CHAPTER 1
INTRODUCTION

1.1. Skin and Activities of the Skin [1-2]

Skin forms the largest organ of the body, accounting for about 16 percent of a person′s weight**.** It performs many vital roles as both a barrier and regulating influence between the outside world and the controlled environment within our bodies. It is very important as it covers and protects everything inside body, such as bones, muscles and organs. It also keeps body at just the right temperature and, through nerve endings close to the surface of the skin, allow us to have the sense of touch. However skin can become infected with bacteria, virus and fungi, and can be irritated by chemicals or other substances.

The skin not just only gives us our appearance and shape, it also serves other important functions:

1. The physical toughness of the skin prevents the ingress of harmful chemicals and invading organisms such as bacteria and viruses. It also provides resistances to shocks for the more sensitive tissue underneath.

2. Skin could warn of harmful extremes of temperature or of other dangers on the other touch can be one of the most powerfully soothing and pleasurable of sensations.

3. Skin regulates body temperature because considerable heat could be transferred through the skin. Even under extreme conditions of high temperature and exercise, our skin tends to make body temperature normal. The internal body temperature (core temperature) needs to be kept constant for normal physiological activity to take place (37°C). It needs to maintain a core temperature for homeostasis.

4. Waste product and excess salt can be excreted from the body through sweat in form of solution. Also, water is lost continuously from our bodies through the skin by evaporization.

5. Skin is the main site of manufacture of vitamin D, which is essential for the growth and maintenance of our bones. The extensive network of nerves within the skin feeds information constantly to the brain concerning our surrounding.

1.2. The Structure of the Skin

The skin composes of three main structural layers as shown in Fig.1.1 [3-5].

- Epidermis

The epidermis is on the outer layer. It serves as a protective barrier and contains no blood vessels. Nourishment is supplied by diffusion of nutrient and essential substances from the dermis layer. The approximate thickness is 0.1 mm and on the palms and soles it is 0.8 to 1.4 mm.

- Dermis

This layer is approximately 1-2 mm thick and is situated immediately below the epidermis. It is composed of a tough, supportive, connective tissue matrix and contains specialized structures. The thin upper layer known as the papillary dermis is composed of loosely inter woven collagen. Next to the deeper layer, thick reticular dermis contains coarser and horizontally running bundles of collagen. The dermis is composed of collagen fibres which impart strength and toughness to the structure.

- Subcutaneous Tissue

There is a layer of fat underneath and in the lower regions of the dermis. The thickness of this layer varies depending on the place in the body and from person to person. This layer consists of spongy connective tissue interspersed with energy-storing fat cells, the adipocytes. Fat cells are grouped together and held in place by collagen fibers in the form of tissue septa. The subcutaneous tissue stores nutrients. It

is useful to the body as insulation and it can be used for energy when the intake of nutrients is insufficient. Being heavily interlaced with blood cells, any kind of injury causes heavy bleeding.

A burn injury is the first and foremost an injury to the skin. Although the burned patient has many problems to face during the various stages of recovery, the major and persisting problems for survivors are those associated with the problems of healing. Burns may be caused by heat (thermal burns), chemical cauterizing agents (chemical burns) or electricity (electrical burns). Most people believe that the medical definition of a burn has more to do with the depth of damage rather than the actual cause [6-8]. These various degree of burns are classiflied in Table 1.1.

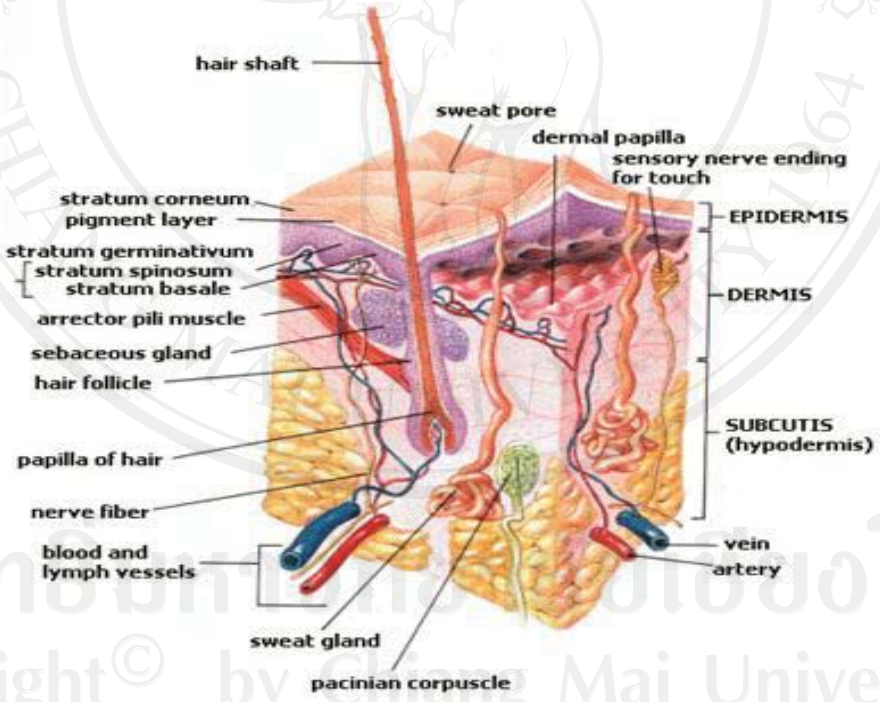


Figure 1.1. The layers and structure of the skin [6].

Table 1.1. Descriptions of the various burn ratings by degree [9-10].

Degree of burn	Descriptions
First-degree burn	Superficial injury involving only the epidermis. Characterized by pain, red skin without blistering, and perhaps swelling. This is a mild partial thickness burn that will heal without scarring.
Second-degree burn	Burn extends from epidermis into the dermis layer. Pain is intense, skin surface will turned to red, blistered, and may have a mottled appearance. This is a partial thickness burn that generally leaves minimal scarring if properly treated.
Third-degree burn	This burn involves destruction of the epidermis and dermis and may extend deep down into fat, muscle, or tissue. This degree of burn give no pain because the nerves have been destroyed. This is a full-thickness burn that may require skin grafting.

1.3. Synthetic Hydrogels and Fabrication

Hydrogels are three dimensional crosslinked hydrophilic polymer networks [11]. They possess the ability to imbibe large quantities of water or biological fluids without dissolving [12]. Hydrophilicity of these gels is attributed to the presence of hydroxy, carbonyl, amide or sulfonic groups along the polymer chain. Crosslinks are formed by covalent bonds, electrostatic, hydrophobic or dipole-dipole interactions prevent the gel from dissolving in any of solvents. The structural and physical integrity of the gels are a result of these interactions. Many different chemical structures can classified as hydrogels. A number of such polymeric compounds are illustrated in Table 1.2. [13-14]. Hydrogels are fabricated into useful forms using a variety of techniques. The specific technique used is primarily dependent upon the nature of the polymer.

However, the objectives in all cases are the same, for example, to prepare a hydrophilic polymer which maintains its integrity in water, which is in precise geometic configuration for the required application and which has sufficient mechanical strength and appropriate compliance to make it useful for that particular application. Hydrogel can be prepared by various methods which are illustrated schematically in Fig. 1.2.

• Cast films

Cast films frequently used as coatings for other materials, usually require non covalent forces (e.g., hydrophobic bonding) to prevent dissolution of the polymer in aqueous media. However, in many cases, films can be covalently crosslinked after casting.

• Crosslinked networks

Crosslinked networks are generally formed by chemical reaction between monomer, crosslinking agent and initiator in the solution. Later the solution becomes gel.

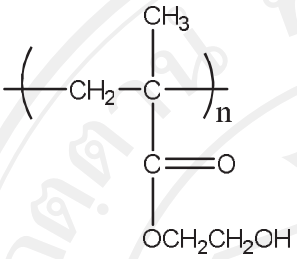
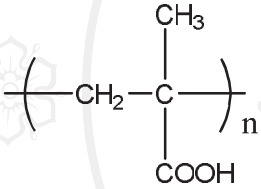
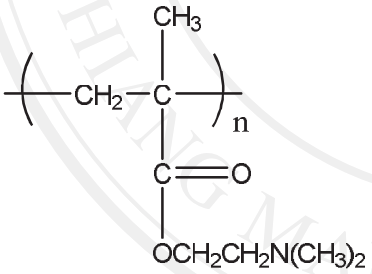
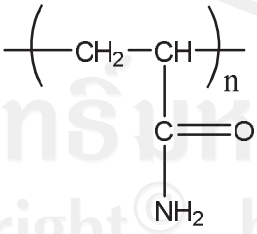
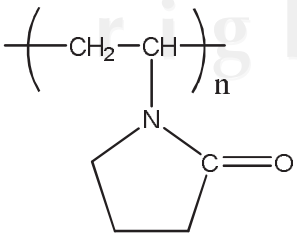
• Surface grafted polymer

The surface grafting of hydrogels to another polymer is attractive as a fabrication technique for a number of reasons, the generally low mechanical strength of hydrogels is improved by bonding to a mechanically stronger polymer, and fabrication is simplified since only the substrate need to be formed into a specific shape. Then, grafting be able to follows the contours of the substrate polymer [15-17].

• Interpenetrating network

Crosslinked hydrogel networks can also be formed within other polymer networks. Both networks are topologically independent and inseparable. This interpenetrating polymer network (IPN) approach represents another technique for mechanically strengthening hydrogel systems [18-19]. Finally, hydrogels can be formed as uniform microspheres in sizes ranging from 0.3 μm to 3.4 μm [20].

Table 1.2. Hydrogel chemical structures [13].

Structure	Name	Hydrogel fabrication
	Poly(2-hydroxyethyl methacrylate)	- Cast - Chemical crosslinked - Surface grafted - IPN
	Poly(methacrylic acid)	- Chemical crosslinked - Surface grafted
	Poly(*N,N′* dimethyl-aminoethyl methacrylate)	- Chemical crosslinked - Surface grafted
	Poly(acryamide)	- Chemical crosslinked - Surface grafted - IPN
	Poly(*N*-vinyl pyrrolidone)	- Chemical crosslinked - Surface grafted

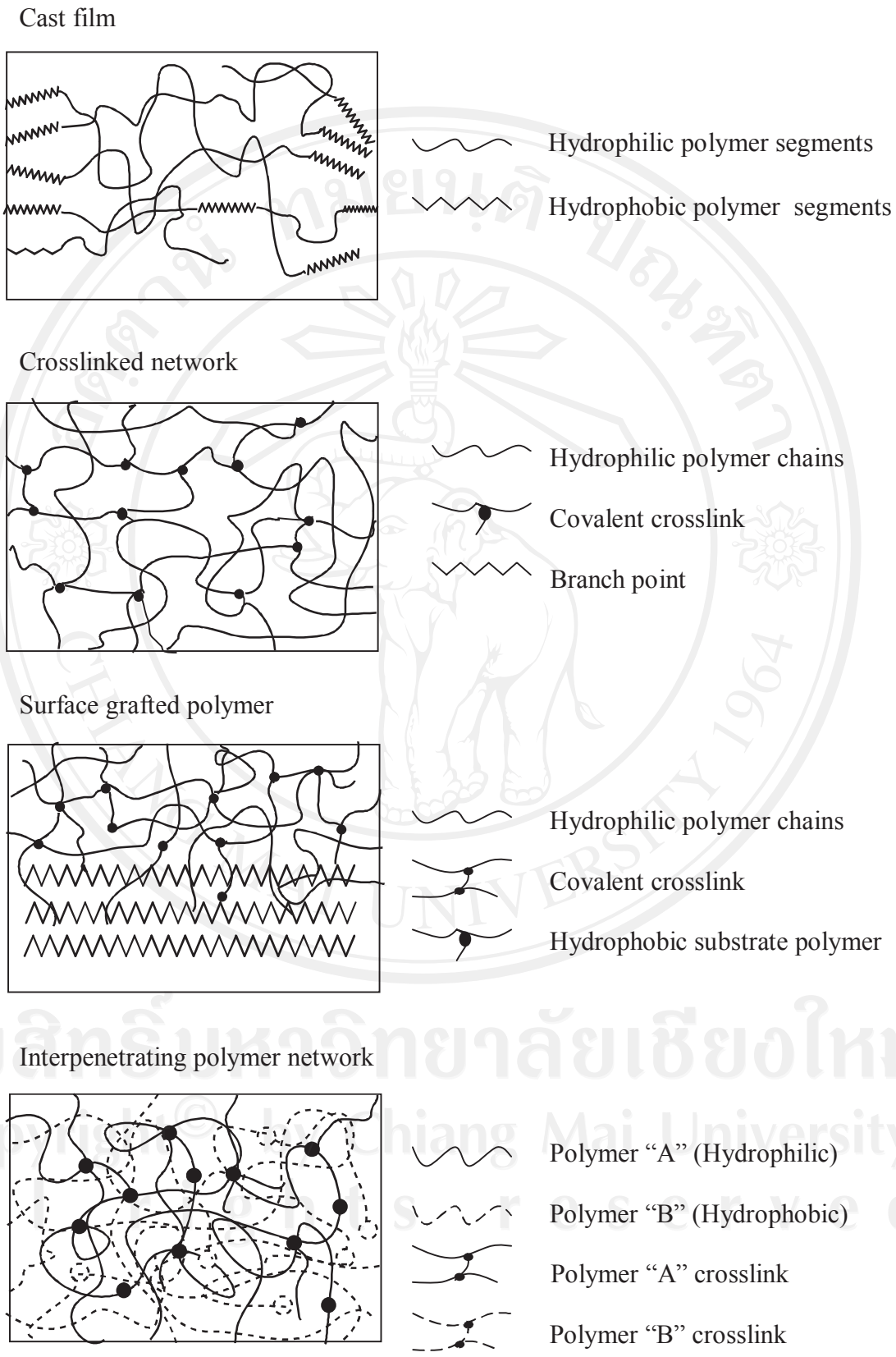


Figure 1.2. Water entrance into a single, idealized hydrogel chain segment.

1.4. Chemical and Physical Properties of Hydrogels

1.4.1. Water in Hydrogels

Water has numerous roles within a hydrogel as a plasticizer, a transport medium within the polymer matrix for dissolved species for example oxygen and a "bridge" between the different surface energies of synthetic polymers. Water absorbed by a hydrogel network structure contributes to the mechanical and physical properties of the gel [21]. The equilibrium water content (EWC) gives a quantitative evaluation of the water content of a gel and is expressed as the ratio of the weight of water in the hydrogel to the weight of water at equilibrium hydration by equation 1.1 [13]:

$$\text{EWC}(\%) = \frac{\text{Wt. of swollen polymer} - \text{Wt. of dry polymer}}{\text{Wt. of swollen polymer}} \times 100 \quad (1.1)$$

An alternative way of expressing a hydrogel´s water content is in terms of its so-called "swelling ratio". The swelling ratio of a hydrogel is given by equation 1.2 [22].

$$\text{Swelling ratio} = \frac{\text{Wt. of swollen polymer} - \text{Wt. of dry polymer}}{\text{Wt. of dry polymer}} \times 100 \quad (1.2)$$

Water present in crosslinked network structure of hydrogel exists between two extreme states as shown in Fig.1.3. Strong association between water and a polymer through hydrogen bonding is known as bound or non freezing water. If the water has a greater degree of mobility and remains unaffected by the polymeric environment it is referred to as free or unbound water.

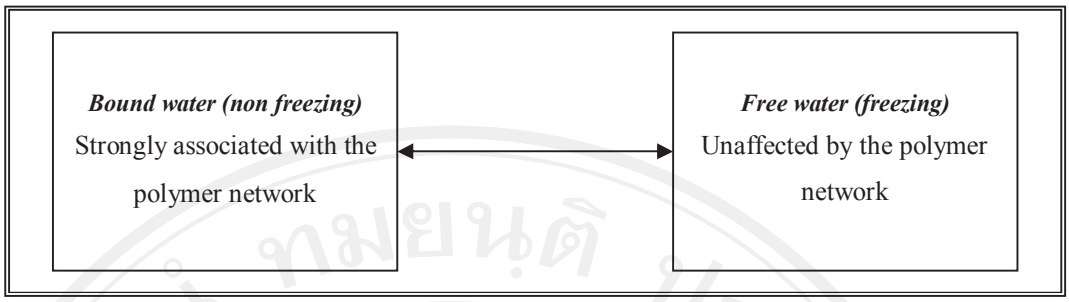


Figure 1.3. Classification of the bound and free water in hydrogels [21].

1.4.2. The Nature of Hydrogel-Water Interactions [23]

The character of the water molecules in a hydrogel can determine the overall permeation of nutrients into and cellular products out of the gel. When a dry hydrogel beings to absorb water, the first water molecules entering the matrix will hydrate the most polar, hydrophilic groups, leading to "*primary bound water*". As the polar groups are hydrated, the network swells, and exposes hydrophobic groups, which also interact with water molecules, leading to hydrophobically-bound water, or "*secondary bound water"*. Primary and secondary bound water are often combined and simply called the *"total bound water"*. After the polar and hydrophobic sites have interacted with the bound water molecules, the network will imbibe additional water, due to the osmotic driving force of the network chains towards infinite dilution. This additional swelling is opposed by the covalent or physical crosslinks, leading to an elastic network retraction force. Thus, the hydrogel will reach an equilibrium swelling level. The additional swelling water that is imbibed after the ionic, polar and hydrophobic groups become saturated with bound water is called *"free water"* or *"bulk water"*, and is assumed to fill the space between the network chains, and/or the center of larger pores, macropores or voids. As the network swells, if the network chains or crosslinks are degradable, the gel will being to disintegrate and dissolve, at a rate depending on its composition. It should be noted that a gel used as a tissue engineering matrix may never be dried, but the total water in the gel is still comprised of *"bound"* and *"free"* water. These different levels of interaction are presented, albeit simplistically, in Fig. 1.4 below.

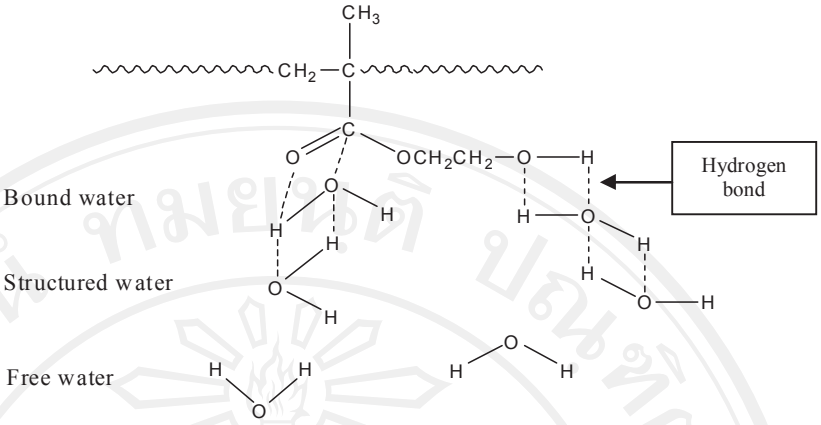


Figure 1.4. Different levels of polymer-water interactions.

where,

bound water	=	absorbed water which is strongly hydrogen bonded directly to the polymer
structured water	=	less tightly bound water interacting only indirectly with the polymer
free water	=	water in more mobile form

The factors which influence the swelling of hydrogels can be divided into two groups which are favorable to the entrance of water into the polymer structure and those which resist or inhibit the water influx. Some of these factors are summarized below:

Favorable to swelling	Inhibit swelling
Osmotic potential	Weak interactions with H_2O
Strong interactions with H_2O	Low free volume
High free volume	Low chain flexibility
High chain flexibility	High cross-linking density
Low cross-linking density	

The different types of water present in hydrogels can be quantified and characterized by using differential scanning calorimetry (DSC). The absorption characteristics of water in hydrophilic polymers vary in accordance to the following factors:

1.4.2.1. Nature of the Hydrophilic Groups

Water molecules interact more strongly with the polar groups within the polymer matrix than the non polar groups in the form of ion-dipole, dipole-dipole or hydrogen bonding. The distribution of bound to free water is affected by the polarity of the functional groups. For example ionic functional groups and strong acids will bind more strongly to the water compared to non-ionic polar groups (on the basis of the number of water molecules per functional group). Amide groups have a stronger binding power compared to hydroxyl or ether groups. Generally higher degrees of hydration are achieved for those hydrogels synthesized from more hydrophilic based monomers or polymers.

1.4.2.2. Crosslink Density of the Polymer Matrix

Increases in the crosslink density result in the formation of rigid and more compact structures with a decrease in the water content of the hydrogel. The amount of free water decreases due to the reduced mobility of the water molecules whilst the relative amounts of bound water increase.

1.4.2.3. Hydrophobic Character

Hydrogen bonding is largely responsible for the amount of bound water. Both interchain and intermolecular hydrogen bonding reduce the number of sites available for water and diminishes the amount of bound water.

1.4.3. Plasticizers

A plasticizer is an additive which makes a polymer material more flexible (by decreasing the glass transition temperature) and more malleable [24]. Generally there

is a reduction in the cohesive intermolecular forces along the polymer chain. In hydrogels, the water acts as placticizer to the gel. There are two principle methods for softening a polymer [25]:

1.4.3.1. Internal Plasticization

The monomer is chemically modified prior to polymerization. Implementing side chain grafting tends to increase the flexibility of the material.

1.4.3.2. External Plasticization

The addition of a suitable plasticizing agent (post polymerization) is a desired solution and reduces overall costs. The plasticizer interacts physically and acts as a solvent for the polymer. There are two groups of external plasticizer [24]. Primary plasticizer – which causes an increase in softness and elongation to take place. Secondary plasticizer – which enhances the performance of the primary plasticizer.

Three major theories, described below, explain how plasticizers alter polymer properties both by internal and external plasticization [26-27].

1.4.3.3. The Lubrication Theory

This theory explains the effect of an external plasticizer on a polymer. An unplasticized polymer is rigid since friction exists between its chains, binding them into a network. Heating results in the weakening of bonds which allows smaller plasticizer molecules to slide between the polymer chains. Upon cooling the plasticizer molecules shield the chains from each other thus preventing the reformation of the rigid network.

1.4.3.4. The Gel Theory

This is an extension of the lubrication theory. It proposes that the plasticizer molecules break up the polymer-polymer interaction by getting between the chains and the interaction sites from the polymer molecules.

1.4.3.5. The Free Volume Theory

This is an extension of both the lubrication and gel theories and explains both internal and external lubrication. The free volume of a polymer is a measure of the internal space available for the movement of the polymer chains. Plasticizers increase the free volume of the polymer, maintaining it as the mixture is cooled. This prevents interactions between neighbouring polymer chains. Motion of the chain, chain ends or attached side chains increase the free volume of the polymer. A plasticizer has a lower molecular weight than the polymer. It has the ability to impart a greater free volume per volume of material since there is an increase in the proportion of end groups and the plasticizer has a glass transition temperature (Tg) lower than that of the polymer.

1.4.4. Crosslinking

The chemical bonds that occur between macromolecules are known as crosslinks. The mechanical and physical properties are influenced by the density of this linkage. Thermoplastics are uncrosslinked polymers which be able to flow at high temperatures. In contrast, crosslinked polymers are unable to melt due to the constraints on the molecular motion, introduced by this particular covalent bond. At temperatures necessary to achieve the flow of thermoplastics, the polymers undergo irreversible degradation. Dissolution behaviour exhibited by uncrosslinked and crosslinked polymers differ. They are dependent upon the nature and extent of the interchain covalent bonds. Polymers without this linkage dissolve if an adequate polymer-solvent compatibility is achieved. In contrast, polymers containing this linkage will not dissolve if the solvation of chain segments cannot overcome the effect of the covalent bonds between the macromolecules. The crosslink density of a polymer controls the amount of solvent suspended within the freespace of its matrix. The swelling is reversible in lightly crosslinked polymers and upon the removal of the solvent it may return to its original size. Polymers that are lightly crosslinked have the tendency to become soft and flexible, particularly above their glass transition temperature. A dense three dimensional network of covalent bonds is achieved by heavily crosslinking a polymer, producing a very brittle material. An increase in the

number of covalent bonds reduces the freedom of motion by the individual segments of the molecules. Stress cannot be taken up by the structure and causes catastrophic failure at a given load with minimal deformation.

Difunctional or multifunctional monomers act as crosslinking agents, forming links between oligomers and other reactive molecules. This provides the polymer with increased elastic response, higher stiffness and a lower value of creep, compared to a non-crosslinked polymer [25].

1.4.4.1. Crosslink Ratio

A characteristic advantage of hydrogels is that their properties can be tailored by modifying the ratio of copolymers to obtain different degrees of hydrophilicity. On the contrary, a disadvantage intrinsic to hydrogels is their low mechanical strength, although this is overcome by altering the degree of crosslink or by forming interpenetrating networks. Hydrogels acquire the majority of their mechanical strength from the crosslinks within the polymer network. An important parameter used to identify crosslinked structures is the crosslink ratio (X). This is the ratio of crosslinker moles to the polymer repeat units. From this, the number average molecular weight between crosslinks, $\overline{M}_c$, is defined by equation 1.3:

$$\overline{M}_c = M_r / 2X \qquad (1.3)$$

Where M_r is the molecular weight of the repeat unit [28]. The crosslinked density, ρ_x, can then be determined from equation 1.4,

$$\rho_x = 1/v\overline{M}_c \qquad (1.4)$$

Where ν is the specific volume of the polymer [29]. These equations can be applied on the assumption that all crosslinking agent has reacted with polymer.

Increasing the concentration of crosslinking agent creates a hydrogel with greater mechanical strength. This, however, causes the hydrogel to become brittle. In addition this causes an increased elastic network retraction force resulting in decreased diffusivity within the hydrogel; thereby causing a reduction in release and swelling rates as well as EWC [30-31]. Thus an optimum degree of crosslinking that achieves a relatively strong, yet elastic hydrogel, will need to be examined for each system.

1.5. pH-Sensitive Hydrogels

"Smart" hydrogels, or stimuli-sensitive hydrogels, are very different from inert hydrogels in that they can exhibit dramatic changes in their swelling behavior, network structure, permeability or mechanical strength in response to changes in the pH or ionic strength of the surrounding fluid, or temperature [32]. Hydrogel exhibiting pH-dependent swelling behavior can be swollen from ionic netwoks. These ionic networks contain either acidic or basic pendant groups [33-44]. In aqueous media of appropriate pH and ionic strength, the pendant groups can ionize, developing fixed charges on the gel. As a result of the electrostatic repulsions, the uptake of solvent in the network is increased. Ionic hydrogels are swollen polymer networks containing pendant groups, such as carboxylic or sulfonic acid, which show sudden or gradual changes in their dynamic and equilibrium swelling behavior as a result of changing the external pH. In these gels, ionization occurs when the pH of the environment is above the pK_a of the ionizable group [33-46]. As the degree of ionization increases (increased system pH), the number of fixed charges increases, resulting in increased electrostatic repulsions between the chains. This, in turn, results in an increased hydrophilicity of the network, and greater swelling ratios. Conversely, cationic materials contain pendant groups such as amines [47-49]. These groups ionize in media which are at a pH below the pk_b of a ionizable species. Thus, in a low

pH environment, ionization increased, causing increased electrostatic repulsions. The hydrogel becomes increasingly hydrophilic and will swell to an increased level.

1.6. Applications of Hydrogels for Used as Wound Dressings

1.6.1. Development of Hydrogel

The earliest hydrogel patents are those of Wichterle and his co-workers [50], whose work has already been mentioned. In subsequent years, these have been expanded and enlarged covering both processes for production and the composition of materials. The claims of the original patents encompass the formation of sparingly crosslinked soft and elastic hydrogels both by polymerization of hydrophilic monomers and alternatively by the crosslinking of performed hydrophilic (water soluble) polymers. The polymerization of hydrophilic monomers, such as 2-hydroxyethyl methacrylate (HEMA) in the presence of ethylene glycol dimethacrylate (EGDM), however, is ideally suited to hydrogel formation. The major forming processes suggested by Wichterle included polymerization of monomer′s. The success of this original work may be judged, however, from the fact that a major proportion of the hydrogel sold today are based, directly or indirectly, on Wichterle′s patents.

Although Wichterle′s original patents envisaged the use of monomers other than HEMA their primary concern is with the technique of manufacture of hydrogels and particularly with their application. The specific advantages of monomers other than HEMA are described and their role has been largely exploited in the later section. Thus, although Wichterle′s patents in the period 1961 to 1968 mention the possible use of some other monomers (for example, acrylamide, methacrylic acid, and dimethylaminoethyl methacrylate) no indication is given of the benefits that these might confer either in terms of the physical properties of hydrogels or improved characteristics of the hydrogel formed from them. It was not until the early 1970s that the patents began to appear in which such benefits were described. The major feature of the number of claims published at this time was the use of *N*-vinyl pyrrolidone

(usually referred to simply as vinyl pyrrolidone and abbreviated as NVP). Two methods of use of vinyl pyrrolidone in hydrogels can be distinguished. These are graft copolymers and random copolymers (more correctly called statistical copolymer). Later, this resulted in the development of the various applications of hydrogels, for example biocompatible polymers and novel drug delivery systems. Ratner and Hoffman [23] suggested that the physical properties of hydrogels resembled those of living tissue more than other classes of synthetic biomaterials. Their relatively high water content and soft rubbery consistency give a strong superficial resemblance to soft living tissue. A wide variety of materials can be used to prepare the hydrogels, including naturally originating materials (proteins such as collagen, polysaccharides such as chitosan or hyaluronic acid) which require modification and synthetic chemicals.

Addition, hydrogels can be classified according to the nature of the side group. Neutral gels possess chains with no side charge and ionic gels have either negatively or positively charged side groups [51]. Morphology is another category used to classify the structure of gels. Amorphous gels possess random polymer chains, semi crystalline gels contain denser regions of ordered polymer chains and hydrogen bonded gels have a three dimensional polymer network held together through this interaction [30]. The shape of a hydrogel is maintained by the balance between the osmotic forces, originating from the entry of the water into the polymer and cohesive forces exerted by the polymer chains which resist expansion, also known as the elastic network retraction force.

1.6.2. Property Requirements of Hydrogel for Used as Wound Dressings

Burn injuries are probably the most traumatic and most difficult to tend of all external injuries with many complications arising from the initial loss of skin. The number of burns accidents is currently increasing. Currently available wound coverings have improved patient care significantly. The properties that serve to produce the hydrogel or an optimally useful general dressing for discard and

replacement as needed during wound care require careful consideration. Thus, the essential requirements for the wound dressing should be [52-53]:

- Controlled water vapour permeability
- Allowance of gaseous exchange
- Absorption of excess exudates
- Impermeable to bacteria
- Easy to apply and remove
- Non-toxic and non-antigenic

1.6.3. Skin Adhesive

The use of crosslink, covalently bonded, synthetic hydrogels has grown considerably in recent years in biomedical applications such as contact lenses, ocular prostheses, synthetic articular cartilage, implant and reconstructive materials, wound dressings, skin adhesives, sensors, perm-selective membranes, drug delivery systems and the list goes on [54-57]. This is attributed to the biocompatibility of hydrogels resulting from their ability to simulate natural tissue by virtue of their high water content and special surface properties [58]. In this research work focused in particular on hydrogels used in applications of temporary wound dressings where skin adhesives were concerned.

Skin adhesives may be defined as pressure sensitive adhesive (PSA) materials that adhere to the skin by applying a light pressure but which should leave no residual adhesive upon their removal [59]. In principal a functional skin adhesive requires peel adhesion. It is used to evaluate the degree of difficulty of removing the adhesive.

1.6.3.1. Mechanism of Adhesion

Adhesion can be defined as the state in which two surfaces are held together by interfacial forces [60]. The interfacial forces may range from valence forces to mechanical interactions, or some combination of chemical and physical interactions.

When one or both of the adherends are of a biological nature this interaction is known as bioadhesion. A bioadhesive can therefore be defined as a substance that has the ability to adhere to a biological material and is capable of being retained on the biological substrate for a protracted period. One distinctive feature of bioadhesion is that adhesion usually occurs in the presence of water. It has been suggested that it is more appropriate to refer to bioadhesion as a phenomenon rather than a mechanism [61]. Hence different types of bioadhesion have been grouped from a phenomenological perspective.

Class I : The adhesion between biological objectes.
Class II : The adhesion of biological substances to an artificial substrate.
Class III : The reverse of class II, that is the adhesion of an artificial substrate (such as a hydrogel) to a biological substrate.

Furthermore this perspective allows for one or more theories to explain the formation of a bioadhesive bond, as the pertinence of each theory is not exclusive as it depends upon the system concerned.

1.6.3.2. Suitability of Hydrogels as Skin Adhesives

A skin adhesive ought to adhere to the skin for the duration of intended use, usually between twenty-four hours and seven days. Additionally it should be easy to apply and remove from the skin and ought to remain adhesive if removed for repositioning. In theory it is feasible to improve the cohesive strength of a hydrogel through crosslinking, yet maintaining its tack and adhesive properties. It is already known that the strength of adhesion is affected by the molecular weight of the adhesive, crosslink density, hydration, hydrophilicity, charge, applied force, initial contact pressure and time and temperature [60, 62]. Hence by controlling the surface chemistry of the adhesion hydrogel, through compositional manipulation, molecular contact and interactions at the interface between the skin the adhesive can be modified.

Three main factors have been identified that are responsible for the importance of hydrogels as skin adhesive materials. These are the presence of a hydrophilic component, the short-range interactions via hydrophobic domains on flexible side chains and the rheological properties [56].

- **Hydrophilicity**

The hydrophilic component is desired in order to remove the lubricating interfacial water layer between the hydrogel and the skin allowing for maximum interaction at the interface. Hydrogels prepared with hydrophilic monomers contain chains with the hydrophilic groups, such as hydroxyl$(-OH)$, carboxyl$(-COOH)$, amide $(-CONH_2)$ and sulphate $(-SO_4^{2-})$ groups. When partially hydrated these hydrogels encompass an outstanding capacity for water uptake; hence potentially a high EWC that promotes their adhesiveness when applied to the skin.

- **Hydrophobic Domains**

In comparison to other polymers, hydrogels have a low crosslink density enhancing the flexibility of the hydrophobic groups on the external hydrogel chain branches. In air, for example, when the environment is relatively hydrophobic compared to that within the hydrogel, the hydrophobic groups rotate thereby exposing their side chains. In contrast to this, in a relatively hydrophilic environment, such as water, the chains rotate thereby exposing their hydrophilic side chains. This feature enhances skin adhesion resulting from "matching" of the hydrophobic side chain with the hydrophobic lipids and proteins of the skin resulting in a hydrophobic interaction.

- **Dynamic Mechanical Properties**

The rheological properties of hydrogels can be tailored to allow close conformation to the skin surface. These properties give the hydrogel sufficient cohesive mechanical strength to remain consistent on remove or for replacing. Ideally a skin adhesive hydrogel should have dominant elastic forces at low frequency stresses, such as when being applied to the skin. This facilitates close shaping of the

hydrogel to the skin contours. The viscous component should be dominant at high frequency stresses, such as during its removal.

In terms of an economically viable product, hydrogels also allow for production of sterile samples, are disposable, have ample shelf life and are aesthetically pleasing.

1.6.4. Wound Healing [63]

1.6.4.1. Physiology of Wound Healing

Injuries occur every day and normally they heal quickly. Wound healing is a complex process in which a variety of mediators and reactions ensure its smooth progression. Wound healing can be divided into 5 phases, which to some extent proceed simultaneously:

- **Blood Clotting for Provisionary Wound Closure**

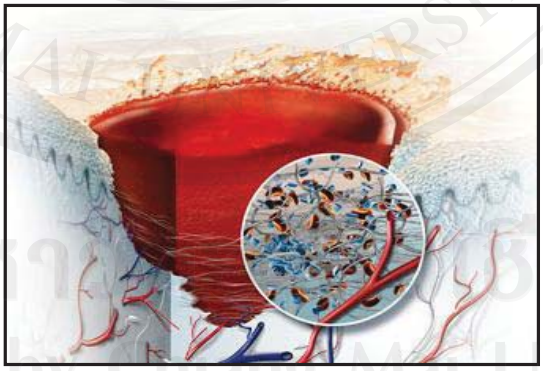


Figure 1.5. Blood Clotting.

Blood vessels are the most likely structure to be damaged in case of injury. First objective of the repair process of the body is to stop the bleeding. Platelet aggregation and activation of the coagulation cascade result in blood clotting.

• **Exudation / Inflammatory Phase**

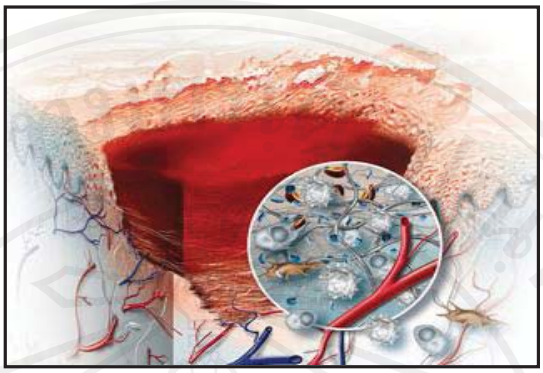


Figure 1.6. Inflammation.

During this phase, the dilation of vessels near the wound leads to a vascular fluid leakage, resulting in an oedema of the wound. This fluid, the so-called exudate, contains a variety of essential substances such as enzymes, antibodies, and growth and inflammation cells, all of them necessary for an undisrupted healing process.

• **Proliferation / Granulation Phase**

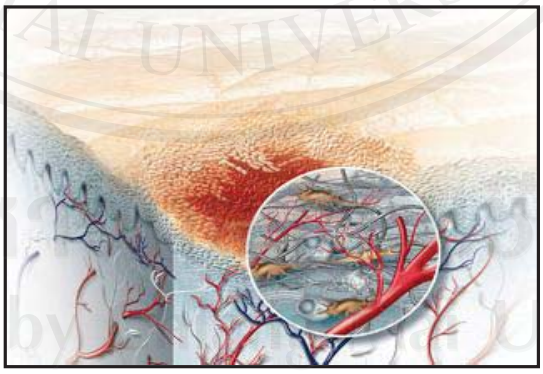


Figure 1.7. Granulation.

The regeneration of new cells – in order to replace the damaged tissue – predominates during the next wound-healing phase. The new tissue is known as granulation tissue. It fills in the wound from underneath and has a bright red

appearance. The build-up of new tissue is performed by fibroblasts (the main cells of the dermis) through synthesis of collagen fibres forming the connective tissue matrix.

- **Epithelization Phase**

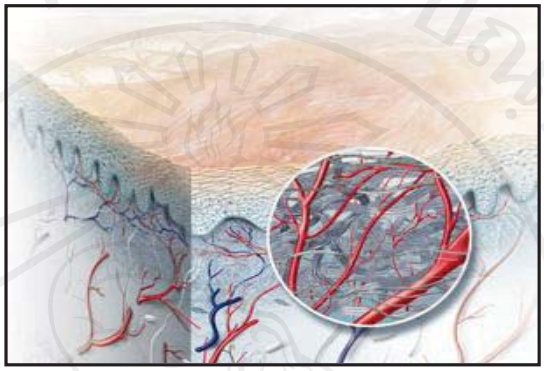


Figure 1.8. Closure (Scar).

While new vessels are responsible for the transportation of nutrients to the regenerating area, granulation tissue fills the wound and creates the basis for epithelialization: the build-up of a new skin layer. The epithelialization of the wound completes the wound healing process. Epithelial cells divide and migrate from the wound edges and close the wound. Once the covering epithelium beneath a scab is renewed, it peels off and newly built, pink-coloured epithelial tissue underneath becomes visible.

- **Maturation Phase**

The proper wound healing phase is followed by the maturation phase in which the collagen fibres are reorganized to acquire more strength to the skin. This newly built tissue however is not identical with the original tissue. Scar tissue does not have good blood circulation, is sometimes uneven and less elastic. In addition major variations in colour are possible and in up to 15% of injuries, hypertrophic scar formation can be observed. The process of remodeling of the scar tissue can take years.

1.6.4.2. Moist Wound Healing

Wound Care

Depending on the type of wound, its size and the phase of wound healing, two different approaches to wound management of minor wounds exist:

i. dry wound healing.
ii. wound treatment in a moist environment, or "moist" wound healing.

i. Dry Wound Healing

Objective of traditional wound care with standard bandages is to cover and protect the wound, to absorb excess wound exudate and to keep the wound dry.

The essential feature of dry healing is that all repair and regenerative processes take place under a protective scab. This firm crust of coagulated blood is the body′s own wound cover protecting the wound against external influences.

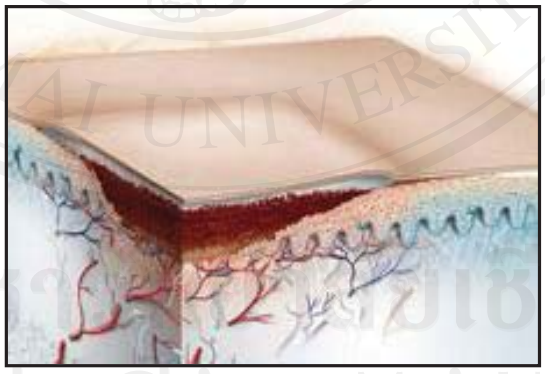


Figure 1.9. Dry wound healing.

ii. Moist Wound Healing-the Advanced Treatment

The idea of moist healing began in 1962 when George D. Winter discovered that healing would proceed twice as fast in a moist environment than under a scab. The principle of moist healing is generally accepted in the professional field, where

many products have already been developed for the healing of chronic wounds via moist wound therapy.

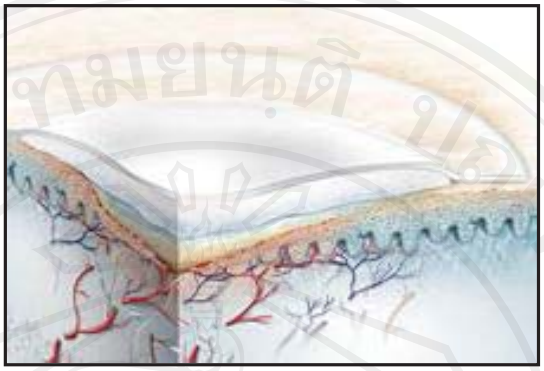


Figure 1.10. Moist wound healing.

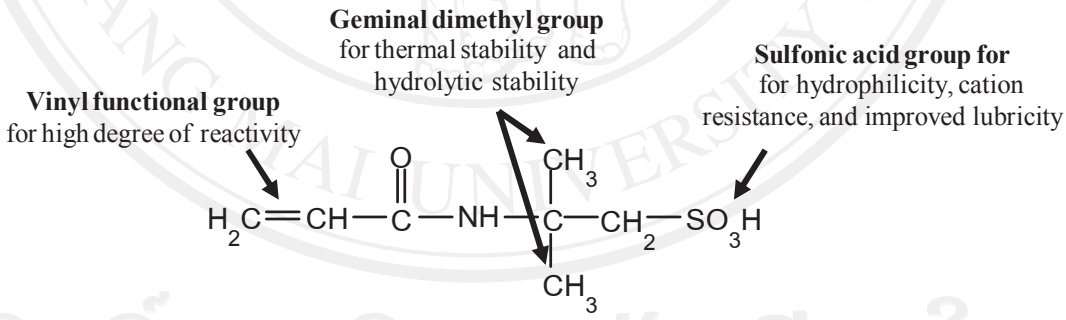
Cell growth needs moisture and the principle aim of moist wound therapy is to create and maintain optimal moist conditions. Cells can grow, divide and migrate at an increased rate to optimize the formation of new tissue. During this phase of wound healing an aqueous medium with several nutrients and vitamins is essential for cell metabolism and growth. The wound exudate serves as a transport medium for a variety of bioactive molecules such as enzymes, growth factors and hormones. The different cells in the wound area communicate with each other via these mediators, making sure that the healing processes proceed in a coordinated manner. Wound exudate also provides the different cells of the immune system with ideal conditions to destroy invading pathogens such as bacteria, foreign bodies and necrotic tissues, diminishing the rate of infection. Moist wound treatment is known to prevent formation of a scab, allowing epithelial cells to spread horizontally outwards through the thin layer of wound exudate to rapidly close the wound.

Benefits of moist wound healing

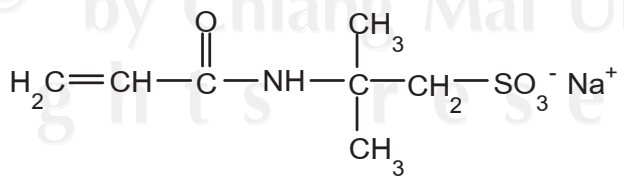
1. Up to 50% faster wound healing (epithelization and dermal repair).
2. Lower rate of infection.
3. Painless removal of the dressing without destroying newly formed tissue.
4. Less scarring and better cosmetic results.

1.7. The Relevance of 2-acrylamido-2-methylpropane sulfonic acid (AMPS), *N*-vinylpyrrolidone (NVP) and methacrylic acid (MAA) monomers with hydrogels

In recent years, the sodium salt of 2-acrylamido-2-methylpropane sulfonic acid (AMPS-Na^+) has received much attention as a replacement for HEMA due to its strongly ionizable sulfonate group, good skin adhesion, and ease of removal of residual monomer. The use of AMPS, or its sodium salt, AMPS-Na^+, as a principal component of hydrogels for medical use is now well established. Their hydrogels are typically stable, 3-dimensional crosslinked polymer networks formed by free radical polymerization in aqueous medium. They can be crosslinked by means of ultraviolet radiation, thermal or redox initiation with a multifunctional crosslinking agent such as Okay and Durmaz [64] studied the AMPS based hydrogels can be used for skin contact electrodes, support carrier in biomedical engineering, and in drug delivery applications. The chemical structures of AMPS and AMPS-Na^+ monomer are shown in Fig. 1.11.



2-acrylamido-2-methylpropane sulfonic acid (AMPS)



sodium salt of 2-acrylamido-2-methylpropane sulfonic acid (AMPS-Na^+)

Figure 1.11. The chemical structures of AMPS and AMPS-Na^+ as main monomers.

In addition, hydrogel synthesized from AMPS-H^+ is easily dissolved in water and the useful properties of poly(AMPS-Na^+) are its hydrophilicity, thermal stability, stability over a broad pH range, resistance to hydrolysis and ionic character. The advantages of this synthetic hydrogel are that it is soft and flexible and the residual unreacted monomer can be removed easily by water due to the polymerization being performed in an aqueous system. The result is that an already hydrated hydrogel is obtained because water is retained within the 3-dimensional hydrogel structure. This synthetic hydrogel is considered to have potential for application as a wound dressing material.

Other monomers such as *N*-vinylpyrrolidone (NVP) and methacrylic acid (MAA) have also received attention as property modifiers for both HEMA and AMPS-Na^+ due to NVP content of the polymers tents to produce hydrogels which are softer and slightly tacky and this may be advantageous, especially where the hydrogel layer is supported on a reinforcing film or fabric, and MAA is classic examples of pH sensitive carriers that exhibit swelling transitions in response to changes in pH, further more, it can improve the gel strength property [65]. The chemical structures of NVP and MAA are shown in Fig. 1.12.

N-vinylpyrrolidone (NVP) Methacrylic acid (MAA)

Figure 1.12. The chemical structures of NVP and MAA.

There are currently many papers and patents citing the hydrogel synthesized from AMPS-Na^+, NVP and MAA monomers. Lui *et al.* [66] synthesized the hydrogels based on acrylamide (AAM) and AMPS by free radical polymerization at 60-80°C. AMPS monomer was partially neutralized by an aqueous NaOH solution under cooling. *N,N'*-methylene-bis-acrylamide (NMBA) and potassium persulfate

(KPS) were used as the crosslink and initiator respectively. It was found that the value of equilibrium swelling of AAM/AMPS is 1,749 g H_2O/g polymer. Abdel-Azim *et al*. [67] synthesized of AMPS/AAM and AMPS/acrylic acid (AA) copolymers. The catalytic initiation polymerization was carried out under a nitrogen atmosphere at 55°C. Cross-linker *1,1,1*-trimethylolpropane trimethacrylate (TPT), and initiator benzoyl peroxide were employed. Their observed that the swelling of AMPS/AAM copolymer is greater than the swelling of AMPS/AA. The reason of this effect is the acrylamide contains more hydrophilic group than the acrylic acid. Walker *et al.* [68] studied the AMPS/HEMA copolymers for applied to proton-conducting polymer membrane. Polymerization was carried out by using 2,2′-azobisisobutyronitrile (AIBN) as the initiator. Their compared the equilibrium swelling of AMPS/HEMA and Nafion 117, commercially available of synthetic wound dressing. It was showed that the AMPS/HEMA exhibiting a higher swelling 4 times than that of Nafion 117. Okay and Durmaz [69] studied the relationship between the mechanism and the swelling of hydrogels. Their hydrogels were prepared by free radical copolymerization of acrylamide (AAM) and AMPS monomers. NMBA and KPS were used as the crosslinker and the initiator respectively. The synthesized hydrogels were characterized in term of their equilibrium swelling in water and NaCl solutions. It was found that, in the absence of AMPS the swelling in water as well as in aqueous NaCl solutions was very low. However, with 5-100% mol AMPS, the swelling increased significantly but decreasing in NaCl as the external solution due to the osmotic pressure effect. Similarly, Kundakci and co-workers [70] compared the swelling properties of pure AAM and AAM/AMPS copolymers prepared by free radical polymerization but with ethylene glycol dimethacrylate (EGDM) as the crosslinker instead of NMBA. Their work showed that the AAM/AMPS exhibited a higher swelling ratio than pure AAM due to the characteristic of hydrophilic groups on the AMPS. Solpan *et al.* [71] investigated water absorption under similar conditions to those of Okay and Durmaz [69] but with the monomers changed from AAM and AMPS to NVP and MAA. From their results, the swelling of the hydrogel depended on the carboxylic acid group in the MAA. It was indicated that increasing the MAA content led to an increase in electrostatic repulsion along the chain resulting

in an expansion of the network structure. Thus, hydrogels containing more MAA produced higher percent swellings.

As the skin is destroyed, the evaporative water loss from the wound surface can be many times greater than from normal skin. This water loss can be reduced by the application of a wound dressing. Thus, Queen and co-workers [72] carried out an *in vitro* evaluation of the water vapour transmission rate (WVTR) under clinical conditions and determined the difference between the values obtained using water and plasma by the American Society for the Testing of Materials (ASTM) procedures. It was found that the water vapour transmissivity of a burn wound dressing should maintain a moisture balance within the repairing wound. In addition, the statistical tests showed no significant difference between the WVTR of a dressing when in contact with plasma rather than water. Therefore, this investigation indicated that the test could be performed using either.

In other work, He and co-workers [73] described the synthesis and swelling properties of pure MAA hydrogels. MAA monomer was used because of its swelling response to changes in pH. In addition, EGDM and Irgacure 651 were used as the crosslinker and photoinitiator respectively, with the photopolymerization carried out in a mixed solvent of water and ethanol of varying ratios. Synthetic hydrogels were synthesized by UV photopolymerization [74-75]. They can be synthesized at temperatures near to physiological conditions and are easily controlled by adjusting the intensity of the UV light. He and co-workers [73] also studied the swelling properties in pH buffer solutions. The results showed that the hydrogels swelled the highest at pH 7.4 due to the electrostatic repulsion between the ionized forms of the carboxylic acid groups, as well as the dissociation of hydrogel bonds between the carboxylic acid groups of the MAA and the oxygen of EGDM. In contrast, at lower pH, the swelling ratio decreased due to the formation of hydrogen bonding. Rosso and co-workers [76] studied new polyelectrolyte hydrogels for biomedical applications synthesized by copolymerization of HEMA/AMPS and HEMA/2-methacryloyloxyethyl ammonium chloride copolymers were anionic and cationic copolymers respectively. Their studies showed that cationic copolymers have good cell adhesion, whereas anionic copolymers have poor cell adhesion.

1.8. Polymerization of Hydrogels

1.8.1. Preparation of Hydrogels

The method used to prepare hydrogels from hydrophilic monomers is free radical polymerization [77-78]. This common technique involves the polymerization of a water soluble monomer functionalized with a radically polymerizable group (carbon-carbon double bonds). An appropriate difunctional crosslinking agent, a low concentration of initiator and water may also be included at this stage. The chemistry of a typical free radical polymerization involves three main steps include initiation, propagation and termination. A 3-dimensional crosslinked polymer is formed when a difunctional monomer is employed [71].

Polymerization can be achieved in bulk or in solution, both methods having advantages and disadvantages. A bulk polymerization is often referred to as a neat polymerization and involves a fast monomer conversion occurring over a few minutes. The main advantage is that no solvent removal from the polymer is required which conserves time, cost and labour. This type of polymerization is normally used for the fabrication of hydrogels sheets. Solution polymerization is mainly used if a large quantity of hydrogel is required.

The free radical addition of two different monomers to one another is known as copolymerization. Problems arise when the reactivity of the monomers are different. The ability to produce polymers containing long sequences of two or more different monomers (block and graft) has led to new products with unique and valuable properties, then the polymer chains can be tailored. The mechanism is analogous to that of homopolymerization. However, the reactivities of monomers may vary considerably from one another. Two monomers M_1 and M_2 can undergo either self-propagation where M_1 reacts with $\sim\sim M_1\bullet$ or M_2 reacts with $\sim\sim M_2\bullet$ or cross-propagation when $\sim\sim M_1\bullet$ reacts with M_2 or $\sim\sim M_2\bullet$ reacts with M_1 [79-81]. Under steady-state conditions, assuming that the radical reactivity is independent of chain length (penultimate effect) and depends only on the nature of the terminal unit, the rate of addition of $\sim\sim M_1\bullet$ to M_2 will equal the rate of addition of $\sim\sim M_2\bullet$ to M_1.

1.8.2. Mechanism for Gelation

It is well known that free-radical polymerization of multifunctional monomers forms heterogeneous polymer networks, leading to microgel formation [82-88]. Such entities are a result of strong intramolecular crosslinking of the growing macroradicals. Eventually, intermolecular reactions among microgels form the network structure. The relative rates of intra- and intermolecular reactions depend on the initial monomer composition, as well as other reaction conditions. The solvent composition is a major factor influencing the gelation kinetics. The mechanism for gelation can be described in five stages: initiation, microgel formation, cluster formation, macro-gelation, and post-gelation. The schematic diagram of structure formation in the photopolymerization describing the first four stages is given in Fig. 1.13. In the first stage, all reactants are mixed together and UV radiation initiates initiator decomposition to form radicals (shown as filled dots).

After initiation, radicals react with C=C monomers to produce monomeric radicals. Because of the presence of multifunctional monomers, the monomeric radicals have chances to link with these molecules to form the growing macroradicals with pendant double bonds, leading to the cyclization or ring formation through intramolecular reactions. This internal crosslinking on the primary polymer chains leads to the formation of "microgels" [89]. Inside the microgels, the Trommsdorff effect may occur because termination is largely hindered due to immobilized polymerical radicals, while the propagation rate is less affected since small monomers are still mobile.

However, the relative viscosity remains nearly unchanged. The greater extent of intramolecular cyclization means less intermolecular crosslinking. This leads to larger mesh size in formed of hydrogels, and the weaker mechanical properties. This mechanism of intramolecular cyclization has been used to explain the network formation influenced by the light intensity [82], the solvent concentration [90], the solvent quality [91-92] and the curing temperature [87].

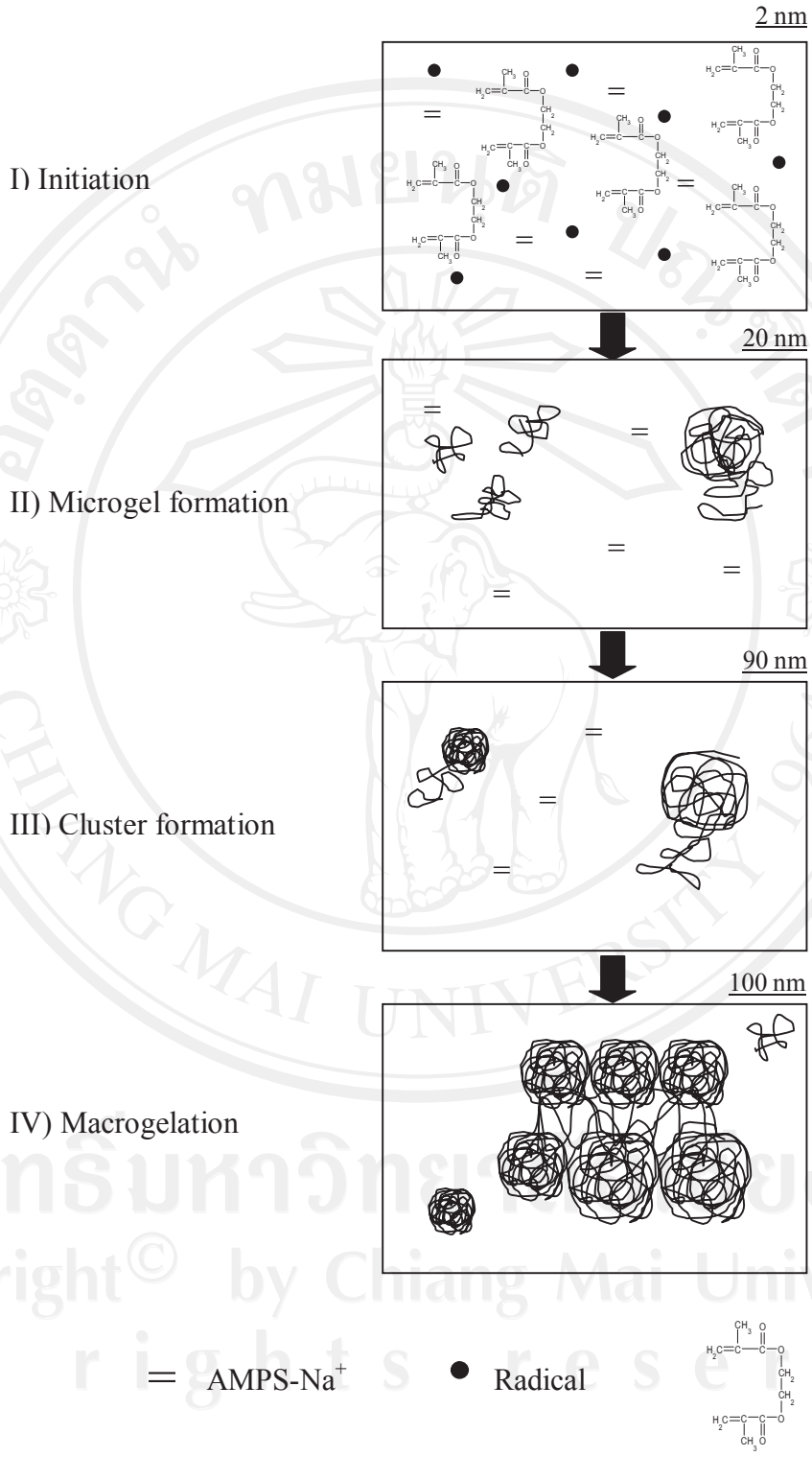


Figure 1.13. The schematic diagram of structure formation of microgels [82].

1.9. Research Aims and Specific Objectives

The aims of this research is to develop the synthetic AMPS-Na^+ based hydrogels which can be used as wound dressing for the second degree burns. This research work will provide knowledge and experience in hydrogel polymer synthesis combined with a sound appreciation of the medical problems involved in wound dressings and the complex property requirements of polymers. To achieve these aims, a number of specific objectives have been addressed including the following:

1. To synthesize the hydrogel sheets by UV- photopolymerization technique with different conditions i.e. AMPS-Na^+ monomer concentrations, EGDM crosslinker concentration, type of crosslinkers (EGDM and NMBA) and copolymerization between AMPS-Na^+ with either comonomers NVP or MAA.

2. To determine the water absorption properties of synthesized hydrogels such as water content and water swelling. Also, to compare water absorption of hydrogels synthesized from AMPS-Na^+ 40% w/v with NMBA crosslinker in deionized water, saline and synthetic body fluid.

3. To study the effect of cationic charges on water swelling of hydrogels using salt solutions with monovalent, divalent and trivalent cation.

4. To investigate the relative weight loss of water from hydrogel sheets synthesized from different conditions. Also, water vapour transmission rate (WVTR) was studied by water cup method at controlled temperature.

5. To determine the diffusion kinetic parameters of hydrogel sheet synthesized from AMPS-Na^+ 40% w/v with different concentrations of NMBA crosslinker.

6. To compare the skin adhesive property of pure AMPS-Na^+ and copolymers of AMPS-Na^+-co-NVP and AMPS-Na^+-co-MAA hydrogels in terms of peel strength.

7. To determine and compare the oxygen permeability of hydrogel sheets synthesized from pure AMPS-Na^+ and copolymer of AMPS-Na^+-co-NVP and AMPS-Na^+-co-MAA.

8. To investigate residual monomers in the hydrogel sheets after synthesized by ion chromatography technique. Also, cytotoxicity test of hydrogel sheets using mouse fibroblast L929 cells was examined.