the photo-initiator to form gels for cosmetic hydrogel patches.

Table 16 Formulations, %EWC and wettability of a series of hydrogels with varies the concentration of photo-initiator into form the gel for cosmetic hydrogel patches

Sample code	Photo-initiator (%w/w)	% EWC	Contact angle (A^O)
1%PEGDA 75PNVF25HEA-1%PI	1.0	88.95	15.8
0.5%MBAAm 75PNVF25HEA-1%PI	1.0	88.92	26.2
0.1%MBAAm 75PNVF25HEA1%PI	1.0	93.02	10.7
1%PEGDA 75PNVF25HEA-0.5%PI	0.5	88.67	18.8
0.5%MBAAm 75PNVF25HEA-0.5%PI	0.5	88.91	26.3
0.1%MBAAm 75PNVF25HEA0.5%PI	0.5	93.40	10.2

The equilibrium water content (%EWC) of each sample were observed and shown in Table 16. The %EWC showed that, 1%PEGDA 75PNVF25HEA-1%PI, 0.5%MBAAm 75PNVF25HEA-1%PI, 0.1%MBAAm 75PNVF25HEA-1%PI, 1%PEGDA 75PNVF25HEA-1%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI were 88.95%, 88.92%, 93.02%, 88.67, 88.91 and 93.40 respectively. The results showed that decreasing the concentration of the photo-initiator in the polymerization system had very little effect on the %EWC of all hydrogels.

From Table 16, the contact angle (A^{O}) of the sample hydrogels showed that, 1%PEGDA 75PNVF25HEA-1%PI, 0.5%MBAAm 75PNVF25HEA-1%PI, 0.1%MBAAm 75PNVF25HEA1%PI, 1%PEGDA 75PNVF25HEA-1%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI were 15.8º, 26.2º, 10.7º, 18.8º, 26.3º and 10.2º, respectively. Importantly, all these values are under 45 and therefore all the hydrogel samples are defined as super hydrophilic. This is typical behaviour for materials of PNVF-co-HEA gels as the surface properties are directly linked to the functional groups present at the surface of the hydrogel in the hydrated state. Again, the differences in contact angle are not significant with an error of between 5-10º is often associated with the technique.

However, the different concentrations of photo-initiator gave similar results, with %EWC and wettability also gave the similar texture of hydrogels but in terms of industry processing concern to use the lower concentrations of photo-initiator at 0.5%w/w by far in term of cost processing that can be used to produce hydrogels at a fraction of the cost to both the environment and industry compared to high concentration of photo-initiator. **Therefore, 1%PEGDA 75PNVF25HEA-0.5%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI were chosen to study further.**

3. Effect of glycerin and preservative on hydrogel patches

Glycerin (glycerin) is one of the most widely used skin moisturizers (humectants) in the cosmetic industry, it is used to improve skin hydration and reduce trans-epidermal water loss (TEWL). Phenoxyethanol is one of the most popular skin care preservatives and is used to prevent the growth of fungi, bacteria, and yeast in hydrogels. It can be found in many cosmetic and personal care products.

This part will be focus on applied gel properties, to discussed and compared the effect of skin moisturizers and preservative loadings on applied gel properties for the most suitable for cosmetic application of hydrogel patches, 10%w/w of glycerin was used as a skin moisturizers and 0.5%w/w of phenoxyethanol was used as a preservative to produce the cosmetic hydrogel patches.

Following the previous studied, 1%PEGDA 75PNVF25HEA-0.5%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI were chosen to studies for comparisons the effect of glycerin and phenoxyethanol loading, defined as added and no added hydrogel samples, %EWC and wettability properties were observed to find the best composition that is most suitable for the application in the field of cosmetic hydrogel patches. Table 17 shows the formulations, %EWC and wettability of a series of hydrogels with added glycerin and phenoxyethanol produce the cosmetic hydrogel patches.

Table 17 Formulations, %EWC and Contact angle of a series of hydrogels with added glycerin and phenoxyethanol produce the cosmetic hydrogel patches

Sample code	Additive loading	% EWC	Contact angle (A^o)
1%PEGDA 75PNVF25HEA-0.5%PI	X	88.67	18.8
0.5%MBAAm 75PNVF25HEA-0.5%PI	X	88.91	26.3
0.1%MBAAm 75PNVF25HEA0.5%PI	X	93.40	10.2
1%PEGDA 75PNVF25HEA-0.5%PI	✓	91.09	13.0
0.5%MBAAm 75PNVF25HEA-0.5%PI	✓	88.61	25.9
0.1%MBAAm 75PNVF25HEA0.5%PI	✓	93.52	15.4

Note: The symbol of ✓ means loading, while X means no additional glycerin and phenoxyethanol to produce the cosmetic hydrogel patches

The equilibrium water content (%EWC) of each sample were measured and shown in Table 17. The results showed that, when loading the glycerin and phenoxyethanol into the polymerization system there was a very small effect to the %EWC of all hydrogels. Therefore, the glycerin and phenoxyethanol do not affect the %EWC of these hydrogels.

From Table 17, when loading the glycerin and phenoxyethanol into the polymerization system, all hydrogels show a very small effect to the contact angle (A^o). Same as the %EWC results, the glycerin and phenoxyethanol did not affect to the contact angle (A^o) of these hydrogels and the contact angle of all these values are under 45 and therefore all the hydrogel samples are defined as super hydrophilic. This is typical behaviour for materials of PNVF co HEA gels as the surface properties are directly linked to the functional groups present at the surface of the hydrogel in the hydrated state.

However, to confirm the most suitable applied gel properties for cosmetic hydrogel patches, mechanical properties is importantly to study in next section.

Tensile properties

For the tensile testing the results are discussed into two parts, first, the hydrogels were compared for the effect of crosslinkers at different concentrations. The second part, the hydrogels are reported that compare the effect of glycerin and phenoxyethanol addition to produce the hydrogels, with both added and no added additives. All compositions of a series of hydrogels were characterized for their tensile properties and reported in term of percentage of elongation at break (%E), tensile strength at break and modulus, the results are shown in table 18.

To study the effect of crosslinkers at different concentrations from the previous results of %EWC (table 15), 1%PEGDA 75PNVF25HEA-0.5%PI and 0.5%MBAAm 75PNVF25HEA-0.5%PI hydrogel showed similar %EWCs of 88.95 and 88.92, respectively and both gave similar textures of the gel. The results show that in terms of industry processing concern, **0.5%MBAAm 75PNVF25HEA** gave the best results in terms of cost processing. However, mechanical properties are also important to study for confirmation of this. From Table 18, 1%PEGDA 75PNVF25HEA-0.5%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI hydrogels were characterized for their tensile properties.

Table 18 Tensile properties of a series of hydrogels with different loading of crosslinker

Samples code	% Elongation at break (%)	Tensile strength at break [MPa]	Modulus [MPa]
1%PEGDA 75PNVF25HEA-0.5%PI	92.5±4.6	30.41±1.5	33.23±1.7
0.5%MBAAm 75PNVF25HEA-0.5%PI	103.0±5.2	27.0±1.4	26.20±1.3
0.1%MBAAm 75PNVF25HEA0.5%PI	117.0±5.9	16.98±0.8	14.68±7.3
0.1%MBAAm 75PNVF25HEA0.5%PI-Ad	134.0±6.7	12.62±0.6	9.52±0.5

Values are mean ± SD (n=3)

Table 18 shows the results of comparison with 1%PEGDA 75PNVF25HEA-0.5%PI and 0.5%MBAAm 75PNVF25HEA-0.5%PI showed that when using 0.5%MBAAm as a crosslinker, it gave a higher %E than 1%PEGDA of crosslinker and shown lower tensile strength at break and modulus. However, both gels are similar, and both have potential to be used in the desired application of cosmetic hydrogels.

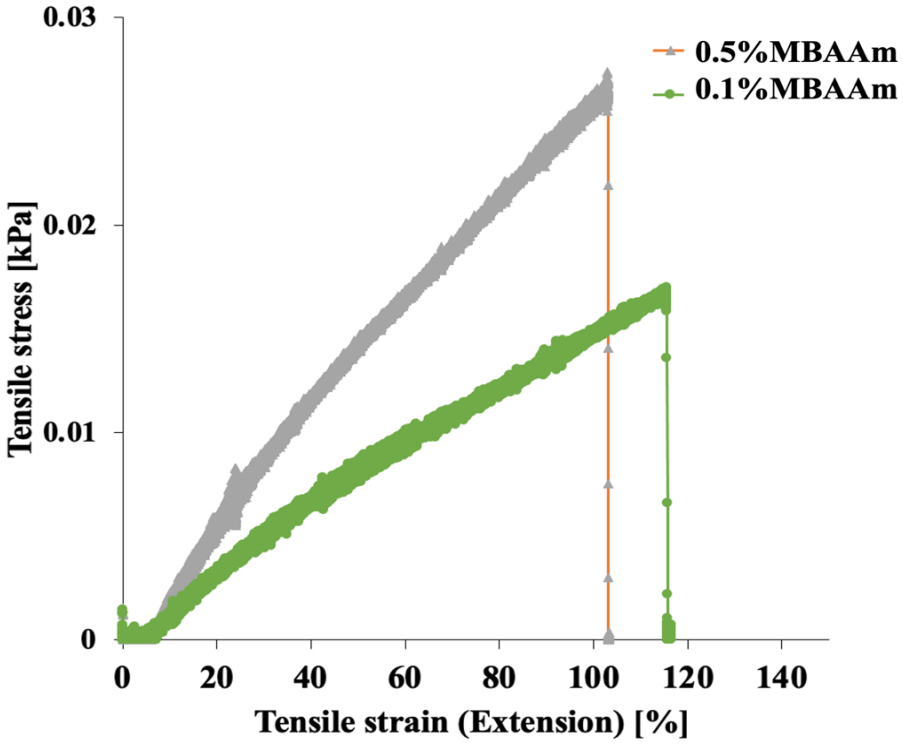


Figure 55 Tensile properties of 75PNVF25HEA-0.5%PI hydrogels with different concentration of MBAAm as a crosslinker

Further investigation of MBAAm system was assessed with %E at different concentration of MBAAm studied. Figure 55, shows that while these hydrogels show an increase of %E when lower amount of MBAAm was used, there is a more pronounced reduction in the tensile modulus. This is due to the degree of crosslinker affected the polymer chains during the polymerization process.

According to the requirement of high %E, high tensile stress, and low modulus value of hydrogels, therefore the sample of 0.1%MBAAm 75PNVF25HEA-0.5%PI

that had %E higher and lower modulus than 0.5%MBAAm. So, the 0.1%MBAAm 75PNVF25HEA-0.5%PI hydrogel were selected for further study about the effect of glycerin and phenoxyethanol loading.

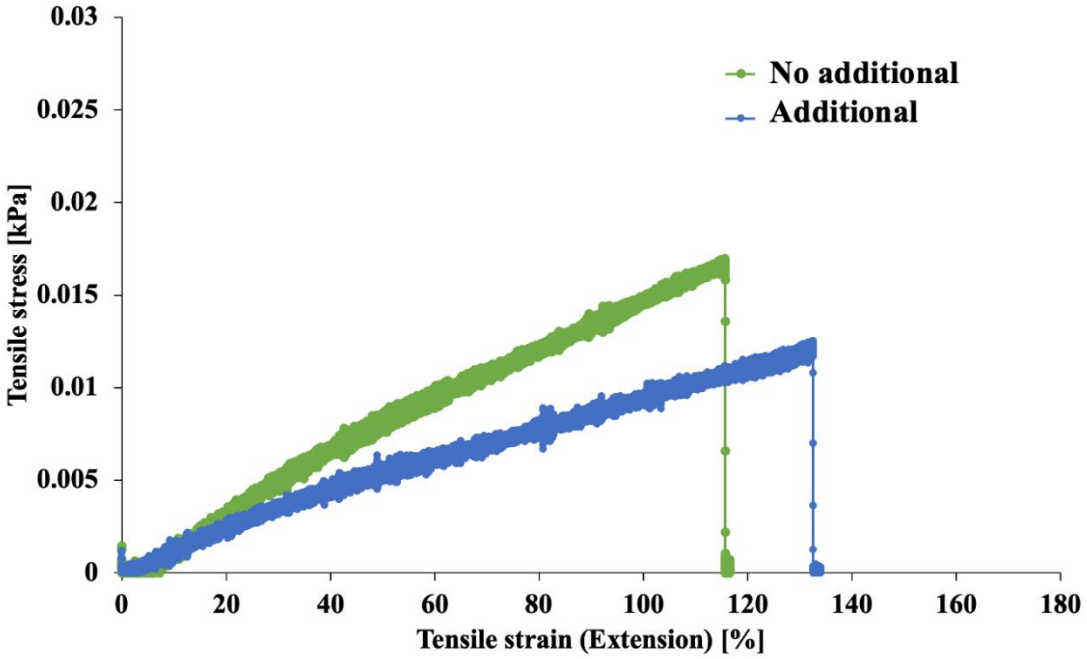


Figure 56 Tensile properties of a series of hydrogels with comparing the effect of additional glycerin and phenoxyethanol loading to the hydrogels

For the part of comparing the effect of glycerin and phenoxyethanol loading to produce the hydrogels. Figure 56 presents the mechanical performance of hydrogels with additive loading and no additional additive - labelled as 0.1%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels, respectively. Figure 57 shows the optical images and characteristic of 0.1%MBAAm 75PNVF25HEA-0.5%PI hydrogel samples with and without additives running on the tensile test using a universal tensile testing machine. A load cell of 500 N and extension rate of 5.0 mm/min was used.

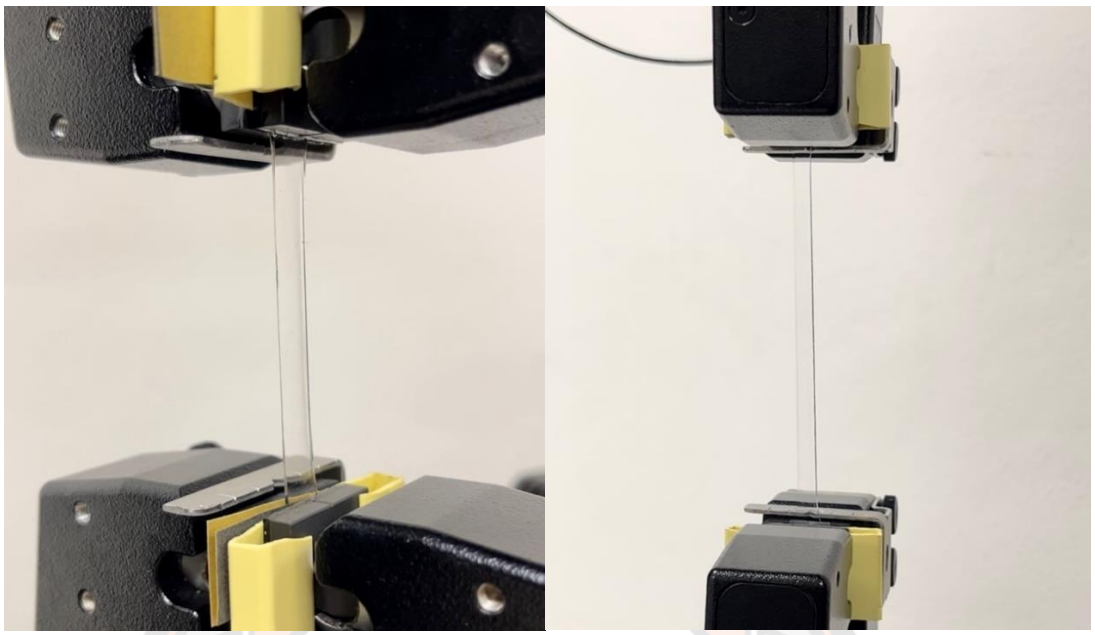


Figure 57 Optical image of tensile testing of hydrogels with different additive loading; (a) 0.1%MBAAm 75PNVF25HEA-0.5%PI and (b) 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad

The %E of each sample were observed and shown in table 18. The %E showed that, 0.1%MBAAm 75PNVF25HEA-0.5%PI and 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels were 117.0 and 134.0, respectively. The results show that, higher tensile strength and elongation at break follow by lower modulus with the addition of glycerin. These results suggested that the improvement of %E of hydrogels were promoted by the addition of glycerin into the hydrogel synthesis system. This is possible because glycerin enhances the gel flexibility.

In summary of this section, **the most suitable applied gel properties for cosmetic hydrogel patches was: 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels, using 0.1% of MBAAm as a crosslinker based on 75% of PNVF co 25% of HEA with glycerin and phenoxyethanol loading via free-radical polymerization via UVLEDs initiation system with diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide was used as photo-initiator.**

Figure 58 Optical image of 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad

In the next section, 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogel (figure 58) were investigated for their applied gel properties in term of *in-vivo* clinic with bio-adhesion ability and skin irritation after application are reported in the next sections.

In vivo testing / Specific testing of applied gel properties

1. **Bio-adhesion ability**

The ability of patch adhesion was evaluated from being adhered to the forearm of the volunteer for a duration of 30 mins for a single application of the hydrogels tested, a total of thirty healthy female volunteers with normal skin properties was included in this study. If there was no slippage or de-adhesion then the gel was successful in adhesion. From figure 59, the results show that, 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels exhibited bio-adhesion to the skin with a long

duration of skin adherence more than 30 minutes. All volunteers were also observed for skin irritation.

In general, the flexibility of the hydrogel patches effected to the adhesive property of the skin. The increase in the flexibility of the hydrogel patches can improve the contact between the hydrogel patches and the skin. Skin adhesion if often linked to several factors, these include the mechanical properties of the gels, surface energy (contact angle) and appetite of water in the skin.

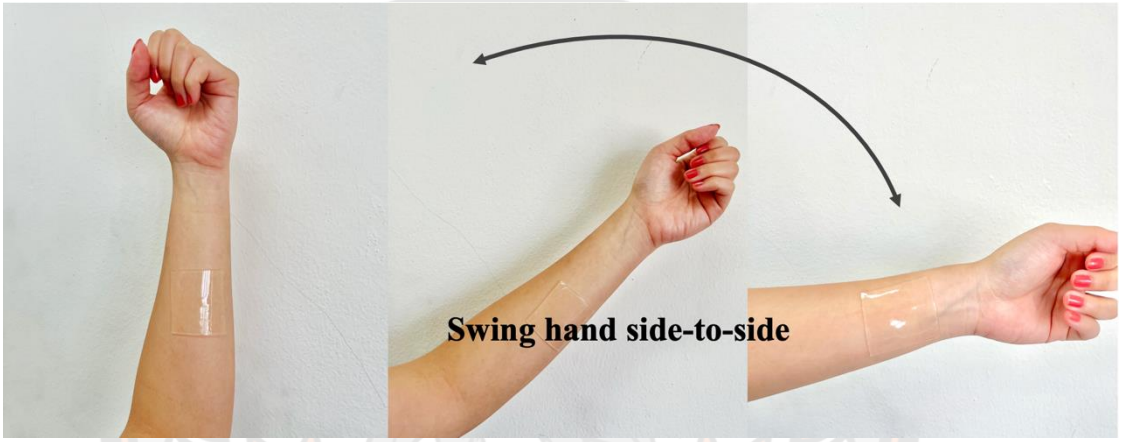


Figure 59 Electronic photograph of patch exhibited bio-adhesion to the skin with a long duration of skin adherence for 30 minutes

2. Skin irritation after application

In this study, the square hydrogel patch of 5x5 cm^{-2} was used for a single application of the hydrogels tested, a total of thirty healthy female volunteers with normal skin properties was included in this study. The age of volunteers averaged 26 years. The skin properties were evaluated by visual inspection after removal of the patch, their skin characteristics around the testing area were recorded in a photograph e.g.

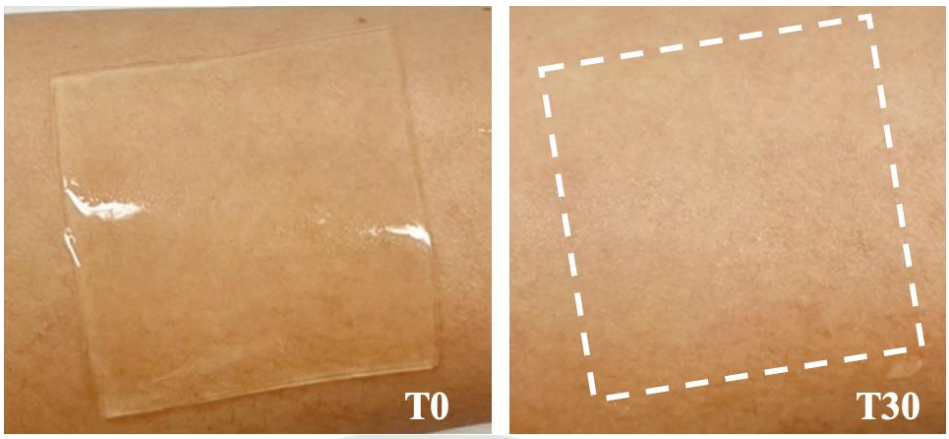


Figure 60 Electronic photograph of the volunteer's forearm for evaluation of skin irritation procedure on application (T0 (left)) and after removal (T30 (right))

Figure 60. Therefore, after single application of the cosmetic hydrogel patch, no signs of skin irritation were observed for all 30 participates during this study, indicating that the developed hydrogels for novel cosmetic hydrogel patches applications are mild to the skin.

Comparison of the novel cosmetic hydrogel patches with the commercial cosmetic products

To confirm the most suitable gel for cosmetic hydrogel patches was the 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels, this gels applied performance was compared to commercial cosmetic products. From the previous studied in part 1, the commercial cosmetic products were characterized for base gel properties as a guideline to produce the synthetic NVF-based cosmetic hydrogels. This section will be reported the comparison the most suitable cosmetic hydrogel patch with commercial cosmetic products. Base gel properties, which are equilibrium water content (EWC), wettability and mechanical analysis of hydrogels were observed for comparison, additional applied gel properties were also reported via a self-perception Questionnaires were test after single application.

Base gel properties

1. **Equilibrium Water Content (%EWC)**

To evaluate and compare these hydrogel patches, 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels and commercial cosmetic products which were Faith in face, Medius heart and Beauty formulas. From figure 61, the %EWC is measured by contact angle testing.

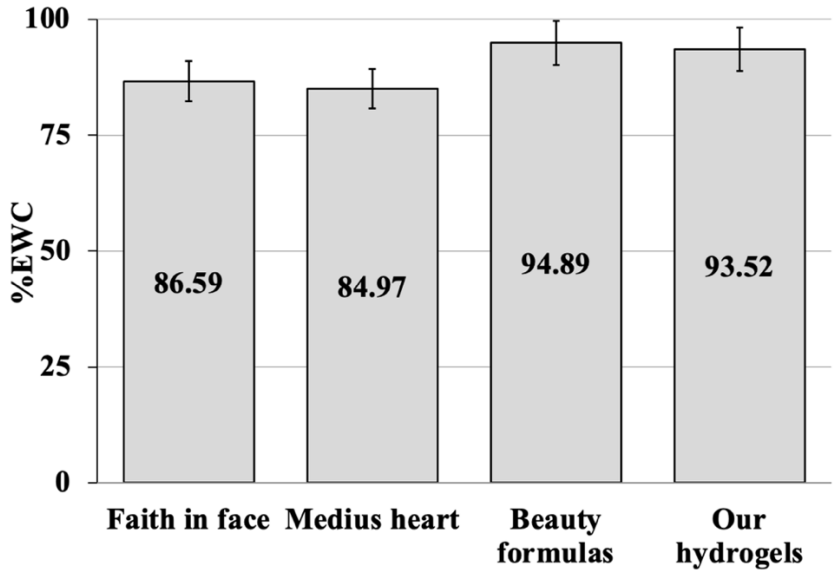


Figure 61 Equilibrium water content (%EWC) of all hydrogel patches

The equilibrium water content (%EWC) of each hydrogel patch were observed and shown in figure 61. The %EWC showed that, Faith in face, Medius heart, Beauty formulas and 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels were 86.59%, 84.97%, 94.89% and 93.52%, respectively. The Beauty formulas showed the highest %EWC was 94.89%, while 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels gave the high %EWC was 93.53%, these high water contents are due to the larger amount of water that can be associated with the polymers present in gel, which were Cetearyl Alcohol and Isopropyl Palmitate for Beauty formulas and N-vinylformamide and N-Hydroxyethyl acrylamide (HEA) for our hydrogel. However, all tested gels gave 'high' %WC (> 84%).

2. Wettability

To compare the surfaces of the hydrated hydrogels, the hydrophilicity of all hydrogel patches were assessed using contact angle measurements. The contact angle of hydrogel patches was observed and shows in figure 62.

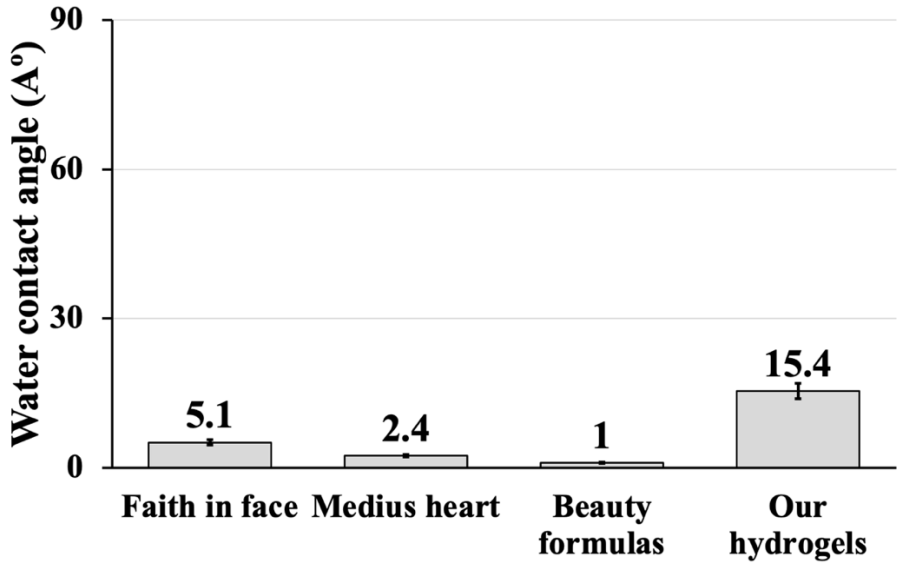


Figure 62 The wettability of the all hydrogel patches

The Faith in face, Medius heart, Beauty formulas and 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels patch is 5.1, 2.4, 1.0 and 15.4, respectively. It

can be seen that, all hydrogel patches showed very low contact angle of a liquid on a solid surface. Importantly, all of these values are below 45^{O} and therefore all the hydrogel samples are defined as hydrophilic, with hydrophilic materials as the surface properties.

3. Tensile properties

All hydrogel patches were characterized for their tensile properties and reported in term of percent elongation at break (%), tensile strength at break (KPa) and modulus (KPa), with the results presented in table 19. This table contains the data of hydrogel patches which, percent elongation at break (%E), tensile strength at break and modulus.

Table 19 Tensile properties of hydrogel patches

Test product	%E (%)	Tensile strength (KPa)	Modulus (KPa)
Faith in face	83.90±4.5	914.85±45.7	1126.20±5.8
Medius heart*	232.70±11.6	174.49±8.7	97.68±4.9
Beauty formulas	252.64±12.6	184.17±9.2	72.90±3.6
0.1%MBAAm 75PNVF25HEA0.5%PI-Ad	134.0±6.7	12.0±0.6	9.52±0.5

Note: *The Medius heart are combine with hydrogel and fibre blend in the compositions

From table 19, tensile strength at break (KPa) and percent elongation at break (%) of all commercial cosmetic products. The Faith in face gave the highest of tensile strength and modulus value are 914 KPa and 1126.20 KPa, respectively due to this product have the fibre blend in their compositions.

The results of comparison with the all of commercial cosmetic products and 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels showed that, all of commercial cosmetic products showed higher %E, although it was not a considerable difference. Whereas, tensile strength at break and modulus shown big different value of

commercial cosmetic products than 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels. However, in term of use in the application, the modulus of hydrogels is of interest to further investigate. The 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels gave the low value of the modulus, its showed good elastic modulus of hydrogels.

In summary of the section, when comparing our hydrogels with the commercial cosmetic products, according to the requirement of good mechanical properties, which high %E, high tensile stress, and low modulus value of hydrogels, the 0.1%MBAAm 75PNVF25HEA-0.5%PI hydrogel shows the good performance and the ability to function as a gel for cosmetic hydrogel patches.

Applied gel properties: Self-perception Questionnaires

The subjects' skin was assessed on their forearms after a single 30-minute application and they were asked to complete a Self-Perception Questionnaire (SPQ). Thirty healthy female volunteers with a mix of skin types were enrolled in the study and completed it. The subjects had varying skin characteristics. The results, as reported in the Self-Perception Questionnaire (SPQ), confirmed the subjects' perception, as shown in Figure 63.

The detail of the Self-Perception Questionnaire form was reported which, more relaxed, more cooled, more hydrated, softer and adhesive hydrogels after 30 minutes applied the hydrogel patches. Combining the level of score (1 is strongly disagree, 2 is Disagree, 3 is neutral, 4 is Agree and 5 is Strongly agree).

Questionnaire for apply product

Number.....Name ..... Phone .....

Code .....

Sexual

☐ male ☐ female

Age (years)

☐ below 20 ☐ 20-30 ☐ 31-40 ☐ above 40

Have you ever used mask hydrogels product?

☐ Yes ☐ No

How do/did you choose your product?

☐ Friend / relative recommendation ☐ Brand

☐ Dermatologist advice ☐ Pricing

☐ Product's ingredients

Details	Level of score				
	5	4	3	2	1
1. More Relaxed					
2. More Cooled					
3. More hydrated					
4. Softer					
5. More adhesive					

Note:

1 is strongly disagree

2 is Disagree

3 is neutral

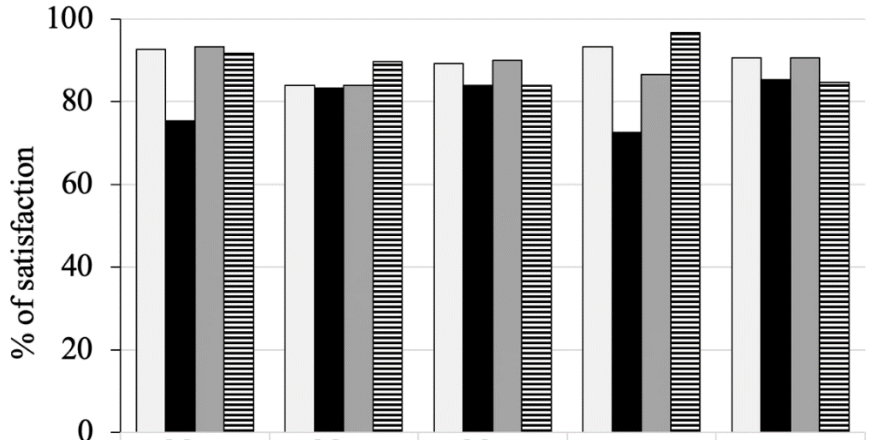
4 is Agree

5 is Strongly agree

Figure 63 Example of the Self-Perception Questionnaire form of the study subjects after one application (T30)

This section will be reported the comparison 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels with commercial cosmetic products are Faith in face, Medius heart and Beauty formulas.

(a)



	More Relaxed	More Cooler	More hydrated	Softer	more Adhesived
Faith in face	92.67	84	89.33	93.33	90.67
Beauty formulas	75.33	83.33	84	72.66	85.33
Midus heart	93.33	84	90	86.67	90.67
Our hydrogels	91.72	89.69	84.01	96.74	84.7

(b)

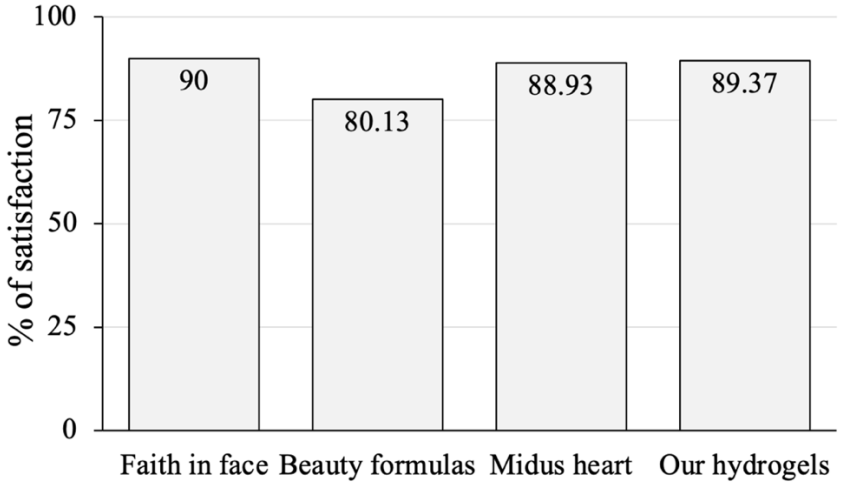


Figure 64 Self-Perception Questionnaire responses of the study subjects after one application of the commercial cosmetic products; (a) each detail and (b) the overall of satisfaction

From figure 64(b), Faith in face gave the highest score of overall of %satisfaction of the study subjects after one application (T30) is 94%. The study subjects felt their skin was more relaxed after treatment is 92.67%. They also felt their skin was more cooled (84%), more hydrated (89.33%), softer (93.33%) and adhesive (90.67%) (Shown in figure 64(a). While the lowest score of overall of %satisfaction is 80.13% (Beauty formulas). Moreover, 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogel shows 89.37% of overall of %satisfaction of the study subjects after one application. The study subjects felt their skin was more relaxed after treatment is 91.72%. They also felt their skin was more cooled (89.69%), more hydrated (84.01%), softer (96.74%) and adhesive (84.7%) (Shown in figure 64(a).

Several studies have demonstrated that glycerin enhances skin hydration and moisturizing properties. Therefore, the 0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogel was selected as the most suitable cosmetic hydrogel patch in this study. This hydrogel-based patch has a high glycerin content, which acts as a humectant, attracting water to the stratum corneum. To maintain healthy skin, it is important to keep it hydrated. A single treatment with the commercial cosmetic products can achieve these effects, but prolonged use can produce even better results.

In conclusion of this part, interestingly, all the instrumental results were confirmed by the subjects' perception in the Self-Perception Questionnaires. The results showed that, when compared with the commercial cosmetic products, 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogel in this study is an effective product to improve skin hydration, providing a cooling, relaxed, softer feel and shows good adhesive properties. Moreover, most of the subjects presented the 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogel is 89.37% of overall of %satisfaction. Therefore, this hydrogel patch is satisfactory for use as the hydrogel patch mask product.

Approach 2: Synthesis of the NVF-copolymer hydrogels containing the cationic poly(vinylamine)

This section focuses on the preparation of pH-responsive hydrogels and their potential use as delivery carriers for cosmetic active ingredients. The hydrogels are synthesized through UV-LED photopolymerization using a copolymer of poly(NVF) and hydroxyethyl acrylamide (HEA) as monomers and N,N-methylenebisacrylamide (MBAAm) as a cross-linker.

The aim of this part is to synthesize poly(NVF-co-HEA) hydrogels containing cationic poly(vinylamine) (PVAm) by partially hydrolyzing the formamide groups present in poly(NVF-co-HEA) under acidic conditions. The properties of the resulting poly(NVF-co-HEA-VAm) hydrogels (hydrolysed hydrogels) will be compared to that of the non-hydrolysed poly(NVF-co-HEA) hydrogels. The study will evaluate the basic properties of the hydrogels, such as their swelling behavior as a function of pH, and their pH-responsive release capabilities to understand their potential as active delivery carriers. Additionally, the study will investigate the conductivity of cosmetic active ingredients.

Preparation of poly(NVF-co-VAm-co-HEA) hydrogel

Figure 65 shows the preparation of the poly(NVF-co-HEA) containing cationic poly(vinylamine) (PVAm). First, poly(NVF) and copolymer hydrogels were prepared. The cross-link network was achieved by using 0.5% of N,N-methylenebisacrylamide (MBAAm) as a crosslinker, 1% of PI and hydroxyethyl acrylamide (HEA) were used as a copolymer. Then, hydrolysis of the poly(NVF-co-HEA) hydrogels by acid hydrolysis condition gave the poly(NVF-co-VAm-co-HEA) hydrogels by partially hydrolysed formamide group. The pH responsive hydrolysed hydrogels were prepared in 2 steps as follows: (1) preparation of poly(NVF-co-HEA) hydrogels and (2) hydrolysis of the formamide group of poly(NVF-co-HEA) hydrogels under HCl acid conditions is shown in figure 65.

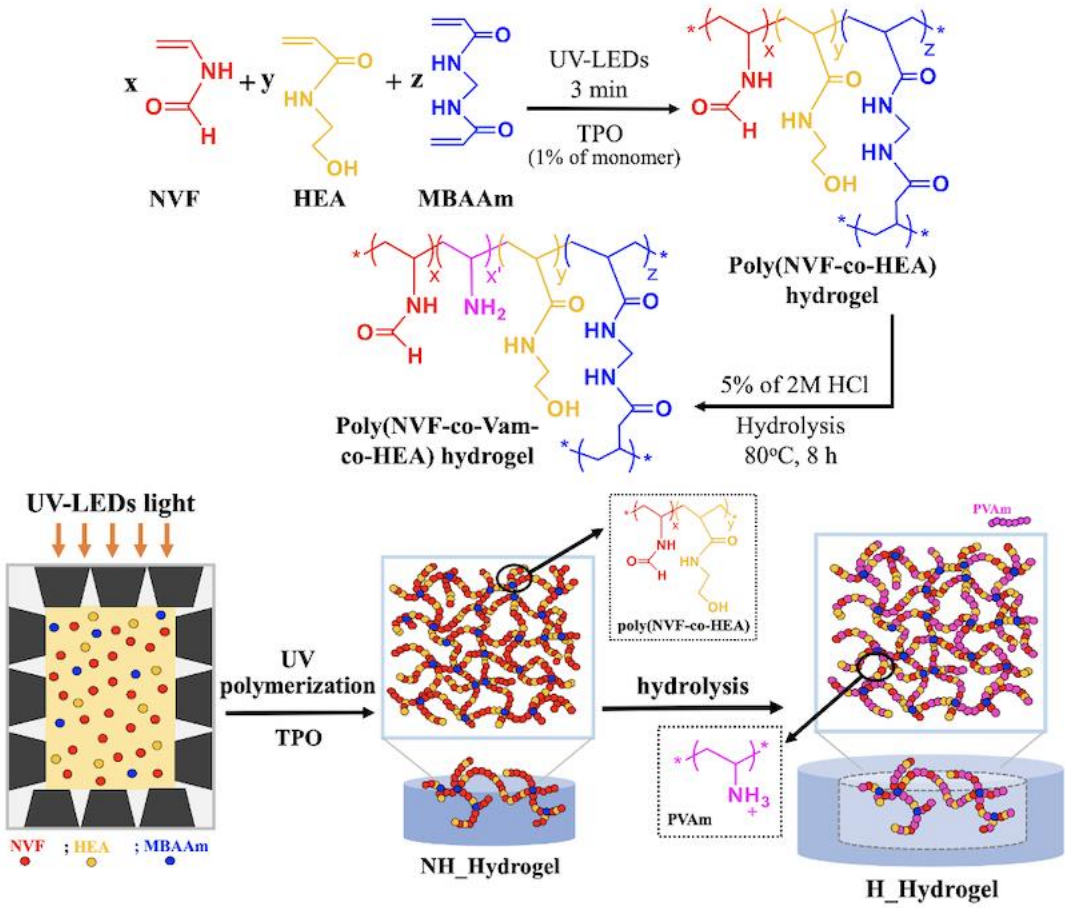


Figure 65 Schematic picture of preparation partway of poly(NVF-co-HEA) and poly(NVF-co-VAm-co-HEA) hydrogels

After finishing of preparation, the poly(NVF-co-VAm-co-HEA) hydrogels in the form of pH responsive hydrogel were immersed in ethanol for 1 minute and the washed with de-ionized water three times to remove any HCl solution. Then, the hydrolysed hydrogels were immersed into de-ionized water for 5 days (the water was changed every day).

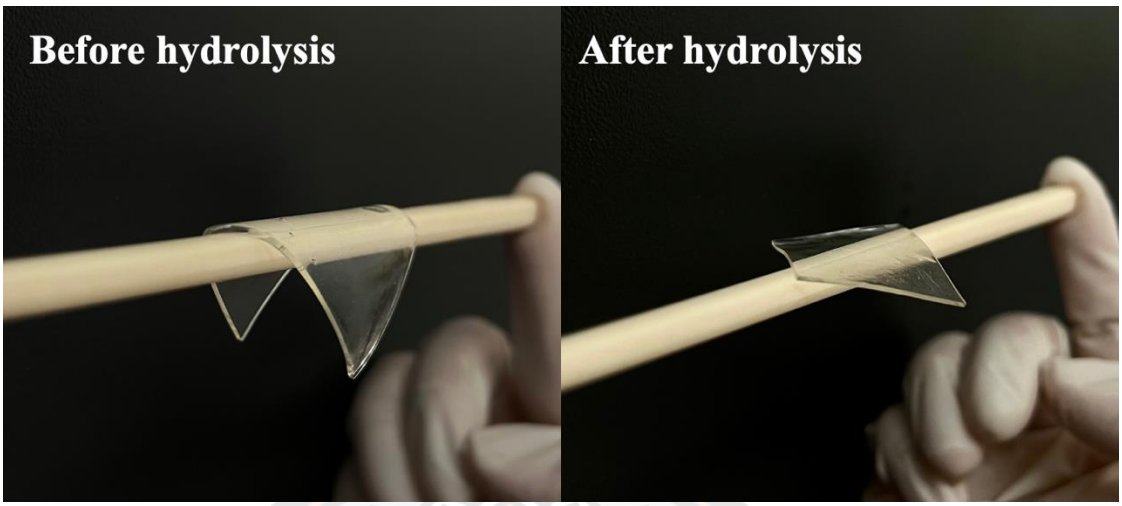


Figure 66 The physical appearance of poly(NVF-co-HEA) hydrogel ; (a) before hydrolysis and (b) after hydrolysis

For synthetic of hydrogels containing the cationic poly(vinylamine), Figure 66 shows the physical appearance of poly(NVF-co-HEA) hydrogel before hydrolysis (66a) and after hydrolysis (66b). The non-hydrolysed poly(NVF-co-HEA) hydrogel (66a) represents the hydrogels are slightly flexible materials, while the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel (66b) represents the hydrogels are more brittle and shows the swollen segment became larger before hydrolysis of hydrogels. This was achieved by producing hydrogels that is expected to contain the polymer vinylamine (VAm).

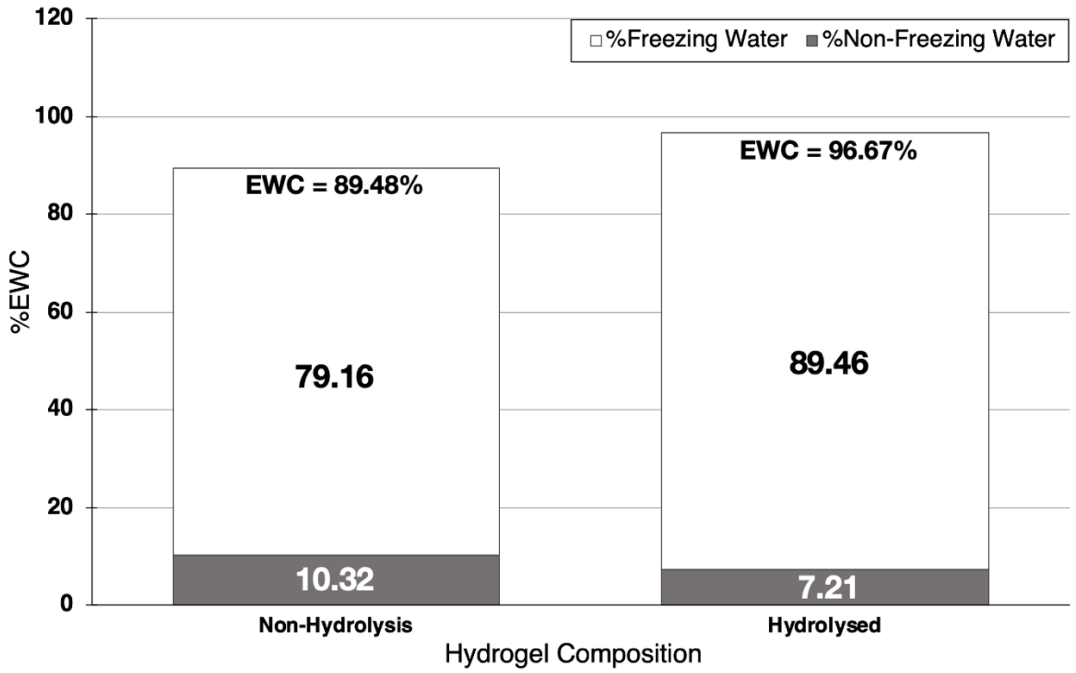
Characterization of NVF-copolymer hydrogels containing cationic poly(vinylamine)

In addition, the characterization of poly(N-vinylformamide) based materials via photopolymerization both hydrolysed and non-hydrolysed hydrogels for the pH responsive hydrogels were compared; with equilibrium water content (%EWC), state of water, tensile properties, the stability of hydrogels at difference time in water-ethanol and the swelling behaviors as a function of pH of hydrogels were studied.

1. Equilibrium Water Content (%EWC) and water structure within the hydrogels

To help confirm the production of hydrogels containing the polymer vinylamine (VAm) %EWC was measured, hydrophilic polymer can absorb varying amounts of water depending on the density of hydrophilic groups present in the polymer. The water bulk content was measured using the equilibrium water content (%EWC). The behavior of water molecules in the hydrogel can be further investigated using Differential Scanning Calorimetry (DSC), as water in hydrogels can exist as two separate types: freezing water (free water) and non-freezing water (bound water). Figure 67 compares the %EWC of the non-hydrolysed and hydrolysed hydrogels. The figure also presents how much of the water is classified as freezing or non-freezing measured using the method presented in chapter 3.

(a)



(b)

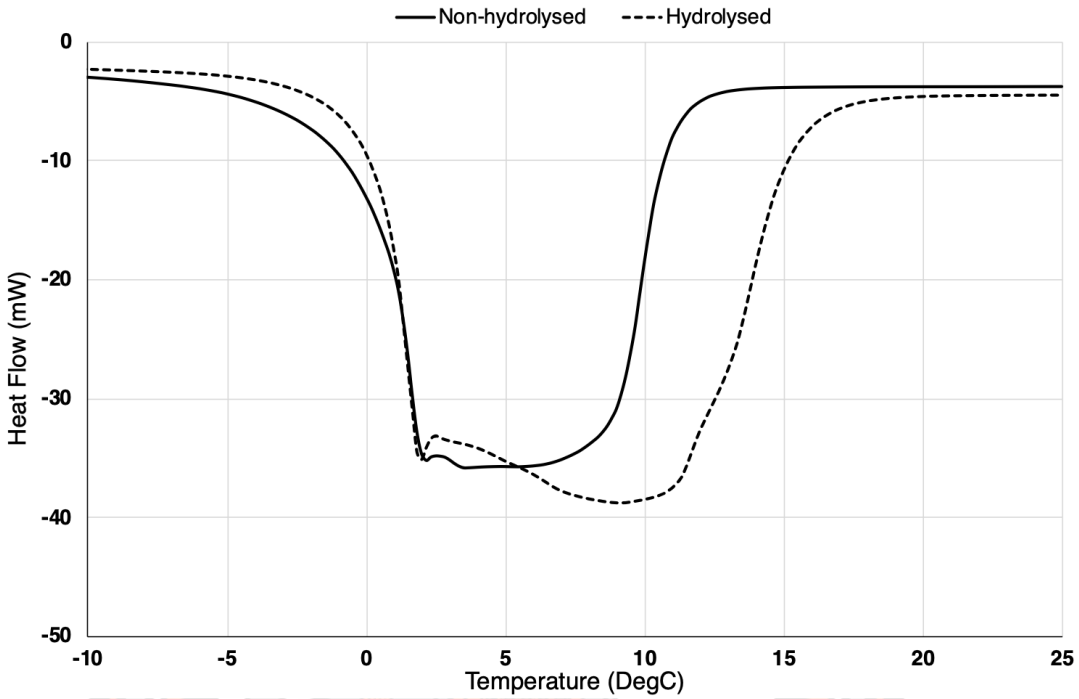


Figure 67 %EWC (a) and about of freezing and non-freezing water (b) of non-hydrolysed poly(NVF-co-HEA) hydrogel and hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel

The water content (%EWC) of each sample was observed and shown in figure 67a. The %EWC of hydrogels showed that non-hydrolysed and hydrolysed hydrogels was 89.48% and 96.67%, respectively. The hydrolysed hydrogels show the highest %EWC if 96.67% after hydrolysis of the hydrogel it is expected to contain the polymer vinylamine (VAm) due to the larger amount of water present in these hydrogels and also has more free water that has the ability to solubilize more hydrophilic species such as drug molecules in the hydrogels.

Figure 67b shows the endothermic peaks of the frozen water, the area under these peaks is the amount of freezing water in the hydrogel. This suggests there is some interaction between the functional groups of both polymers that enable more water to bind to the copolymer backbone. However, the intensity of freezing water peak increase after hydrolysis of hydrogel due to the water molecules of hydrogel

have the different behaviours in the hydrogel depending on the composition of functional groups present in the hydrogel structure, as these gels should contain polyvinylamine (PVAm) the interactions between water molecules and hydrogel structure are enhanced.

This result corresponds to the results from %EWC, when the water content increasing the freezing water is also increase for hydrolysed hydrogels. In summary, it was found that the hydrogel after hydrolysis was increase water content are due to the present of polyvinylamine (PVAm) as a cationic functionality to copolymerization and the amount of freezing water follows the same trend as %EWC.

2. Tensile properties

To studies the characteristic of hydrolysed hydrogels, mechanical properties are importantly to study for producing the pH responsive hydrogels. In this study, non-hydrolysed poly(NVF-co-HEA) hydrogels and hydrolysed poly(NVF-co-VAm-co-HEA) hydrogels were characterized for their tensile properties.

Table 20 Tensile properties of hydrogels

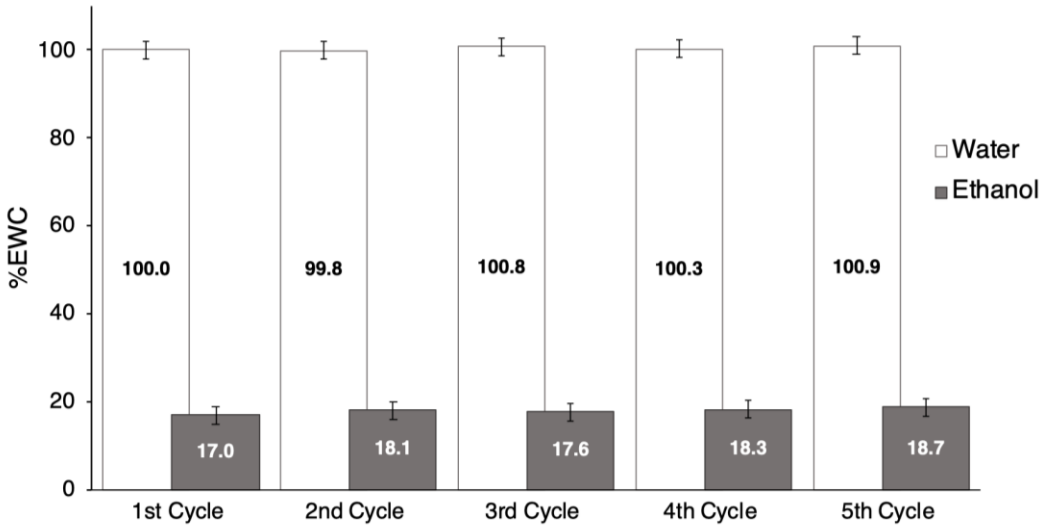
Samples code	% Elongation at break (%)	Tensile strength at break [MPa]	Modulus [MPa]
Non-hydrolysed hydrogel	103.0	27.0	26.20
Hydrolysed hydrogel	37.6	38.56	105.68

The %E of each sample was observed and is shown in Table 20. The %E of the non-hydrolysed and hydrolysed hydrogel was 103.0% and 37.6%, respectively. Comparing the non-hydrolysed and hydrolysed gels, we see that the hydrolysed hydrogel has a lower elongation at break, but a higher tensile strength at break and modulus, indicating that hydrolysis resulted in these changes. Therefore, the hydrolysed hydrogel should be used for applications that require a stringer but less flexible gel.

3. Stability of hydrogel over time in water and ethanol

The swellings of these hydrogels were also examined by first looking at their swelling by cycling between soaking in DI water and ethanol solutions and in different pH solutions. For this study, the stability of hydrogels in water and ethanol over time was observed. Non-hydrolysed poly(NVF-co-HEA) hydrogels and hydrolysed poly(NVF-co-VAm-co-HEA) were chosen for comparison in this study. The initial weight of the hydrogel samples was measured and then they were soaked in water for 24 hours, removed, and re-weighed, then the hydrogels were placed into ethanol for 24 hours and then the process was repeated for 5 cycles. The results show the water content (%EWC) of the non-hydrolysed and hydrolysed hydrogels over time in water and ethanol as shown in Figure 68.

(a)



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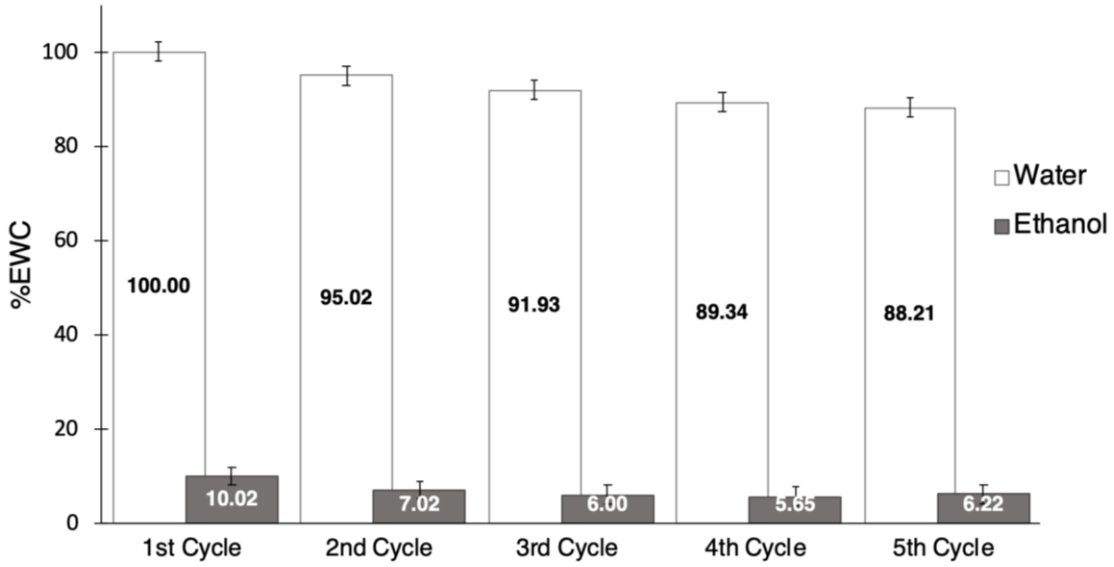

Figure 68 Stability of hydrogel over time in water and; (a) poly(NVF-co-VAm-co-HEA) hydrogel and (b) poly(NVF-co-HEA) hydrogel

The hydrogels showed that after adsorption reached its equilibrium in ethanol, all the samples became opaque and shrunken, which was a different result compared to when the hydrogels were soaked in water, where they became swollen and transparent upon reaching their equilibrium time. Figures 68a and 68b show the differences between the non-hydrolysed and hydrolysed hydrogels.

In Figure 68 the non-hydrolysed hydrogel swelling ratio is very consistent across the pH range (figure 68a), whereas the hydrolysed hydrogel shows varied swelling behaviors (figure 68b). There are significant reductions in the swelling ratio at basic pH's (10 and 12) and a slight reduction at acidic pH 2 and less so pH 4. When the solvent was cycled between water for 24 h and ethanol for 24 h again there are differences between the gels. Taking the EWC as 100% when the non-hydrolysed hydrogel was cycled between water and ethanol the results showed that the gel returns to EWC after being placed in water for 24 h and after being soaked in ethanol for 24 h had a water content that was around 16-18%. The hydrolysed hydrogel saw its water content reduced to 4-5% of it EWC when soaked in ethanol and this then failed to return to EWC of the gel at 24 h of soaking in water. Thus, upon cycling the overall

maximum EWC gradually reduced with the 4th cycle having 92% of the original water content may be due to the hydrolysed hydrogels need to do a long resoak to returns to EWC after being placed in water for 24 h. Therefore, both non-hydrolysed and hydrolysed hydrogels represent the ability of hydrogels to re-equilibrate in water and ethanol.

2. The swelling behavior as a function of pH

The swelling ratio of non-hydrolysed poly(NVF-co-HEA) and hydrolysed poly(NVF-co-VAm-co-HEA) hydrogels as a function of pH is shown in Figure 69. In this study, the hydrogel samples were kept in solution at room temperature until reaching equilibrium (5 days). As shown in the figure 69, the non-hydrolysed poly(NVF-co-HEA) hydrogel displayed a constant swelling behavior at any pH solution, reaching 100% EWC.

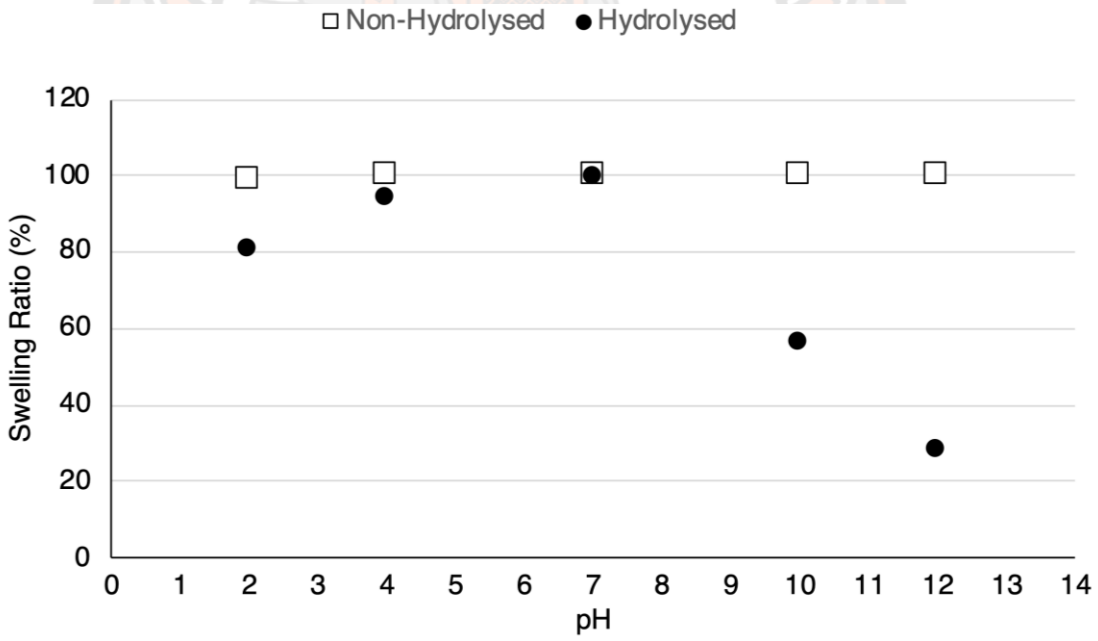

Figure 69 The swelling behavior as a function of pH of poly(NVF-co-VAm-co-HEA) hydrogel and poly(NVF-co-HEA) hydrogel

The results clearly shows that the non-hydrolysed poly(NVF-co-HEA) hydrogels swelling ratio remained very consistent over the different pH values. While

after hydrolysis, the EWC of hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel showed the maximum value at pH 7.0. The hydrolysed hydrogels showed the smaller swelling ratios at pH 2 and pH 12, when compared with non-hydrolysed poly(NVF-co-HEA) hydrogels are due to the presence of polyvinylamine (PVAm) as a cationic functionality in the hydrogel that behaves differently at different pHs.

It was found that the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel showed different swelling behavior at different pH values. At low pH (pH 2), electrostatic repulsive forces between like-charged regions may have reduced and collapses the polymer network. At high pH (pH 10 and 12), electrostatic interactions between oppositely charged regions may have occurred, along with the deprotonation of ammonia ions into amines in the polymer chain, causing the network to shrink. As a result, the water capacity decreased with further increases in pH (10, 12).

For summary of this section, these pH sensitive hydrogels were characterized by EWC, DSC, tensile properties and swelling behavior in both water and ethanol. The hydrogel swelling at different pHs was observed, non-hydrolysed hydrogels showed consistent swelling across the entire pH range but after hydrolysis, the hydrogels showed different behavior with reduced swelling ratios observed at acidic and basic conditions (pH2 and pH 10 and 12).

Next section, in our design of a controlled release system, dye and Lactobionic acid was incorporated as model ingredients.

***In vitro* study of releasing of the hydrogels**

1. **pH Responsive Release of dye solution**

For control drug release studies, each hydrogel was soaked in a dye solution of know concentration (0.0001M) for 96 hours, three dyes were used (Orange II sodium salt, crystal violet and Congo red). Table 21 shows information of organic dye that we used.

Table 21 Information of three surrogate dyes

Name	Dye type	MW	pKa
Orange II sodium salt	Anionic azo dye	350.32	8.26, 11.4
Crystal violet	Cationic dye	407.99	9.4
Congo red	*neutral-ionic azo dye*	696.68	4

The *in vitro* release profiles of dye solutions from the hydrogels at room temperature were studied in DI water solution at different pHs. Figure 70 shows the *in vitro* uptake of three dye into the non-hydrolysed and hydrolysed hydrogels at pH 7. The results show that, the colour of each gel changes considerably and dependent on dye incorporated as expected.

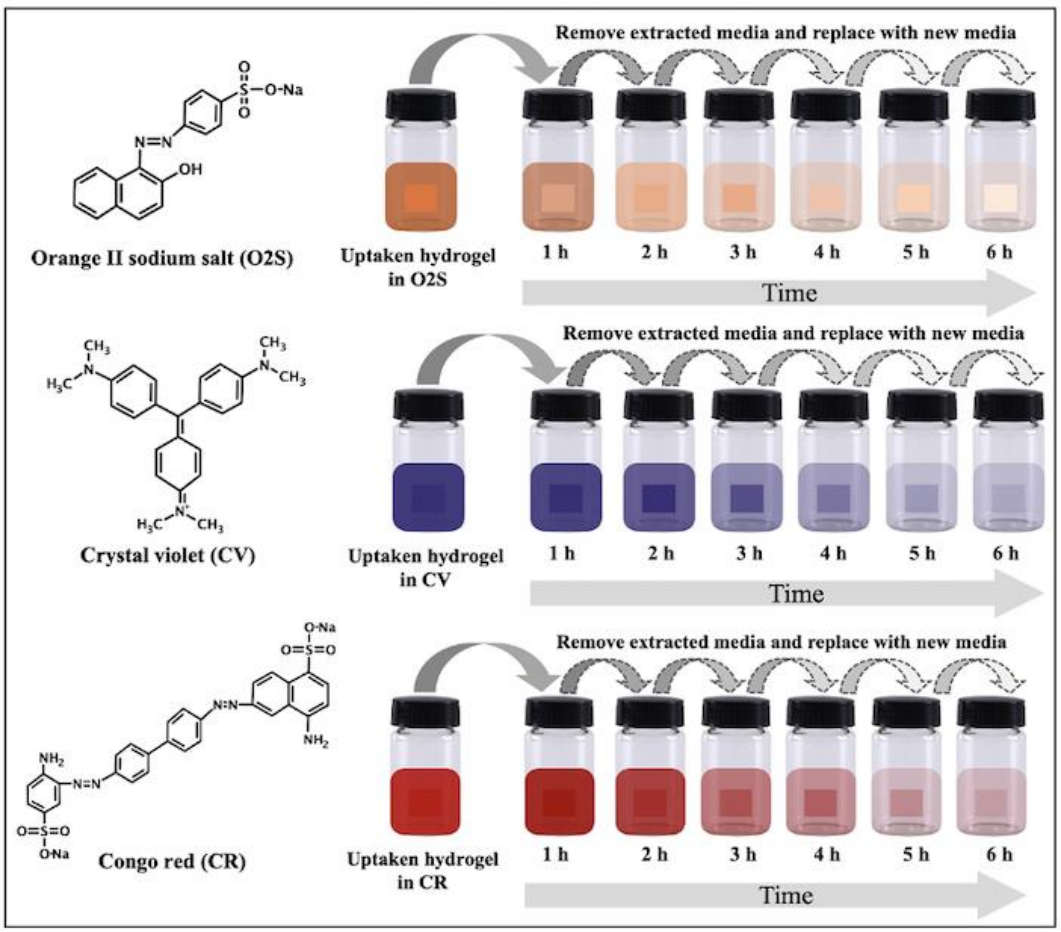

Figure 70 Digital photographs of non-hydrolysed (left) and hydrolsed hydrogels (right) in difference dye uptake; (a) Orange II sodium salt (O2S), (b) Congo red (CR) and (c) Crystal violet (CV) at pH 7 solution

The results showed that the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel had complete uptake when compared to the non-hydrolysed hydrogel with regards to the anionic dye, Orange II sodium salt. This is due to the formation of an ionic pair, with the presence of PVAm from the hydrogel having vinylamine as a cationic functionality, and the presence of many anionic groups from the Orange II sodium salt. Both the non-hydrolysed and hydrolysed hydrogels showed excellent dye adsorption capacity for CR dyes from their Congo red solution**.** Figures 71, 72 and 73 shows the cumulative amount (ug/ml) of dye release from the hydrogels in comparing the non-hydrolysed and hydrolysed hydrogels in DI water under various pH values. The release profiles of the hydrolysed hydrogels show the potential ability for pH responsive release due to hydrolysed poly(NVF-co-VAm-co-HEA) hydrogels which contained PVAm as a pH responsive functionality. In case of the non-hydrolysed hydrogels, the release profiles of these hydrogels did not show many differences under various pH conditions, apart from with Congo red at pH2 (Figures 72).

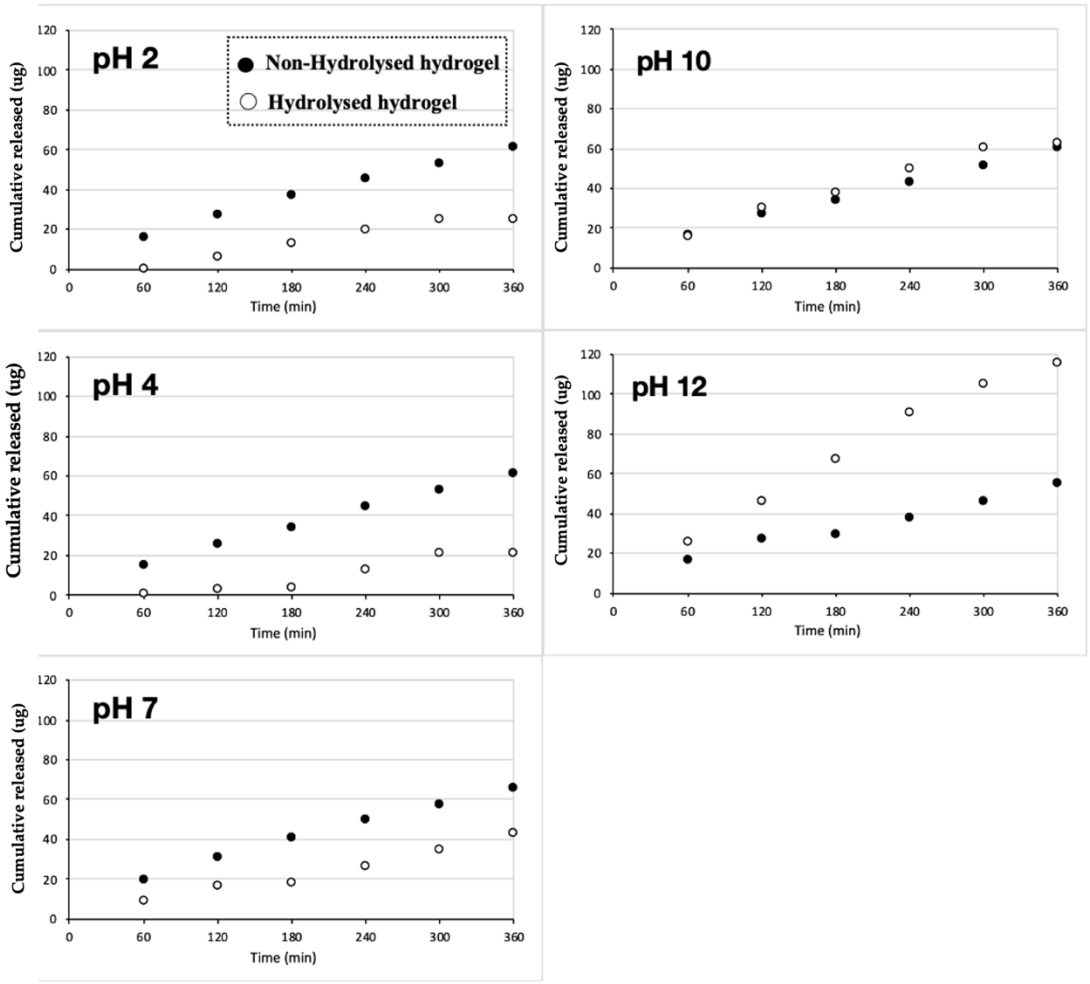

Figure 71 The comparison of release profiles of non-hydrolysed (black) and hydrolsed hydrogels (white) in O2S under various pH conditions

Figure 71 shows the results for O2S, when comparing the non-hydrolysed and hydrolysed gels, we see that the non-hydrolysed gel shows similar release at most pH values but with slightly less release at pH 12. The hydrolysed gel, however, this gel does show significant pH sensitivity.

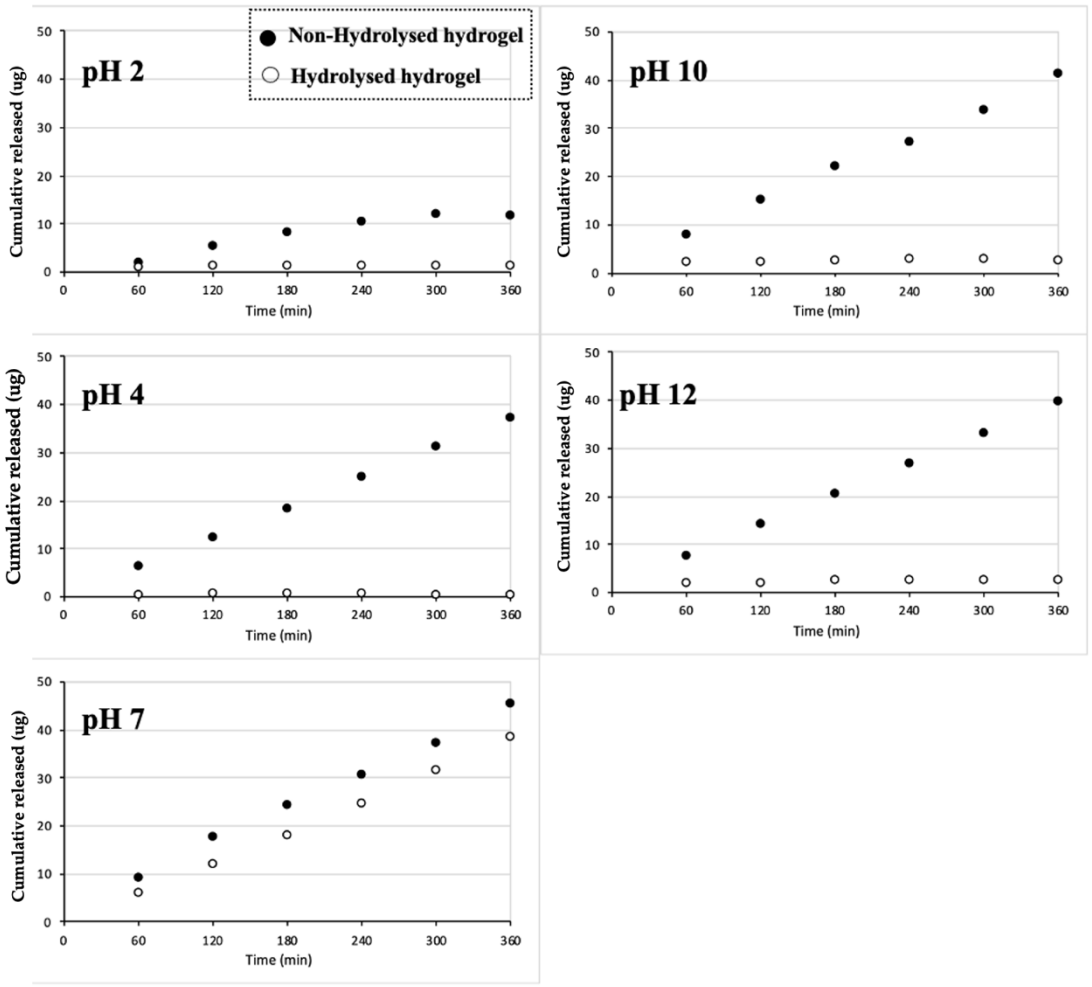

Figure 72 The comparison of release profiles of non-hydrolysed (black) and hydrolysed hydrogels (white) in CR under various pH conditions

Figure 72 presents the release of Congo Red, the results are quite different to that of the previous dye, with the non-hydrolysed gel releasing similar amounts apart from at the most acidic pH value (pH 2), which shows much lower release. For the hydrolysed gels the release at different pH values is much more pronounced, at both acidic and basic pH values showing very low release. Whereas, at neutral pH 7 the gel shows release.

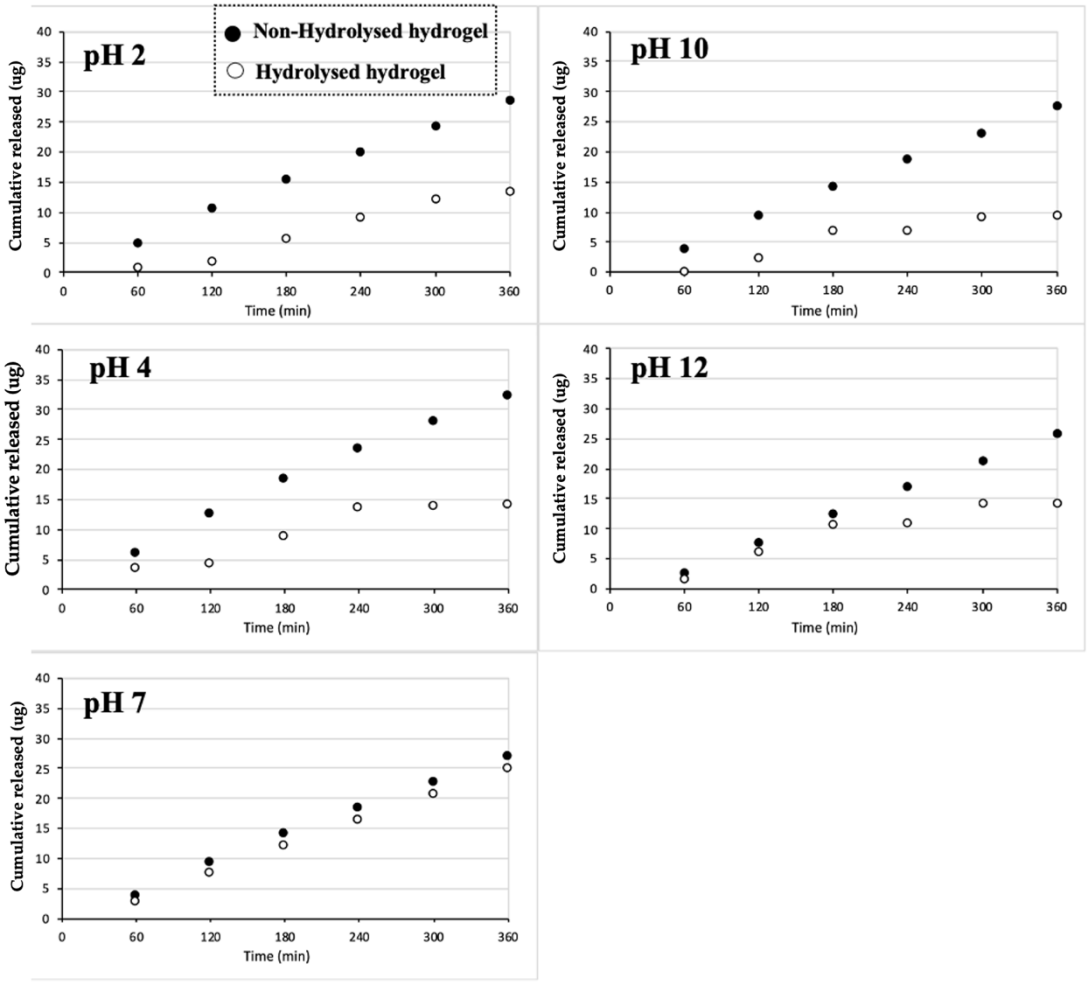

Figure 73 The comparison of release profiles of non-hydrolysed (black) and hydrolsed hydrogels (white) in CV under various pH conditions

Figure 73 shows the release of crystal violet, for the non-hydrolysed gels the release profile at different pH values shows lower release amounts as the pH of the system is increase to more basic values. The hydrolysed gel shows lower values of release at acidic and basic conditions with pH 7 showing the highest release. The release of crystal violet for both non-hydrolysed and hydrolysed is low.

Figures 74 show the amount of three surrogate dyes (O2S, CR and CV) released from both the non-hydrolysed and hydrolysed hydrogels over a 6 h time period at 5 different pH's (pH 2, 4, 7, 10 and 12).

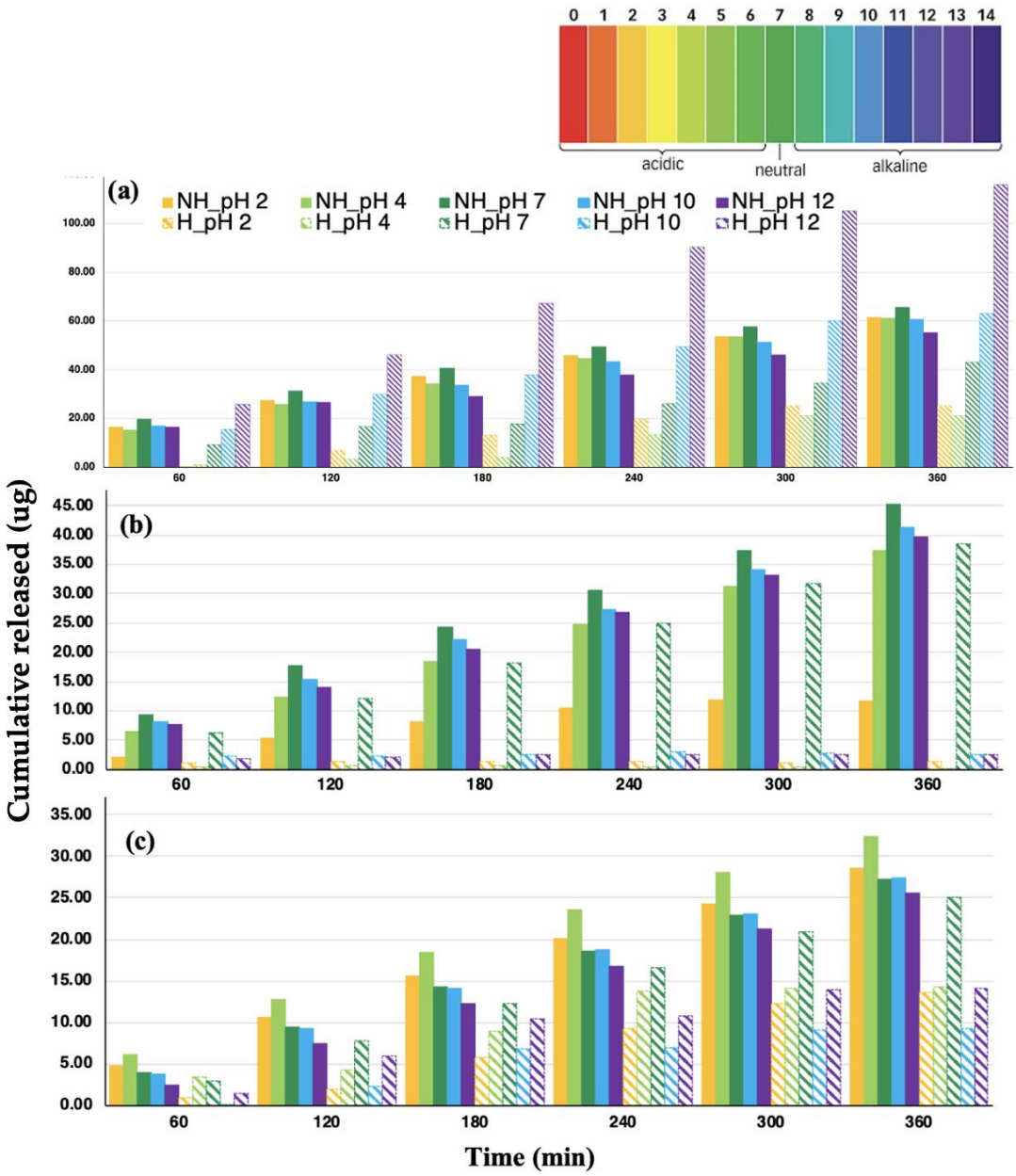

Figure 74 pH Responsive release profiles in difference dye; (a) Orange II sodium salt (O2S), (b) Congo red (CR) and (c) Crystal violet (CV) under various pH conditions

Figure 74a shows the results for O2S, when comparing the non-hydrolysed and hydrolysed gels. The differences in the release between the non-hydrolysed and hydrolysed hydrogels are the most pronounced with O2S dye, we see that the non-hydrolysed gel shows similar release at most pH values but with slightly less release

at pH 12. On the other hand, the hydrolysed hydrogels that contain PVAm demonstrate the potential for pH-responsive release, this hydrogel does show significant pH sensitivity. The release at acidic and neutral pH values is low while the release at the basic pH values (pH 10 and 12) shows greater release with pH 12 showing an amount released 60% higher than any other sample.

Figure 74b shows the release profiles of the hydrogels after uptake of Congo Red. There is a significant difference between the release of non-hydrolysed and hydrolysed hydrogels, especially for Congo Red among the dyes tested. The release profiles of non-hydrolysed hydrogels do not vary under different pH conditions, with only a minor volume change of Congo Red at pH 2. On the other hand, the hydrolysed hydrogels showed the lowest release of Congo Red, which may be due to the presence of amine groups in the non-hydrolysed hydrogels that interact with the amide groups of Congo Red. It was noted that the release from hydrolysed hydrogels was highest at pH 7. From previous studies, it was found that none of the homopolymers fully released Congo Red during the 6-hour release period, however, their release profiles, though low, are the most linear. These findings can be attributed to the inherent properties of Congo Red, which contains both sulfonic acid and amine groups, giving it the ability to interact with all three of the polymers present in the different gels.

Figure 74c shows the release profiles of the non-hydrolysed and hydrolysed hydrogels after uptake of crystal violet dye (CV). The values are similar for the non-hydrolysed and hydrolysed hydrogels. In the case of the non-hydrolysed hydrogels, the release profiles of these hydrogels show no difference under various pH conditions. On the other hand, the hydrolysed hydrogels containing PVAm show potential for pH-responsive release. The results indicate that the highest amount of release is observed for hydrolysed hydrogels at pH 7.

2. pH Responsive Release of an active ingredient

To investigate the pH-responsive release behavior of an active ingredient from the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel, Lactobionic acid was used as a model ingredient. Lactobionic acid is a well-known whitening agents used in skin lightening cosmetic products. A 5% w/w solution of Lactobionic acid was incorporated into the hydrogels. The cumulative percentage of Lactobionic acid release from the hydrogels at different pH values is shown in Figure 75.

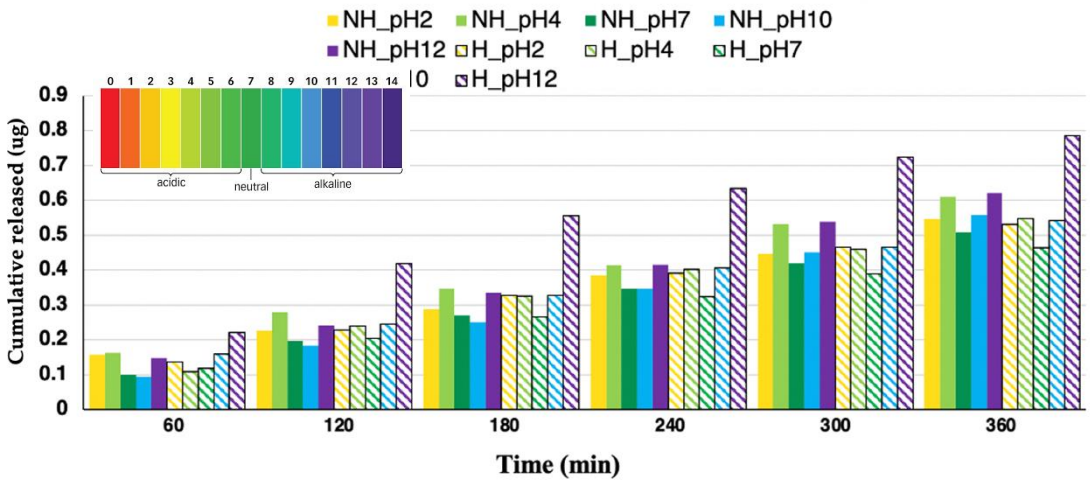

Figure 75 pH Responsive release profiles of an active ingredient; Lactobionic acid at different of pH

At low pH conditions (pH 2 and 4), only small amounts of Lactobionic acid was released from both the non-hydrolysed and hydrolysed hydrogels, due to the collapse of the hydrogel network. The release profile of these hydrogels shows a slight increase when the pH is increased. When the active ingredient was incorporated with the hydrogel and applied to human skin (pH 6-7), the increase in the surrounding pH might lead to the release profile of the active ingredient from the hydrogel material.

Interestingly, the highest amount of release is seen at the pH of 12, this is perhaps from the reduction in the hydrogel network at this pH with the hydrolysed gel.

Summary : Synthesis of the NVF-copolymer hydrogels containing the cationic poly(vinylamine)

All the results show that the functionalised hydrogel was successfully prepared to form the poly(NVF-co-VAm-co-HEA) hydrogel synthesized in this study have the potential to be used as a smart carrier for cosmetic ingredients triggered by an external pH change for cosmetic applications and this hydrogel exhibits pH dependent behavior.

CHAPTER V

CONCLUSIONS

This thesis focuses on developing a technology platform for producing novel cosmetic hydrogels that incorporate cosmetic active agents. The hydrogels were fabricated from Poly(N-vinylformamide) and copolymers through photopolymerization to create cosmetic hydrogel patches. To guide the selection of suitable properties for cosmetic hydrogels, a series of commercial cosmetic hydrogel masks were first studied. The PNVF-based materials were then evaluated and compared with the commercial products. The study also evaluated the impact of varying the ingredient ratio in the formula for producing PNVF materials and NVF-copolymer hydrogels. To prepare pH-responsive hydrogels and assess their potential use as delivery carriers for cosmetic active ingredients, hydrogels were synthesized through UV-LED photopolymerization using a copolymer of poly(NVF) and hydroxyethyl acrylamide (HEA) as monomers. The results of pH-responsive release provide insight into the active delivery properties and help identify the best conductive cosmetic active ingredients.

For approach I, the hydrogels have been designed to produce cosmetic hydrogel patches. We first characterized the effects of commercial cosmetic products to determine the most suitable options to pursue further. The work has been separated into three parts to find the most suitable options for the production of PNVF-based materials and copolymer hydrogels: (1) preparation of poly(NVF)-copolymer hydrogels, (2) development of hydrogels for novel cosmetic hydrogel patch applications, and (3) comparison of the novel cosmetic hydrogel patches with commercial cosmetic products.

The first part of this study focused on the preparation of poly(NVF)-copolymer hydrogels, which depended on several factors such as the initiator system, monomer, crosslinker type, and crosslink density. These factors were crucial in controlling the hydrogel polymerization and the final properties of the hydrogels. The

physical properties of hydrogels prepared using three different initiation systems were compared, including thermal initiation, UV-A initiation, and UV-LEDs initiation. Both photo-initiation methods gave very similar results for the %EWC (equilibrium water content), tensile properties, and contact angle. However, there were some differences observed in the thermally cured hydrogels due to the longer polymerization time required for this process. The UV-LEDs method was found to be the most energy-efficient and environmentally friendly method for producing hydrogels compared to thermally initiated hydrogels.

The study also investigated a series of Poly(N-vinylformamide) (PNVF) and various comonomers of two hydrophilic monomers, N-hydroxyethyl acrylamide (HEA) and 2-Carboxyethyl acrylate (CEA). The highest %EWC was observed when using 0% copolymer loading, i.e. the poly(NVF) homopolymer, due to the larger amount of water present in these hydrogels, which may have led to more free solubilized drug in the hydrogels. In contrast, 100PCEA presented with the lowest %EWC. Samples of poly(NVF) copolymerized with HEA presented a decrease in %EWC values as the HEA content increased. When comparing the surface of the hydrated hydrogels, increasing the ratio of either HEA or CEA led to an increase in the contact angle of the gel towards the higher angle of the homopolymer. For controlled drug release studies, each hydrogel was soaked in a dye solution of known concentration, using three surrogate dyes (Orange II sodium salt, crystal violet, and Congo red). The release profiles of the homopolymers showed that 100PNVF released a higher amount of dye than the other polymers for each of the three dyes. This could be due to 100PNVF processing the highest water content as well as high freezing water content (free water). In contrast, 100PCEA gave the lowest release across the range of dyes. CEA showed the most differences in both the release profiles and also the color of the gels before and after release, likely due to CEA being an anionic hydrogel and the dye being strongly held in the hydrogel structure through charge attraction. The release profiles of the copolymers showed that as the amount of either HEA or CEA increased (25 or 50%wt), the amount of orange II sodium salt (O2S) decreased after dye uptake. The differences in the release between poly(NVF-co-HEA) and poly(NVF-co-CEA) hydrogels were the most pronounced for crystal violet (CV), with CEA containing a carboxylate group that could strongly interact with CV

structure. In contrast, HEA contained a hydroxyl group that could interact with the amide groups of CV but not as intensely. However, Congo red (CR) gave the lowest release of all the tested dyes, and the release profiles, although low, were also the most linear, likely due to the inherent properties of CR containing both sulfonic acid and amine groups, allowing it to interact with all three of the polymers present in the different gels.

For the second part, to development of hydrogels for novel cosmetic hydrogel patches application. The fabrication and characterization will be presented into three parts; (1) Effect of cross-linker on hydrogel patches, (2) Effect of Photo-initiator on hydrogel patches (3) Effect of glycerin and preservative on hydrogel patches.

In the section on the effect of crosslinkers on hydrogel patches, the results will be discussed and compared among three crosslinkers at different concentrations ranging from 1.0 to 4.0 %w/w, namely Di(ethylene glycol) diacrylate (DEGDA), Poly(ethylene glycol) diacrylate (PEGDA), and N,N-methylenebisacrylamide (MBAAm). While all hydrogel samples were successfully fabricated, only some were suitable for the desired application as cosmetic patches due to being brittle, weak gel, or non-adhesive to the skin. The highest %EWC was 93.02% when using MBAAm as the crosslinker at a concentration of 0.1%, which can be explained by its hydrophilic nature. However, the %EWC of 1%PEGDA 75PNVF25HEA and 0.5%MBAAm 75PNVF25HEA showed similar values of 88.95 and 88.92, respectively. Although both samples had similar hydrogel texture, for industry processing purposes, 0.5%MBAAm 75PNVF25HEA had the best results in terms of cost processing, which can produce hydrogels at a fraction of the cost to both the environment and industry compared to high concentrations of crosslinker. Importantly, all these values were under 45, and therefore, all hydrogel samples were defined as super hydrophilic.

In the section on the effect of photo-initiators on hydrogel patches, the different concentrations of photo-initiator gave similar results in terms of %EWC and wettability, as well as texture of hydrogels. However, in terms of industry processing concerns, using lower concentrations of photo-initiator at 0.5%w/w was by far more cost-effective, producing hydrogels at a fraction of the cost to both the environment and industry compared to high concentrations of photo-initiator. Therefore,

1%PEGDA 75PNVF25HEA-0.5%PI, 0.5%MBAAm 75PNVF25HEA-0.5%PI, and 0.1%MBAAm 75PNVF25HEA-0.5%PI were chosen for further study.

In the section on the effect of glycerin and preservative on hydrogel patches, we discussed and compared the effect of skin moisturizers and preservative loadings on the properties of the gel to find the most suitable hydrogel patches for cosmetic applications. To produce cosmetic hydrogel patches, we used 10%w/w glycerin as a skin moisturizer and 0.5%w/w phenoxyethanol as a preservative. The results showed that glycerin and phenoxyethanol did not affect the %EWC and wettability of the hydrogels. However, to confirm the most suitable applied gel properties for cosmetic hydrogel patches, we needed to study the mechanical properties. Comparison with 1%PEGDA 75PNVF25HEA-0.5%PI and 0.5%MBAAm 75PNVF25HEA-0.5%PI showed that when using 0.5%MBAAm as a crosslinker, it gave a higher %E than 1%PEGDA of crosslinker and lower tensile strength at break and modulus. However, for applied hydrogels, the modulus of 0.5%MBAAm hydrogel was lower than that of 1%PEGDA, indicating good elastic modulus hydrogels. Therefore, we were interested in studying %E at different concentrations of MBAAm. We found that these hydrogels showed an increase in %E when a lower amount of MBAAm was used. At 0.1% MBAAm, the tensile stress and modulus value of hydrogels decreased with decreasing MBAAm. High %E, high tensile stress, and low modulus value of hydrogels are required. Therefore, we selected the sample of 0.1%MBAAm 75PNVF25HEA-0.5%PI hydrogel for comparing the effect of glycerin and phenoxyethanol loading to produce the hydrogels. The results showed that higher tensile strength and elongation at break followed by a lower modulus were the result of adding glycerin. These results suggested that the dramatic improvement of %E of hydrogels was promoted by the addition of glycerin into the hydrogel synthesis system, which enhances the gel flexibility. It is possible to believe that this improvement in %E and elastic modulus is useful for the applied gel properties of hydrogel patches. In summary, the most suitable applied gel properties for cosmetic hydrogel patches are 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels, using 0.1% of MBAAm as a crosslinker based on 75% of PNVF co 25% of HEA with glycerin and phenoxyethanol loading via free-radical polymerization via UVLEDs initiation system with diphenyl(2,4,6-trimethylbenzoyl) phosphine oxide used as the

photo-initiator. So, we investigated the applied gel properties of 0.1%MBAAm 75PNVF25HEA-0.5%PI-Ad hydrogels in terms of in-vivo bio-adhesion ability and skin irritation after application. We evaluated the ability of patch adhesion by adhering it to the forearm of the volunteer for 30 minutes. The result was successful in adhesion, with no slippage of the hydrogels from the applied side. Additionally, no signs of skin irritation were observed during this study, indicating that the developed hydrogels for novel cosmetic hydrogel patches applications are mild to the skin.

For the third part, to compare the novel cosmetic hydrogel patches with commercial cosmetic products, 0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogels were chosen as the most suitable cosmetic hydrogel patch. The results of the comparison with all commercial cosmetic products and 0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogels showed that our hydrogels gave similar results in %EWC and wettability, with high %EWC and low contact angle of a liquid on a solid surface. Considering the requirement of good mechanical properties, such as high %E, high tensile stress, and low modulus value of hydrogels, the 0.1% MBAAm 75PNVF25HEA-0.5% PI hydrogel showed good results in terms of applied gel properties for cosmetic hydrogel patches. For *in vivo* study, thirty healthy female volunteers were asked to complete a Self-Perception Questionnaire (SPQ) and report which hydrogels felt more relaxed, more cooled, more hydrated, softer, and adhesive after 30 minutes of applying the hydrogel patches. Faith in Face (commercial cosmetic products) received the highest overall satisfaction score of the study subjects after one application (T30), at 94%. The study subjects felt that their skin was more relaxed after treatment, at 92.67%, and they also felt that their skin was more cooled (84%), more hydrated (89.33%), softer (93.33%), and adhesive (90.67%). In contrast, 0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogel showed an overall satisfaction score of 89.37% of the study subjects after one application. The study subjects felt that their skin was more relaxed after treatment, at 91.72%, and they also felt that their skin was more cooled (89.69%), more hydrated (84.01%), softer (96.74%), and adhesive (84.7%).

In summary, all instrumental results were confirmed by the subjects' perception in the Self-Perception Questionnaires. The results showed that the

0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogel in this study is an effective product to improve skin hydration, more cooled, more relaxed, softer, and adhesive of hydrogels. Moreover, the report highlighted that the 0.1% MBAAm 75PNVF25HEA-0.5% PI-Ad hydrogel received an overall satisfaction score of 89.37%. Therefore, this hydrogel patch is satisfactory for use as a hydrogel patch mask product. Although there is still some room for improvement which could be achieved by altering the hydrogel composition.

Overall, the aim of Approach II was to synthesize pH-responsive hydrogels containing cationic poly(vinylamine) (PVAm) by partially hydrolyzing the amide group of poly(NVF-co-HEA) under acidic conditions, for potential use as delivery carriers for cosmetic active ingredients. The hydrolysed hydrogels were successfully synthesized through UV-LEDs photopolymerization, using a copolymer of 75% poly(NVF) and 25% hydroxyethyl acrylamide (HEA) as monomers, and 0.5% N,N-methylenebisacrylamide (MBAAm) as a crosslinker. The hydrolysed hydrogels showed the highest %EWC at 96.67% after hydrolysis, and the state of water corresponded to the results from %EWC. As the water content increased, the freezing water content also increased for the hydrolysed hydrogels. It was found that the increase in water content after hydrolysis was due to the presence of cationic functionality, PVAm, in copolymerization, and the amount of freezing water followed the same trend as %EWC. To study the applied gel properties, both non-hydrolysed and hydrolysed hydrogels were tested for their ability to re-equilibrate in water and ethanol for the stability of the hydrogels. Non-hydrolysed hydrogels showed consistent swelling across the entire pH range, but after hydrolysis, the hydrogels showed different behavior with reduced swelling ratios observed at acidic and basic conditions (pH 2, 10, and 12).

For the controlled drug release studies, pH-responsive release of three surrogate dye solutions and an active ingredient were investigated. The results showed that the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel had a superior uptake ability compared to the non-hydrolysed hydrogel with regards to the anionic dye Orange II sodium salt. However, the release profile of the active ingredient from the hydrogels did not differ significantly under various pH conditions for the non-

hydrolysed hydrogels. On the other hand, the hydrolysed hydrogels containing PVAm demonstrated the potential for pH-responsive release. The results indicate that the amount of release from hydrolysed hydrogels was highest at pH 12. For the release of CR dye, the release profiles of non-hydrolysed hydrogels did not vary under different pH conditions, with only a minor volume change of Congo Red at pH 2. However, the hydrolysed hydrogels showed the lowest release of Congo Red, which may be due to the presence of amine groups in the non-hydrolyzed hydrogels that interact with the amide groups of Congo Red. The release from hydrolysed hydrogels was highest at pH 7. In the case of the release of CV dye, the values were similar for the non-hydrolysed and hydrolysed hydrogels. The release profiles of the non-hydrolysed hydrogels did not show any difference under various pH conditions. However, the hydrolysed hydrogels containing PVAm showed potential for pH-responsive release. The results indicated that the highest amount of release was observed for hydrolysed hydrogels at pH 7.

To investigate the pH-responsive release behavior of an active ingredient from the hydrolysed poly(NVF-co-VAm-co-HEA) hydrogel, 5% w/w Lactobionic acid was used as a model ingredient. In low pH conditions (pH 2 and 4), only small amounts of Lactobionic acid were released from both the non-hydrolysed and hydrolysed hydrogels due to the collapse of the hydrogel network. The release profile showed a slight increase when the pH was increased. When the active ingredient was incorporated with the hydrogel and applied to human skin (pH 6-7), the increase in the surrounding pH might lead to the release of the active ingredient from the hydrogel material.

In summary, all the results showed that the functionalized hydrogel was successfully prepared. The poly(NVF-co-VAm-co-HEA-) hydrogel synthesized in this study has the potential to be used as a smart carrier for cosmetic ingredients triggered by an external pH change for cosmetic applications and this hydrogel exhibits pH some dependent behavior.

Recommendations

1. This work will be further studied by continuing *in vivo* clinic testing at COSNAT, Naresuan University to observe skin irritation.
2. To confirm the amount of active releasing from the hydrogels, skin permeability studies of cosmetic ingredients will be further studied using Franz diffusion cell and human epidermis. The amount of cosmetic ingredients concentration diffused through the patch and then accumulated in the receptor medium can be analyzed by HPLC.
3. For the long-term research, stability studies of the hydrogel patch containing cosmetic ingredient should be studied further.
4. The amount of hydrolysis of hydrolysed hydrogels can be further calculated.
5. To confirm the %EWC of hydrogels, calculate the %EWC in air will be further studied.
6. To confirm the network stability testing of hydrogels, the time of %EWC should be increase for 48 hrs. for studied.

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