# CHAPTER 4

DISCUSSION AND CONCLUSIONS

In this final Chapter 4, the following conclusions can be drawn from the experimental results obtained and presented in the previous Chapter 3. As the results have shown, the main aims of this research, as stated at the end of Chapter 1, have been achieved in this initial study of synthetic hydrogels as temporary skin substitutes.

4.1 Hydrogel Synthesis

The primary objective of this project has been to synthesize hydrogels based on poly(2-hydroxyethyl methacrylate), P(HEMA), by free radical polymerization. Ethylene glycol dimethacrylate, EGDMA, was used as a crosslinking agent both to improve mechanical strength and also as a convenient means of controlling water absorption properties.

2-hydroxyethyl methacrylate
HEMA

poly(2-hydroxyethyl methacrylate)
P(HEMA)

ethylene glycol dimethacrylate
EGDMA
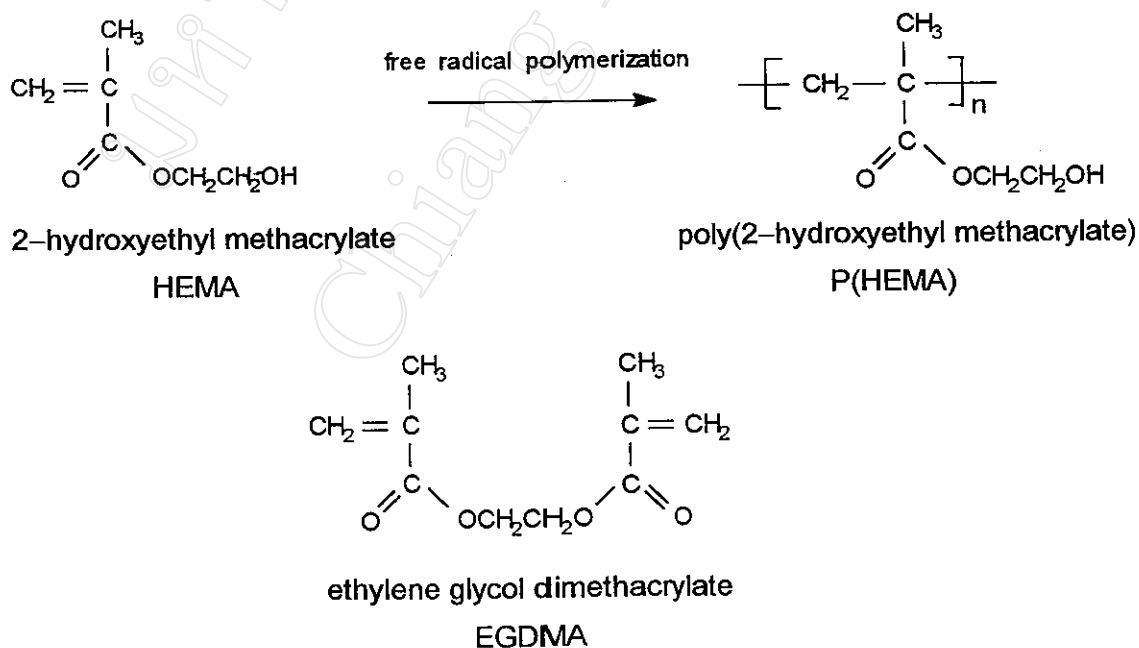
The 3-dimensional network structure of crosslinked P(HEMA) is represented below in Fig. 4.1 showing how the difunctional EGDMA units serve to join the P(HEMA) chains together at random points throughout the matrix. The structural similarity between EGDMA and HEMA ensures that the crosslinking does not seriously affect the chemical nature of the polymer.
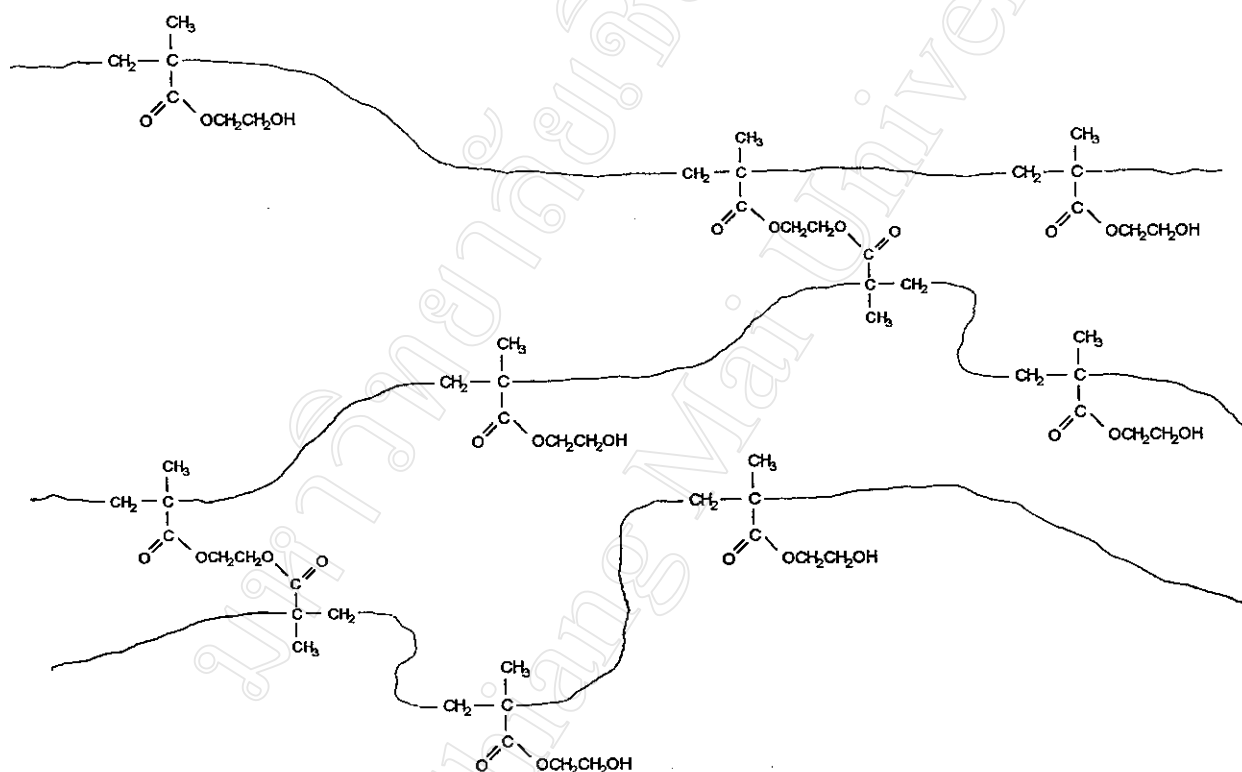
Fig. 4.1 : Structural representation of P(HEMA) crosslinked by EGDMA.

4.2 The Nature of Hydrogel–Water Interactions

The nature of the interaction between water and P(HEMA), which gives the polymer its so-called **"hydrogel"** properties, has been discussed at length in the literature [43]. The general concensus of opinion seems to be that there are different "levels" of interaction depending upon the proximity of the water molecule to the polymer chain. Since this interaction is clearly **"hydrogen–bonding"** in nature, the strength of interaction (or force of attraction) will obviously increase as this proximity increases. These different levels of interaction are represented, albeit simplistically, in Fig. 4.2 below.
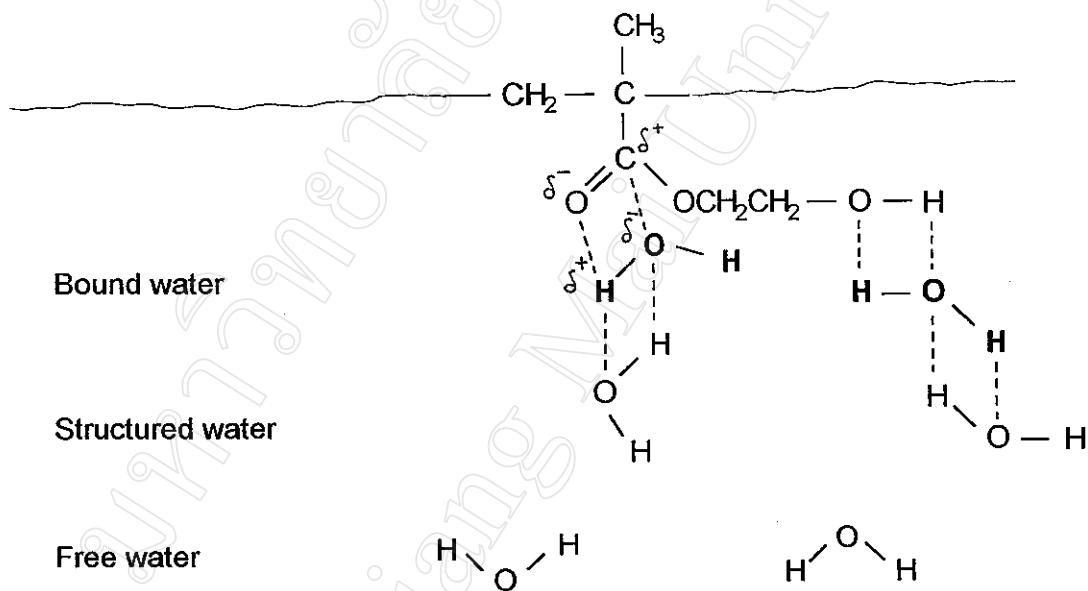
where :

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

Fig. 4.2 : Different levels of polymer–water interactions in P(HEMA).

This simplistic view in Fig. 4.2 of the water binding process suggests that in **bound water**, the water molecules are hydrogen bonded to hydrophilic groups on the polymer chain, while **structured water** molecules are hydrogen bonded to both **free** and **bound** water molecules. Using the concept of **hydration shells**, it is possible to envisage the bound water hydrogen bonded to the hydrophilic sites (in this case, hydroxyl and carbonyl groups) on the polymer chain, with other hydration shells of structured and free water molecules surrounding the inner hydration shell. Also, as the number of monomer units between crosslinks decreases, the network becomes more tightly bound and there is a marked decrease in free water content with increasing crosslink concentration, together with a small decrease in the number of moles of bound water per mole of hydrophilic repeat unit. Meanwhile, water that is characterized by differential scanning calorimetry (DSC) as free water is observed to have a marked **plasticizing** effect upon the hydrogel, whereas bound water has little such effect [53].
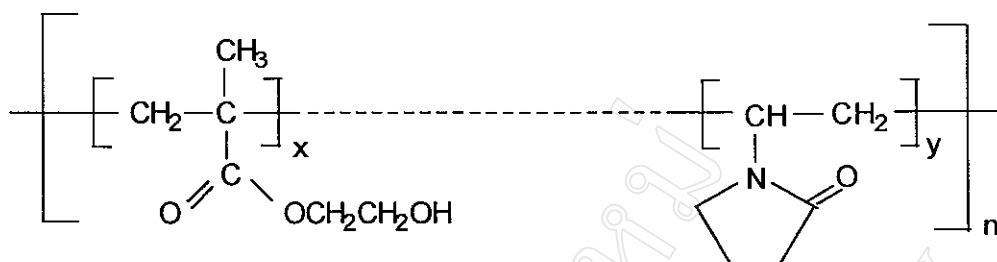
4.3 Effects of Crosslinking

In this study, the effects of ethylene glycol dimethacrylate, EGDMA, crosslinker (1% w/w) on the water uptake and equilibrium water content, EWC, of P(HEMA) were studied and the results shown in Fig. 3.23 (on page 59) and Table 3.16 (on page 61). The results indicated that, with or without crosslinking, the EWC was reached after about 2 hours immersion in distilled water at room temperature. However, the EWC of the uncrosslinked P(HEMA) (38.2 %) was higher than that of the crosslinked P(HEMA) (34.7 %). This is as would be expected since the crosslinks have the effect of tieing the P(HEMA) chains together in a 3–dimensional network, thereby stiffening the matrix and decreasing its capacity to expand or swell. This, in turn, decreases its porosity towards the uptake of water molecules. Thus, the crosslink density, which simply depends on the amount of crosslinking agent added, provides an easy and convenient means of controlling the EWC of the hydrogel polymer.

In addition to its effect on water absorption properties, crosslinking also has an important effect on mechanical properties. Indeed, the main reason why

synthetic hydrogels such as P(HEMA) are usually crosslinked is to increase their otherwise rather poor mechanical strength. As the results of this work have shown (Table 3.46 on page 102), the incorporation of EGDMA (1% w/w) crosslinks, as represented in Fig. 4.1, does indeed increase the tensile strength of P(HEMA) by about 16%. At the same time, the Young's Modulus (stiffness) also increases while the elongation at break (extensibility) decreases due to the constraining influence of the crosslinks on matrix deformation.

4.4 Effects of Copolymerization

Apart from crosslinking, the water binding behaviour of hydrogels can be controlled to a certain extent by copolymerization. In this project, N–vinylpyrrolidone, VP, was chosen as the modifying comonomer over the 0 – 20 wt.% range. VP was chosen because PVP is known to be a more hydrophilic polymer than P(HEMA); PVP is water–soluble whereas P(HEMA) is only water–swellable. Hence, the incorporation of VP units into the P(HEMA) chain should lead to an increase in hydrophilicity and, therefore, water absorption. The reason why PVP is water–soluble whereas P(HEMA) is not is probably due to the fact that there is less intermolecular interaction in PVP and therefore the chains are easier to solvate. In P(HEMA), the intermolecular hydrogen bonding between the hydroxyethyl groups is obviously stronger than the solvating power of water with the result that the polymer can only swell but not dissolve. Indeed, it was found in this work that P(HEMA) was extremely difficult to dissolve in any common organic solvent, hot or cold (Table 3.9 on page 43). It has also been reported in the literature that VP has been the main hydrophilic monomer used to obtain high water content hydrogels where stability to temperature, pH and ionicity is required [54].

poly(2–hydroxyethyl methacrylate–co–vinylpyrrolidone)
P(HEMA–co–VP)

The effects of copolymer composition on the water uptake and EWC of crosslinked P(HEMA–co–VP) hydrogels were shown previously in Fig. 3.24 (on page 59) and Table 3.16 (on page 61). While the EWC of crosslinked P(HEMA) is 34.7%, the EWCs of the crosslinked P(HEMA–co–VP) copolymers are in the range 35.6 – 40.9 % as the VP content in the comonomer feed increases from 5 – 20 wt.%. From elemental analysis of the actual copolymer compositions (Table 3.11 on page 51), the VP contents in the copolymers increase from 8 – 19 wt.%. Thus, as the VP content increases, the water uptake and EWC also increase. It was also observed that the copolymers became more pliable. This latter effect is probably due to the steric effect of the bulky VP groups decreasing the intermolecular interaction between the polymer chains. Also, of course, an increase in EWC effectively means an increase in the amount of **free water** between the chains. Since this free water acts like a **plasticizer**, it is reasonable to expect that a higher EWC will result in a more pliable material.

Therefore, the EWC of a P(HEMA–co–VP) copolymer at a given composition can be considered to depend upon the balance of contributing steric and polar effects. The polar effect arises predominantly from the hydroxyl group in the HEMA units and, to a lesser extent, from the carbonyl groups in both HEMA and VP, whereas the steric effect arises from the combined contributions of the bulky pyrrolidone group in VP together with the α–methyl group (alkyl side chain) in HEMA. Thus, in the P(HEMA–co–VP) copolymers, in which HEMA is by far the major component, polar effects predominate. However, as the proportion of VP units increases, the contribution from steric effects increases which, as the results show, leads to an increase in EWC.

4.5 Water Vapour Transmission Studies

Fluid retention (pooling) behind a burn wound dressing or, conversely, the dehydration of the granulating wound bed are serious problems in the treatment of burn injuries. It is therefore essential that the water vapour transmissivity, WVT, of a burn wound dressing should be able to maintain a satisfactory moisture balance within the repairing wound.

Lamke et al [48] investigated the water vapour transmission across normal skin. They also reported the evaporative water loss for burns, granulating wounds and donor sites along with the average surface temperature (Table 4.1). Their WVT rates, shown below, are useful to the study of temporary skin substitutes by laying down guidelines as to the required WVT of a dressing for a particular injury.

Table 4.1 : Water vapour transmission (WVT) rates and surface temperatures for normal skin, burns, granulating wounds and donor sites [48].

Area Studied	WVT (g / hr m^2)	Surface Temperature (°C)
Normal skin	8.5	35.8 ± 0.2
First degree burn	11.6	35.3 ± 0.1
Second degree burn	178.1	35.3 ± 0.4
Third degree burn	143.2	34.5 ± 0.4
Granulating wound	214.1	34.7 ± 0.2
Donor site	149.6	35.3 ± 0.2

These WVT rates in Table 4.1 can be usefully compared with those obtained for the samples prepared in this work, as listed in Table 4.2.

Table 4.2 : Water vapour transmission (WVT) rates for the P(HEMA) and
P(HEMA-co-VP) samples prepared in this work.

Sample[a]	WVT[b] $(g / hr \ m^2)$	
	Water Cup	Inverted Cup
No sample (open cup)	141.8	–
P(HEMA) uncrosslinked	11.8	78.7
P(HEMA) crosslinked	14.2	79.5
P(HEMA-co-VP) / 95 : 5	–	93.2
P(HEMA-co-VP) / 90 : 10	–	80.3
P(HEMA-co-VP) / 85 : 15	–	86.3
P(HEMA-co-VP) / 80 : 20	–	104.7
PVP uncrosslinked	42.9	–
PVP crosslinked	44.4	–

[a] average sample thickness = 0.5 ± 0.1 mm

[b] measured at $35.0 \pm 1.0°C$

When the WVT rates in Table 4.2 are compared with those in Table 4.1, it is probably more "realistic" to consider the 'Inverted Cup' values. This is because, in the 'Inverted Cup' method, the water is in direct contact with the sample, the same as the wound fluid (exudate) would be with a temporary skin substitute. On this basis, the 'Inverted Cup' WVT rates of between 78 – 105 g/hr m^2 are seen to be intermediate between the WVT rates for first and second degree burns. This would seem to be a reasonable level of transmission, restricting the WVT rate to below those for second and third degree burns without restricting it so much that pooling would occur. In medical practice, a balance needs to be struck between fluid evaporation and fluid retention so that the wound surface is just moist enough to

promote the healing process. It is also clear from the results in Table 4.2 that, as expected, the WVT rate increases with increasing VP content in the P(HEMA–co–VP) copolymer. This is useful insofar that the copolymer composition of a P(HEMA–co–VP) wound dressing could be "tailored" to meet the demands imposed by the severity of the burn.

The 'Water Cup' WVT rates in Table 4.2 support the above conclusions. Compared with the evaporative water loss from a free water surface (i.e., in the absence of any sample) of 141.8 g/hr m^2, the WVT rates for the P(HEMA) samples are lower by a factor of 10. (This comparison is not possible, of course, in the 'Inverted Cup' method.) Thus, P(HEMA) is seen to provide an effective partial barrier to evaporative water loss, even in films as thin as 0.5 mm. Also, the WVT rates for PVP in Table 4.2, being 3 – 4 times higher than those for P(HEMA), further support the previous conclusion that the more hydrophilic the polymer, the higher will be its water vapour transmissivity.

Clearly, the hydrogel samples prepared in this work are able to control water vapour transmission within reasonable limits. Since this ability is a vital function of a temporary wound covering, the samples would appear to have sufficient potential in this area of biomedical application to warrant further study.

4.6 Balance of Hydrophilicity and Mechanical Properties

In designing a polymer for use as a temporary skin substitute, many property requirements need to be taken into consideration. These requirements were listed in the **Introduction** to this thesis (Table 1.1 on page 7). Apart from its obvious need to be biocompatible and non–toxic (which both P(HEMA) and PVP are), perhaps the most important properties which determine its performance characteristics are its **hydrophilicity** and **mechanical properties**. Expressed more specifically, the polymer must be able to absorb water and allow water vapour to be transmitted, and at the same time be strong and flexible. To obtain a satisfactory balance of these properties is fundamental to any research project such as this.

A summary of the equilibrium water contents (EWC) alongside the mechanical properties of the hydrogel sheets prepared in this work is given in Table 4.3.

Table 4.3 : A summary of the equilibrium water contents (EWC) and mechanical properties of the crosslinked P(HEMA) and P(HEMA–co–VP) hydrogels prepared in this work.

Hydrogel	EWC (%)	Tensile Strength (Pa)	Elongation at Break (%)	Young's Modulus (Pa)
P(HEMA)	34.7	3.40×10^4	45.9	7.42×10^4
P(HEMA–co–VP) / 95 : 5	35.6	4.30×10^4	62.9	6.89×10^4
P(HEMA–co–VP) / 90 : 10	37.8	2.96×10^4	42.5	6.93×10^4
P(HEMA–co–VP) / 85 : 15	39.4	2.80×10^4	36.0	7.83×10^4
P(HEMA–co–VP) / 80 : 20	40.9	3.43×10^4	56.8	6.03×10^4

As concluded at the end of previous Chapter 3 (page 103), increasing the VP content in the copolymer has a more clearly defined effect on the EWC than on the mechanical properties. Whereas the EWC increases consistently with VP content, the mechanical properties show no definite trend. But how do these results compare with those of similar studies ? Corkhill and Tighe [44] have also examined P(HEMA–co–VP) hydrogels as part of a much wider study. As their results in Table 4.4 show, while their EWC values agree quite closely with those obtained in this work, their mechanical property values are significantly higher. However, care should be taken in interpreting these comparisons since mechanical test data can vary quite considerably with sample preparation, sample dimensions (especially thickness) and testing conditions.

Table 4.4 : **Equilibrium water contents (EWC) and mechanical properties of crosslinked P(HEMA) and P(HEMA-co-VP) hydrogels obtained by Corkhill and Tighe [44].**

Hydrogel[a]	EWC (%)	Tensile Strength (Pa)	Elongation at Break (%)	Young's Modulus (Pa)
P(HEMA)	37.6	5.0×10^5	198	2.5×10^5
P(HEMA-co-VP) / 95 : 5	38.7	4.8×10^5	194	2.5×10^5
P(HEMA-co-VP) / 90 : 10	41.1	5.3×10^5	174	3.0×10^5
P(HEMA-co-VP) / 80 : 20	43.8	3.2×10^5	74	4.3×10^5

[a] also crosslinked with 1% w/w EGDMA
sample thickness (before hydration) = 0.2 mm [43]

In overall conclusion, the hydrogels which have been synthesized and characterized in this project, in accordance with its objectives, have been shown to possess hydration properties suitable for use as a wound covering. Furthermore, there is plenty of scope in their design for these properties to be controlled through variables such as film thickness, degree of crosslinking and copolymer composition. Less convincing, however, are their mechanical properties, especially tensile strength and pliability. Poor mechanical strength when hydrated has traditionally been the main disadvantage of synthetic hydrogels. How to improve this aspect of their performance is clearly one of the main objectives for further work. Some suggestions for further work are now made.

SUGGESTIONS FOR FURTHER WORK

In continuation of the research initiated in this project, the following suggestions for further work are now made :

1. Although copolymerization with N–vinylpyrrolidone, VP, increases the hydrophilicity of P(HEMA), thereby enhancing both its water absorption and transmission properties, it does not appear to bring about any significant improvement in mechanical properties. Since their rather poor mechanical strength when hydrated is the main weakness of P(HEMA) hydrogels, **other modifying comonomers** should be considered. Some possibilities are mentioned in the literature [3].

2. In this work, hydrogel sheets of uniform thickness 0.5 ± 0.1 mm were studied. Since sample thickness can be expected to have an effect on both water absorption / vapour transmission and mechanical properties (e.g., flexibility, pliability), **sheets of different thicknesses** should be produced and their properties compared.

3. The mechanical testing in this project had to be carried out using a 5 kgf load cell due to the unavailability of a smaller one. However, the maximum load at break of the hydrated hydrogel samples was of the order of only about 10 gf. Consequently, the level of sensitivity of the mechanical test results obtained here must be considered to be rather low, especially to relatively small changes in copolymer composition. A **smaller, more sensitive load cell**, capable of more precise and accurate measurements, is essential if the effects of microstructural changes are to be ascertained.

4. It would be useful at this initial stage of the research to compare the properties of samples prepared with those of **commercial materials**, both hydrogel–type and nonhydrogel–type wound dressings. This would indicate the level of performance that needs to be achieved.

5. An alternative method to elemental analysis for the determination of **copolymer composition** is the rapidly developing technique of **FT–Raman Spectroscopy**. Since FT–Raman spectra, amenable to quantitative analysis, can be conviniently obtained from solid samples, homopolymer mixtures of known compositions could be used as calibrants. Copolymer compositions could then be determined by the type of "absorbance ratio" method commonly used in IR spectroscopy.

6. It has been suggested [55] that a small amount of a **volatile solvent** added to the polymerization mixture can increase the **membrane macroporosity** of the hydrogel as it forms which, in turn, increases its capacity for subsequent water absorption. This is an interesting possibility which is worth investigating.

7. Finally, a **thermal analysis study of the water** in the P(HEMA) hydrogels, perhaps similar to that reported by Roorda et al [56], would shed light on the different levels of polymer–water interaction, as discussed in section 4.2 of this thesis and represented structurally in Fig. 4.2 on page 106. **Differential scanning calorimetry (DSC)** would be an ideal technique for this type of study. Although not in the mainstream of the project, this would be an interesting side study which would contribute to our knowledge and understanding of the thermodynamic nature of hydrogels.