Synthesis and Characterization of Hydrogels Based on Poly (N-vinyl formamide)

by

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Abstract

It is important to synthesize gels with lower toxicity and higher processibility to widen their applications. In this regard, the monomer N-vinyl formamide has an advantage over its widely used isomer acrylamide. A novel hydrogel, poly (N-vinyl formamide) (PNVF) was synthesized and its properties were measured when swollen to equilibrium in water. This gel was synthesized through free radical copolymerization/crosslinking with (N-vinyl formamido) ethyl ether (NVEE) initiated by VAZO-44 at 50˚C. Transparent gels of reproducible, uniform properties were obtained using NVEE, apparently due to nearly random copolymerization with NVF, likely due to similarity in the polarity if their propagating radicals.

These gels were characterized by varying monomer concentration from 10% to 25%. In addition to an increase in monomer concentration, crosslinker concentration was also varied from 1% to 3% on a mass basis, to study the effect on swelling degree and mechanical properties. Mechanical properties were calculated by conducting tensile tests to measure fracture stress, fracture strain, Young's modulus (E) and shear modulus (G). Young's modulus of PNVF gels increased from 138 kPa to 609 kPa, shear modulus increased from 49 to 212 kPa, fracture stress increased from 17 to 66 kPa where as the fracture strain decreased from 14.8 to 11.1 with an increase in crosslinking density. Other parameters like solubility parameter of the polymer and crosslink density were also calculated using established theories. The crosslink density was calculated from the shear modulus using the phantom network model of rubber elasticity and was on the order of 10^{-4}mol/cm^3. Combining this with measurements of the swelling degree, and Flory- Huggins solubility parameter (χ) of these polymers, was calculated to be ~0.42. PNVF gel

properties were found to be generally comparable to polyacrylamide, suggesting that PNVF gels crosslinked with NVEE could substitute for PAAm gels in applications.

PNVF hybrid gels were also synthesized by adding PNVF nanogels to PNVF single network. It was hypothesized that nanogels will improve fracture properties of these gels. Addition of PNVF nanogels to PNVF macrogels, however, did not have a significant effect on the fracture properties in the limited ranges tested.

Acknowledgements

I am deeply indebted to my supervisor, Dr. Stevin H. Gehrke, whose encouragement, guidance and support from the beginning to the end enabled me to develop an understanding of this subject. I appreciate him helping me finish my thesis even after I moved away from my university. In addition to my mentor, there are numerous people who have contributed to this work whom I would like to mention by name. I am extremely grateful to my committee members: Dr. Detamore and Dr. Berkland for their insight and suggestions. I would also like to thank all members of hydrogel research group, especially Patricia Huber, Tiffany Suekama, Anisha Patel, and Joe Lomakin for their immense help and support throughout this work. I want to thank Zahra Mohammadi for helping me with PNVF nanogel synthesis and Nathan Dormer for helping me with statistical analysis. I appreciate the financial support provided by the Department of Bio-engineering and the National Science Foundation (Grant DMR 0805264). I also want to thank all my close friends at University of Kansas for making life outside of the lab enjoyable. Finally, I would like to thank my family, especially my husband, Saboor Rashid, my sister, Yummna Aziz, and my parents for being with me every step of the way. This would not have been possible without their support. I dedicate this work to them.

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Chapter 1

Introduction

There is a constant struggle to discover and synthesize polymers from monomers with low toxicity and better mechanical properties.[1] Hydrogels are swollen materials which retain their shape due to a permanent three dimensional structure. These gels or polymer networks are cross-linked polymers that expand when in contact with a compatible solvent but cannot dissolve because of their cross-linked chemical structure.[1] These networks of polymer chains are water-insoluble, sometimes found as a colloidal gel in which water is the dispersion medium. Hydrogels are highly absorbent (they can contain over 99% water) natural or synthetic polymers. Hydrogels also possess a degree of flexibility very similar to natural tissue, due to their significant water content.

These water-insoluble gels have a wide range of applications. They are widely used in tissue engineering, drug delivery application, superabsorbent, protein separation via gel electrophoresis and soft contact lenses. The utility of the gels lies in their response to physiochemical stimuli, elastic and mass transfer properties.[1]

In this project, efforts were made to synthesize and characterize poly (N-vinyl formamide) (PNVF) gels. NVF is an isomer of acrylamide which is an extremely water soluble monomer and has a vast range of applications listed in Table 2-1. Polyacrylamide hydrogels are formed by the free radical copolymerization of the monomer acrylamide (AAm) and the cross linker N, N'-methylenebisacrylamide (bisacrylamide) in an aqueous solution.[2] However, the potential applications are hindered because of high monomer toxicity. Although polymers are usually inert, monomers are toxic and/or carcinogenic; hence it may be difficult to obtain

governmental approval for usage of such gels in biomedical applications.[3] Therefore, the main goal of this dissertation was to study other families of polyacrylamide with same mechanical properties but lower monomer toxicity.

N-Vinyl formamide (NVF), a water soluble isomer of acrylamide, is of particular interest due to its low toxicity and high reactivity for homo- and copolymerization[4] with numerous other polymers and crosslinkers. PNVF has a range of applications, and is used in water treatment, textile finishes, personal care products, adhesives, oil field chemicals, paper making, and radiation cure coatings etc.[5, 6] Due to the high biocompatibility of NVF, PNVF gels were synthesized in this thesis using a custom made crosslinker, (N-vinylformamido) ethyl ether (NVEE). In this project, mechanical properties of this gel were studied and reported in detail. Swelling degree and mechanical properties are vital to know before it becomes possible to use a novel hydrogel. These mechanical characteristics like fracture stress, fracture strain, Young's modulus, crosslink density, describes the mechanical limits of a material application. The equilibrium swelling degree or sorption capacity is the most important property of a hydrogel and directly influences other properties. Swelling degree also gave an idea of the mechanical properties of the gel. Swelling degree varies depending upon the formulation. Increasing the monomer concentration decreases the swelling degree of the gel because the number of entanglements increases in the network, and increases the crosslinking efficiency. Static properties of PNVF gels were compared with polyacrylamide gels to determine the differences in their mechanical strengths. All tests were done in tension. These tests needed extra care as hydrogels are weak in nature and tend to rupture while hanging from the mounting clamps. Self lubrication and slippage from the mounting clamp was also a common hindrance in reproducibility.

There is not a clear established procedure for the synthesis and characterization of PNVF gels in literature. Hence, it was important to characterize several different parameters to give an idea of the mechanical limits of this hydrogel. Several gel samples with varying monomer/ crosslinker concentrations were synthesized and characterized in Chapter 4.

The first three chapters of this dissertation provide detailed background information. Chapters 2 and 3 discuss a few major topics, including the synthesis of hydrogels, formation of a network structure through crosslinking and general background of poly (N-vinyl formamide), polyacrylamide and poly (N-vinyl pyrrolidone). Chapter 2 provides an in depth discussion on why these polymers are important and how they are related to each other. Chapter 3 is structured around mechanical properties as well as swelling degree. Swelling degree is one of the most important data points of this study as it can be compared to the literature value. Other fracture properties are not commonly listed in the literature and the theory behind them is undeveloped. Two fundamental classical theories are discussed in this chapter which differs from each other by a factor of 2. The affine model is more commonly used in the literature but from this study, it was concluded that phantom model gave more realistic results. Results were tabulated using both models to compare the difference between the models.

In addition to PNVF macrogels, PNVF nanogels were also synthesized and added to the network to improve mechanical properties. Fillers are added to elastomers to improve tear strength, static strength and improve abrasion resistance. Addition of PNVF nanogels increased the cross-linking among the polymer chains making the gel more elastic. Mechanical properties of hybrid gels were tested and compared against the single network PNVF gels.

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Chapter 2

Synthesis of Hydrogels

2.1 SYNTHESIS OF HYDROGELS

Hydrogels have been used in different application fields from pharmaceutical industry to agriculture and drug delivery systems. They can be made by a number of methods, but generally, they are synthesized by direct cross-linking of polymers or copolymers in solution using a small amount of cross-linking agent. Though, sometimes they are synthesized by treating the aqueous solution of a chosen water-soluble polymer with γ rays. One of the most widely used methods is a use of a chemical initiator to initiate the free radical polymerization. The focus of this dissertation is to synthesize poly (N-vinyl formamide) hydrogels via a custom made crosslinker (N-vinylformamido) ethyl ether (NVEE) and to compare results with the more well known polyacrylamide gels. In these preparation techniques, a chemical initiator is used which successfully initiates the reaction and a small amount of NVEE becomes part of two different polymer chains, leading to formation of a complete network structure.

This chapter provides background information on poly (N-vinyl formamide) in detail which was the main polymer under study. Poly (N-vinyl formamide) gels have only been synthesized in the industry a few times and hence there is no established technique in the literature which details the steps required for PNVF gel synthesis or characterization. This chapter also provides background information on polyacrylamide which is a better known polymer and is one of the best characterized hydrogel in the industry. The properties of these two gels are compared in Chapter 4.

2.2 POLY (N-VINYL FORMAMIDE)

NVF is the key compound in the synthesis of linear cationic polymers with primary amine groups. It has a molecular weight of 71 g/mol and a high boiling point of 80˚C at 10mbar. NVF is a water soluble monomer that provides a wider range of application due to its lower toxicity and higher reactivity than the more common acrylamide monomer. NVF, being a liquid monomer, makes it is easier to scale up and is commercially available in pure form. Usually, NVF is also soluble in a large variety of organic solvents. As NVF is an amide, its solution maybe expected to have considerable compatibility or solvency for a variety of materials such as inorganic salts. Polymerization of NVF is carried out by free radical polymerization. Free radical polymerization has three principal steps:

1. Initiation of the active monomer
2. Propagation or growth of the active (free-radical) chain by sequential addition of monomers.
3. Termination of the active chain to give the final polymer product.

Propagation rate of free-radical polymerization is very rapid. This may be due to the ratio $k_p/k_t^{1/2}$ (where k_p is the propagation rate constant and k_t is the termination rate constant) being extremely high in a free-radical polymerization.

In this study, PNVF gels were synthesized in the hope that they could replace the more widely used polyacrylamide gels. In these preparation techniques, VA-44 successfully initiated the reaction and a small amount NVEE entangled the chains into a complete network structure. Vinyl formamide (VF) is an isomer of acrylamide, which readily polymerizes to poly (N-vinyl formamide). Polymers incorporating NVF can be used in many of the same applications as acrylamide polymers. NVF is neither a carcinogen nor a neurotoxin. PNVF was copolymerized

with the well known acrylamide crosslinker, bisacrylamide, but that resulted in weak, pliant, transparent gels, which were hard to handle and led to irreproducible results. This reaction can be explained by the following discussion. When two monomers, M_1 and M_2 are copolymerized, there are four main propagation reactions as follows

$$P_1^{\cdot} + M_1 \xrightarrow{k_{11}} P_1^{\cdot} \qquad \textbf{(1)}$$

$$P_1^{\cdot} + M_2 \xrightarrow{k_{12}} P_2^{\cdot} \qquad \textbf{(2)}$$

$$P_2^{\cdot} + M_1 \xrightarrow{k_{21}} P_1^{\cdot} \qquad \textbf{(3)}$$

$$P_2^{\cdot} + M_2 \xrightarrow{k_{22}} P_2^{\cdot} \qquad \textbf{(4)}$$

Where P represents a propagating chain with the terminal unit designated by the respective monomer and k represents the reactivity ratio of the respective reaction.

From this, reaction constants for monomers 1 and 2 can be defined

$$r_1 = \frac{k_{11}}{k_{12}} \qquad \text{[Eq. 2-1]}$$

$$r_2 = \frac{k_{22}}{k_{21}} \qquad \text{[Eq. 2-2]}$$

For 'ideal' copolymerization it is important for all four of these reactions to proceed. Three main types of copolymer structures that can result from a combination of these reactions are alternating, random and block copolymers. Reactions (1) and (4) will strictly yield alternating copolymer regime. This is when both reactivity ratios are zero (i.e., $k_{11} = k_{22} = 0$). The two polymer chains will rarely interact with each other. On the other hand, if both reactivity ratios are small but not zero, the comonomer sequence will not be completely alternating but will have an alternating tendency.[2] This is more commonly known as a block copolymer structure.

Finally, when both reactivity ratios equal unity, there is no preferential monomer incorporation into the propagation chain. This results in a random or 'ideal' copolymerization. Random copolymerization leads to equal amounts of both monomers incorporated in the network. However, reactivity ratios for less common monomers like NVF can be calculated using a *Q-e* scheme proposed by Alfrey and Price.[2] Using this scheme, the propagation rate constants can be calculated using the following equations

$$r_1 = \frac{k_{11}}{k_{12}} = \left(\frac{Q_1}{Q_2}\right)\exp[-e_1(e_1 - e_2)] \qquad \text{[Eq. 2-3]}$$

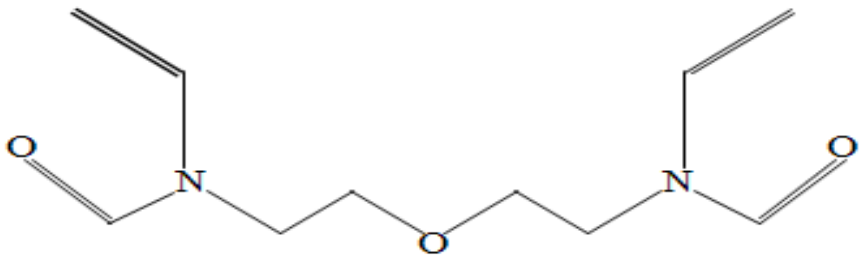
$$r_2 = \frac{k_{22}}{k_{21}} = \left(\frac{Q_2}{Q_1}\right)\exp[-e_2(e_2 - e_1)] \qquad \text{[Eq. 2-4]}$$

Values of *Q* and *e* are tabulated for some commercially important monomers from which the values of less common monomers can be estimated. The rate constant, in general, is independent of the nature of the initiator and solvent in a free radical copolymerization.[1] Using equations 2-3 and 2-4, the amount of monomer in the solution f_i, and the polymer composition in the solution F_i can be calculated. Therefore, the relationship between f_i, F_i and r_i can be compressed into equation 3-5.

$$F_1 = \frac{r_1 f_1^2 + f_1 f_2}{r_1 f_1^2 + 2 f_1 f_2 + r_2 f_2^2} \qquad \text{[Eq. 2-5]}$$

This equation shows that when reactivity ratios equal unity $F_1 = f_1$ which is the case of ideal or random copolymerization as mentioned above.

To form a well-defined gel, it is vital to have a generic crosslinker which randomly copolymerizes, reduces side reactions and increases mechanical strength. (N-vinylformamido) ethyl ether (NVEE) is a custom made crosslinker which proved to work much better with PNVF than most of the generic polyacrylamide crosslinkers in the industry. Mechanical properties of PNVF hydrogels synthesized from this crosslinker were comparable to polyacrylamide gels as shown in the results section in Chapter 4.



NVEE

Fig. 2-1-(N-vinylformamido)ethyl ether (NVEE) is a derivative of NVF. It increased mechanical properties and resulted in well-defined PNVF gels.

High purity NVF can undergo radical polymerization very easily and can copolymerize with a large number of other monomers to form products of various molar masses. In addition to this, polymers bearing primary amine functionalities are of great interest due to their high reactivity for various post derivatization reactions as well as their cationic nature in appropriate pH regimes.[3] Strongly basic polyvinylamines (PVAm) can be easily synthesized by cleaving the formyl group by hydrolysis from the mostly water-soluble polymers. NVF also gains a lot of attention due to favorable environmental aspects because the polymer solutions are completely free of residual monomer after hydrolysis to polyvinylamines.[4] There are numerous applications for such cationic polymers because of their high charge density and high reactivity of the

primary amine groups. PVAm are important for their biological applications and are used as a support for enzymes and other active components. These cationic forms show excellent adhesion to anionically charged biological surfaces, such as cellulose, skin, and hair.[5] They are also used for chromatographic support, heterogeneous catalysis and biocompatible implant layers.[5] These cationically active polyelectrolytes are also desired for modification of inorganic substrates for the synthesis of functional polymer-inorganic hybrid materials with well-defined properties. Unfortunately, the simplest precursor monomer to PVAm, vinylamine, is unavailable because it tautomerizes to acetaldehyde imine.[6] Therefore, PVAm can only be synthesized through indirect routes, such as hydrolysis of polyacylamide, poly (N-vinylcarbamate), and poly (N-vinylacetamide). Out of all these reactions vinyl formamide is an excellent precursor monomer with short reaction times and relatively mild hydrolysis conditions. The formamide group readily undergoes hydrolysis resulting in a controlled amount of PVAm segments. Figure 2-1 shows the process of hydrolysis. The basicity, hydrophilicity, solubility or reactivity can be controlled by the use of suitable comonomers and by varying hydrolysis conditions.

NVF is a multifunctional molecule: on one hand the C=C double bond is available for addition and polymerization reactions, and on the other hand there is the weakly acidic proton on the nitrogen flanked by the formyl and vinyl groups.[4] This allows NVF to undergo several different chemical reactions and eases the hydrolysis process.

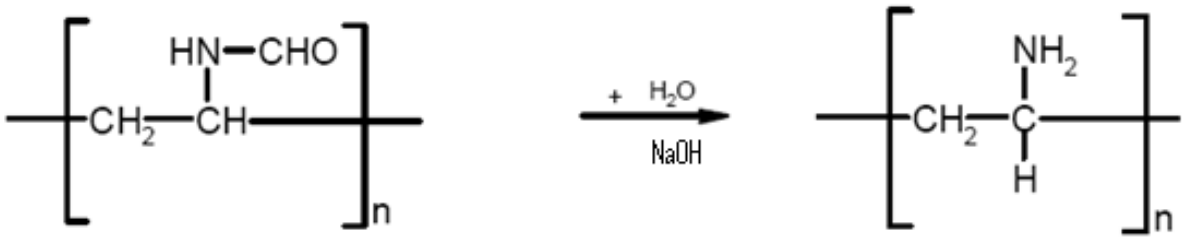


Fig. 2-2 Basic hydrolysis of PNVF to yield polyvinylamines.

2.3 POLYACRYLAMIDE

Amines of acrylic and methacrylic acids have been widely studied in the past. Methacrylamide, acrylamide and diacetoneacrylamide are some of the derivatives of methacrylic and acrylic acids.[7] Among these, acrylamide by far is the most important monomer of this group. Polyacrylamide is an extremely reactive water soluble molecule. This polymer has numerous applications, as listed in Table 2-1.

Table 2-1 Advantages of polyacrylamide.

Advantages of polyacrylamide	
soil conditioner	surface coatings
in waste water treatment	textile treatments
adhesives	improvements of cements
dispersants	water purification
flocculants	paper treatment
printing plates	soil stabilization
viscosity modifiers and thickeners	well drilling
fiber dying and modification	boiler water treatment
leather substitutes	hair sprays,
in the cosmetic	ion-exchange resins
paper sizing	pigment binders
protective colloids in photographic emulsions	polyester binding resins
gel electrophoresis	textile industries

Due to this vast list of applications of this monomer, attempts are being made to increase its application in the biological industry as well. Acrylamide, methylacrylamide, and many of the simple nonfluorinated, related monomers are generally water soluble, which makes them easy to work with. Usually, they are also soluble in a large variety of organic solvents. As they are amides, their solutions are expected to have considerable compatibility or solvency in inorganic salts. Polymerization of acrylamide is usually carried out by free radical polymerization and anionic means using a variety of initiating systems.[7] These polymers of acrylamide usually yield high molecular weights. These are covalently cross-linked materials, whose chemical and physical bases are well understood. The network formation of such gels depends on many factors such as monomer and crosslinker concentration and the time of reaction. Mechanical properties of polyacrylamide (PAAm) gels have been studied extensively.[7] It is important to study fracture properties as they give a better estimate of what kind of application the gels are suitable for, or for an approximation of how much load the gels can bear before deformation. Figures 2-3 and 2-4 show fracture properties of PAAm gels from the literature.[9]

2.4 POLY (N-VINYL PYRROLIDONE)

N-vinyl pyrrolidone (NVP, **1**) is a yellowish which is highly miscible in water. It is a precursor to poly (N- vinyl pyrrolidone) (PVP), an important synthetic material. Polyvinylpyrrolidone, also called by the trade name Povidone or the acronyms PNVP or PVP, is one of the numerous products of acetylene chemistry. PVP is normally prepared by thermal polymerization of N-vinyl pyrrolidone.[10] PVP is a nontoxic polymer which crosslinks into an important class of hydrogels useful for a variety of applications, especially in the medical industry.

Medical applications of PNVP hydrogels are now wide spread due to their excellent blood compatibility, and have therefore been widely used in applications including drug delivery

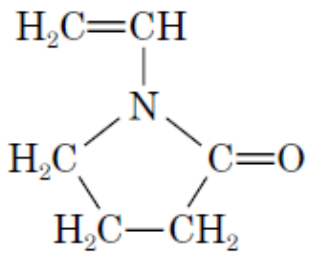
systems, artificial muscles, wound dressing and tissue engineering.[11,12] Hydrogels formed from copolymerization of PNVP and methacrylates are used in many applications in biomedical fields, especially in contact lenses.[13] PVP is biocompatible and attracts a lot of attention in the biomedical field due to its low toxicity. These copolymers have been prepared *via* photo- and thermal polymerization using ethylene glycol dimethacrylate (EGDMA). These lenses include high polarity/ hydrophilicity of NVP.[14] For example, copolymers of NVP and 2-hydroxyethyl methacrylate (HEMA, **2)** gives hydrogel contact lens material with water content in the ranges of 40-60%.[15] But, copolymerization of NVP and methacrylates monomers, in the presence of crosslinker such as ethylene glycol dimethacrylate (EGDMA, **3**),[16] often show inconsistent quality due to compositional drift during the course of polymerization,[17] such as poor control lens dimension, modulus, and water content.

The inconsistency is quality shows there is a need to synthesize and develop a procedure to copolymerize NVP. As discussed earlier, commonly used crosslinkers like EGDMA and allyl methacrylate (AMA, **5**)[18] poorly copolymerizes NVP and methacrylate monomers. AMA results in high water content due to low crosslink density. It was demonstrated by Bambury *et al.* that NVP UV copolymerizes well with vinyl carbamates and carbonates in the presence of a photo-polymerization initiator, 2-hydroxy-2,2-dimethyl acetophenone.[19] This led to the discovery of a crosslinker which contained a vinyl carbonate and a methacrylate group, methacryloxyethyl vinyl carbonate (HEMAVC, **4**) which had both methacrylate and vinyl carbonate groups allowing it to copolymerize well with both methacrylate and NVP. Hydrogels synthesized from this crosslinker were compared to the more generic EGDMA, with a combination of a UV initiator (BME) and a thermal initiator (TBO). HEMAVC enhanced copolymerization of NVP and methacrylates under UV and showed better incorporation than AMA.[15] EGDMA, on the

other hand, worked better for copolymerizing HEMA/NVP, but was not good enough for NVP alone.

A series of experiments carried by Yu-Chin Lai at Bausch & Lomb showed vinyl carbonate copolymerizes NVP better than a methacrylate group. HEMAVC consistently gave NVP based hydrogels of better quality than those synthesized by EGDMA. According to this study, the ability of incorporating NVP decreases in the order of HEMAVC, AMA, and EGDMA. Researchers also noticed a wide difference in reactivity during the copolymerization of NVP and methacrylates, including EGDMA, all methacrylate groups were used up before a significant number of NVP molecules incorporated with a methacrylate[7] suggesting block copolymerizing pattern. Though, a stiff hydrogel film was obtained when high (50%) amount of crosslinker was used.

Arndt *et al*. also established that N-vinyl-2-pyrrolidone (VP) cannot copolymerize well with methacrylates due to compositional drift during the course of polymerization. This is due to the difference in reactivity of the two monomers. The presence of a significant amount of water soluble fractions can impact the end use of copolymers.[13] This suggests a need for chemical initiator to crosslink NVP with methacrylate groups.



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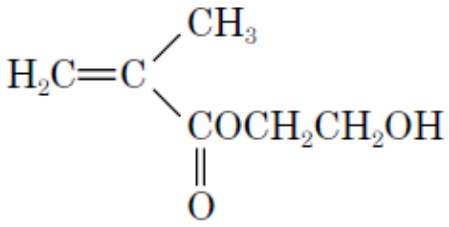

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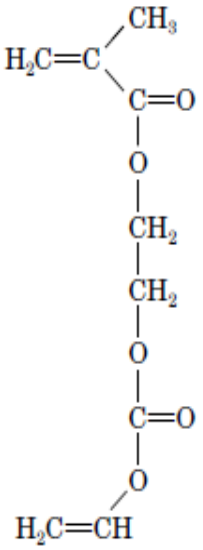

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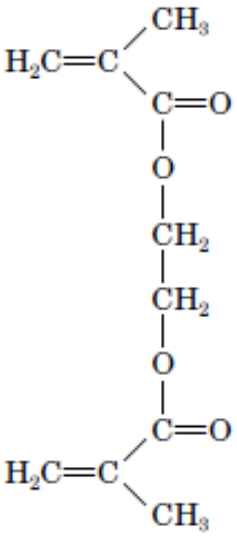

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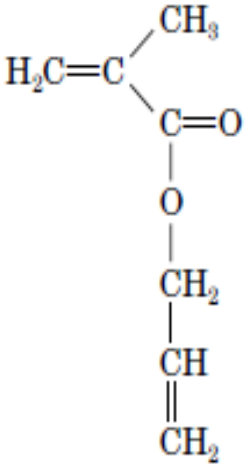

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Fig. 2-3. This figure shows the structures of various monomers under study. N-vinyl pyrrolidone (NVP,1), 2-hydroxyethyl methacrylate (HEMA, 2),ethylene glycol dimethacrylate (EGDMA, 3), methacryloxyethyl vinyl carbonate (HEMAVC, 4) and allyl methacrylate (AMA, 5)

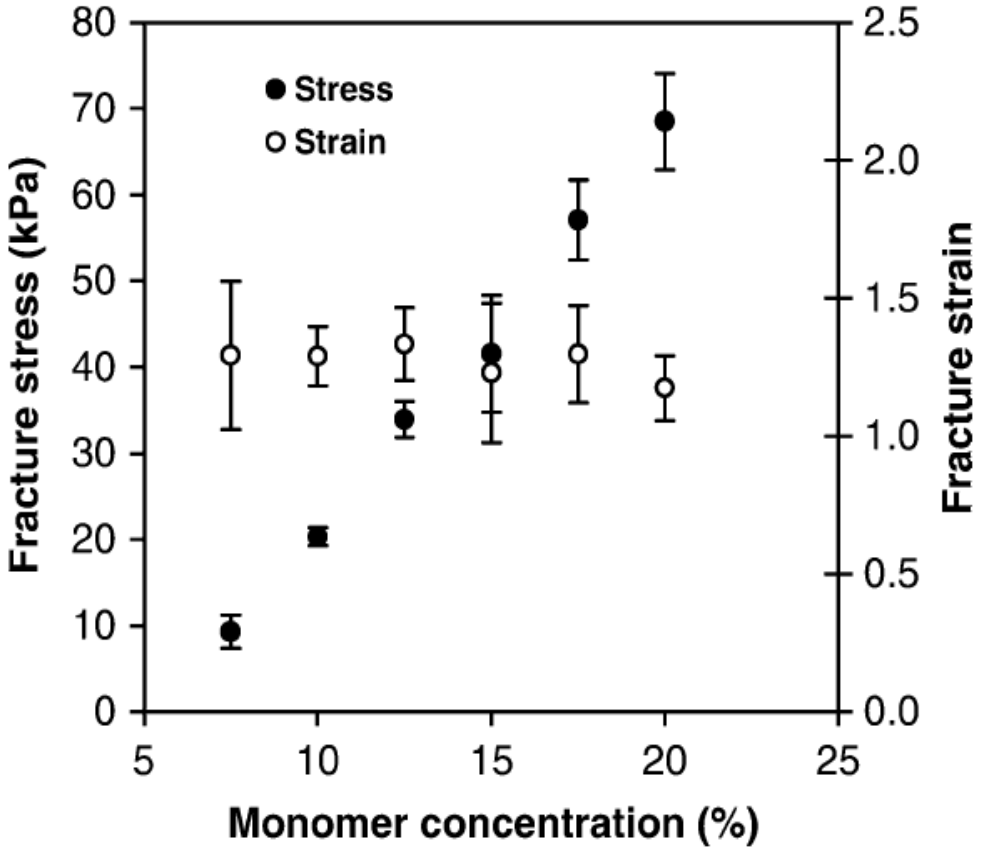

Fig. 2-4. Fracture stress and strain for PAAm gels (M/C: 40) associated with gel concentration. Taken from [9].

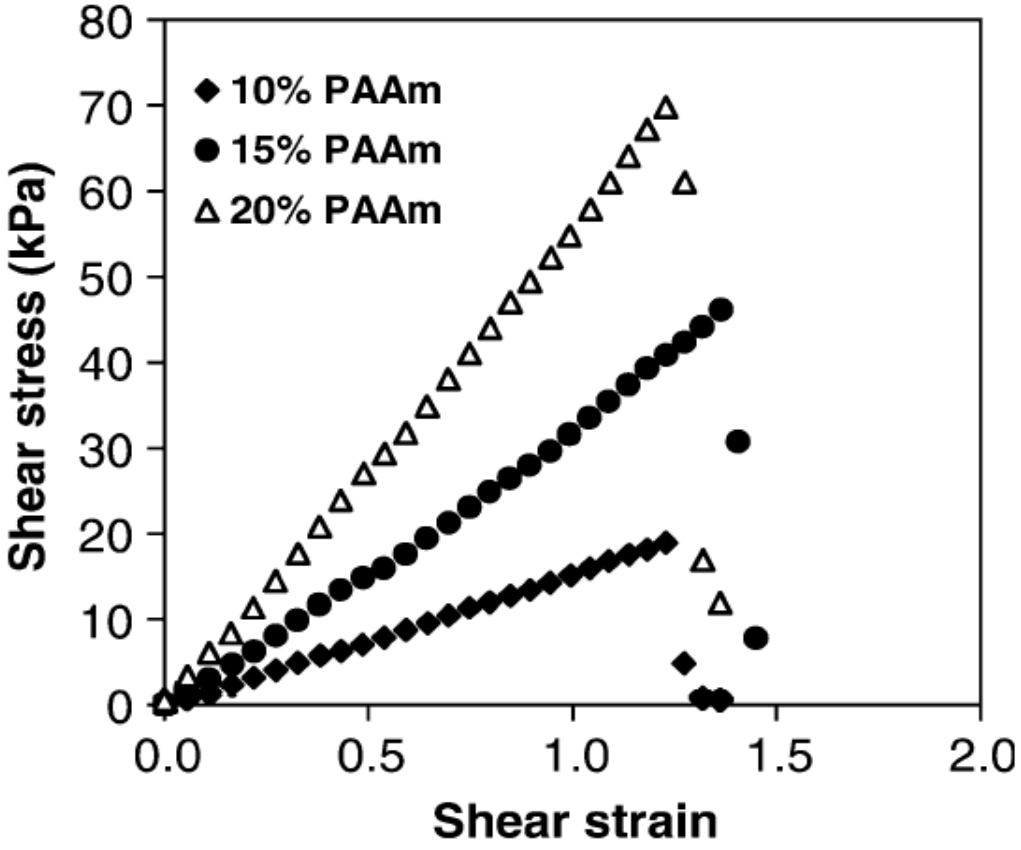

Fig. 2-5 Shear stress versus shear rate at increasing monomer (PAAm) concentration at room temperature.Taken from [9].

PVP have molecular weights from 2,500 to about 1 million. Higher molecular weight products are polymerized in aqueous solution mostly using hydrogen peroxide initiator.[19] The polymers thus obtained have unstable hydroxyl and carbonyl end groups. In addition to this, crosslinking of linear PVP is achieved by gamma radiation or by the treatment of the polymer with persulfate or with hydrazine and hydrogen peroxide. The products obtained are only slightly cross-linked and form soft gels.[19] Popcorn polymerization is suggested by Straub et al., which leads to highly cross-linked PVP. A radical mechanism is assumed, however, no radical initiator is required. The main source of radical comes from the rupture of polymer chains already formed.

This technique of polymerization uses dangerously high temperature to form the bifunctional group. The rupture of polymer chains cannot be controlled and shows an independent relation between the swelling and crosslinking of polymer chains.

Potassium persulfate is a well known initiator for polymerization of vinyl type monomer. Thermal decomposition of persulfate anions produces sulfate ion radicals, which causes chain propagation. However, in case of PVP, crosslinking and/or chain scission can be observed in aqueous solutions of polymers in the presence of persulfate depending on the content of persulfate in the mixture.[20]

Some other competing reactions, such as oxidative degradation and ring opening of lactam ring have also been seen. Such complications were also seen when persulfate was used to initiate polymerization in NVF. On the other hand, 2, 2'-azobis (N,N'-dimethylene isobutyramidine) dihydro chloride(VA-044) proved to be a better choice of initiator with no apparent side reactions.

NVP slightly differs from NVF in structure. PNVP is widely used in soft contact lenses and in drug delivery applications but its use is limited due to the poor understanding of its characteristic behavior and network structure. Literature suggests a need for a crosslinker which could crosslink these polymers without using UV-initiated polymerization. There have been numerous cases where researchers have tried to crosslink PNVP with Bis, with no success. NVEE, the novel crosslinker incorporated N-vinyl formamide exceptionally well in a network. The polarity of NVF is similar to that of NVEE as they have similar structures. It is thought that vinyl groups on both ends of this structure helps grow the chains in a random pattern which EGDMA, *N,N'*-methylenebisacryalmide (bisacrylamide) and other commonly used crosslinkers lack. A thermal chemical initiator, VA-44, is used to initiate NVF polymerization and is highly suggested for NVP polymerization as well. NVEE is expected to copolymerize well with NVP just as it does with NVF, forming a stable hydrogel.

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Chapter 3

Swelling Degree and Mechanical Properties of Hydrogels

3.1 SWELLING DEGREE

Swelling degree is the most important property of a hydrogel which directly influences the rate of water sorption, the permeability of drugs, and the mechanical strength of the gel.[1] It represents the amount of solvent a gel will absorb at a given temperature and is defined as the ratio of swollen gel volume to the volume of dry polymer. This is usually quantified by Q. Swelling leads to a three- dimensional expansion in which the network absorbs solvent and reaches an equilibrium degree of swelling at which the decrease in free energy due to mixing of the solvent with the network chains is perfectly balanced by the increase in free energy accompanying the stretching of the chains (elasticity).[2]

According to Flory, total free energy, and therefore swelling pressure, is represented as the sum of the individual contributions from polymer-solvent interactions, network elasticity, ionic osmotic pressure and electrostatic effects.[3] Swelling pressure is therefore represented as follows:

$$P_{ext} = \Pi_{mix} + \Pi_{elas} + \Pi_{ion} + \Pi_{elec} \qquad \text{[Eq. 3-1]}$$

Where Π_{mix} represents the contribution from polymer-solvent interactions, which if favorable, increases swelling. Π_{elas} represents the contribution due to network elasticity which arises from restraints on swelling imposed by cross-links, opposing dissolution. For ionized gels, Π_{ion} represents the osmotic pressure of the counter-ions and Π_{elec} accounts for the interactions between the charged groups in the gel.

Flory-Rehner then went on and combined these terms to define the free swelling of a nonionic gel in equilibrium with a solvent by

$$ln(1-\varphi_2)+\varphi_2+\chi\varphi_2{}^2+V_1\rho_x\varphi_{2,r}\left[\left(\frac{\varphi_2}{\varphi_{2,r}}\right)^{\frac{1}{3}}-\frac{1}{2}\left(\frac{\varphi_2}{\varphi_{2,r}}\right)\right]=0 \qquad \text{[Eq. 3-2]}$$

Where V_1 is the solvent molar volume, χ is the polymer-solvent interaction parameter, ρ_x is the effective crosslink density, φ_2 is the polymer volume fraction ($\varphi_2 = 1/Q$) and $\varphi_{2,r}$ is the polymer volume fraction at network formation. This equation shows that equilibrium swelling degree relies mainly on χ, the polymer-solvent interaction parameter, and ρ_x, the effective crosslink density. The effective crosslink density is the concentration of elastically active chains in the polymer network and is reported on the basis of moles of chains per cubic centimeter of dry polymer.[4] Most studies of networks in swelling equilibrium give values for the crosslink density or related quantities that are in agreement with those obtained from measurements of mechanical properties.[5] By measuring the equilibrium swelling behavior of a hydrogel, its modulus maybe predicted and vice versa. Crosslink density is directly related to polymer deformation. A stress-strain response of ideal network under uni-axial extension is given by

$$\tau = {}^{F}/_{A} = G(\lambda-\lambda^{-2}) \qquad \text{[Eq. 3-3]}$$

where τ is the engineering stress; F is the applied force; A_0 is the cross-sectional area of the swollen polymer in unstrained state and G is the shear modulus. $\lambda = {}^{L}/_{L_0}$, where L is the sample length under strain and L_0 is the un-deformed sample length. G was calculated from plots of stress versus $(\lambda-\lambda^{-2})$ at low strains. At low strains, this yields a straight line, the slope of the

line representing the modulus. The effective crosslink density was then calculated from the modulus by:

$$\rho_x = {2G}\Big/{RT\varphi_{2,r}^{2/3}\varphi_2^{\frac{1}{3}}} \qquad \text{[Eq. 3-4]}$$

ρ_x is the crosslink density calculated from the dry polymer volume. This equation accounts for the polymer volume fraction at the time of synthesis as well as the swelling ratio.

In addition to these parameters, other swelling parameters like average molecular weight between cross-links (M_c) and mesh size were also calculated from swelling degree and modulus using the following equation

$$M_c = \frac{RT\rho_2}{GQ^{1/3}} = \frac{1}{\rho_x V_2} \qquad \text{[Eq. 3-5]}$$

Where ρ_2 is the polymer density (ρ_2= 1.2g/cm^3) and V_2 is the polymer molar volume.

3.2 MECHANICAL PROPERTIES

The prime consideration in determining the general use of a hydrogel is by determining its swelling degree and mechanical properties, that is, its deformation and fracture under stress. Hydrogels vary widely in their mechanical behavior depending on the degree of crystallinity, degree of cross-linking, and the values of glass transition T_g and crystalline melting temperature T_m. Crystalline melting temperature is the melting temperature of the crystalline domains of a polymer sample while glass transition temperature is the temperature at which amorphous domains of a polymer takes on characteristic properties of a glassy state. Strength is lost at or near T_g for an amorphous polymer and at or near T_m for a crystalline polymer. High degrees of

crystallinity or crosslinking are characterized by high T_g which results in a network of high strength and low extensibility. Whereas, high extensibility and low strength in polymers translates into low degrees of crystallinity and crosslinking and low T_g values.[6]

Mechanical behavior of a polymer can be characterized by its stress-strain properties [7].This often involves observing the behavior of a polymer as one applies tensile stress to it in order to elongate (strain) it to a point where it ruptures. Three important quantities characterize the stress-strain behavior of a polymer:[6]

1. Modulus (E). The resistances to deformation as measure by initial stress divided by strain $\Delta L/L$. An ideal elastic solid obeys Hooke's Law; $\sigma = E*e$, where the linear strain e is the change in length divided by the original length when a static stress σ, force per unit cross-sectional area, is applied to stretch a piece of the material of uniform cross-section.
2. Fracture or Ultimate Stress: The stress required to rupture the sample.
3. Fracture or Ultimate Strain (Elongation): The extent of elongation at the point where the sample ruptures.

In an ideal elastic solid, stress σ leads to an instantaneous strain ε, and on removal of the stress; the strain instantaneously reverts to zero. Strain is normally restricted to small values before fracture. There are five important ways in which the mechanical behavior of a polymer may deviate from this ideal behavior.[8]

1. time-dependence of response
2. non-recovery of strain on removal of stress
3. non-linearity of response (e not proportional to σ)
4. large strains without fracture

5. anisotropy of response

3.2.1 Static Properties

Static mechanical testing involves applying a constant stress or strain to a gel sample in tension or compression. These tests are often referred as stress-strain tests. It is important to understand what causes polymer chain failure or rupture. Fig. 3-1 shows the qualitative difference between a ductile and a brittle material. The type of fracture dealt in this study is called brittle fracture. The process of brittle fraction involves two stages, crack initiation and crack propagation. Although, only samples without any obvious flaws or bubbles were used, it is assumed in practice that minor cracks always exist in real polymer networks. Stress increases the tip of the crack in a gel to which a uniform static stress is applied normal to the crack.

Three main sections can be recognized on this plot (Fig.3-1). The initial linear relationship between stress and strain, this linear region is known as the elastic (as opposed to the rubber elastic) region. This is mostly a reversible deformation. The slope of this initial region is referred as Young's modulus (E). As strain increases, the curve starts deviating from the linear region and enters a second, non-linear region of the curve. This is more commonly known as the plastic region. This deformation by definition is irreversible and may lead to a fracture point. Finally the point at which the curve abruptly falls down is known as the fracture point. This is where the fracture stress and strain is recorded and it is at this point where the material no longer holds any structural integrity and loses the ability to extend any further. Fracture stress and strain are two ultimate properties. Young's modulus can be determined from this stress-strain curved.

Material toughness can also be calculated from these plots. Toughness is the resistance of a material to failure when suddenly exposed to stress. It is more commonly defined as the area

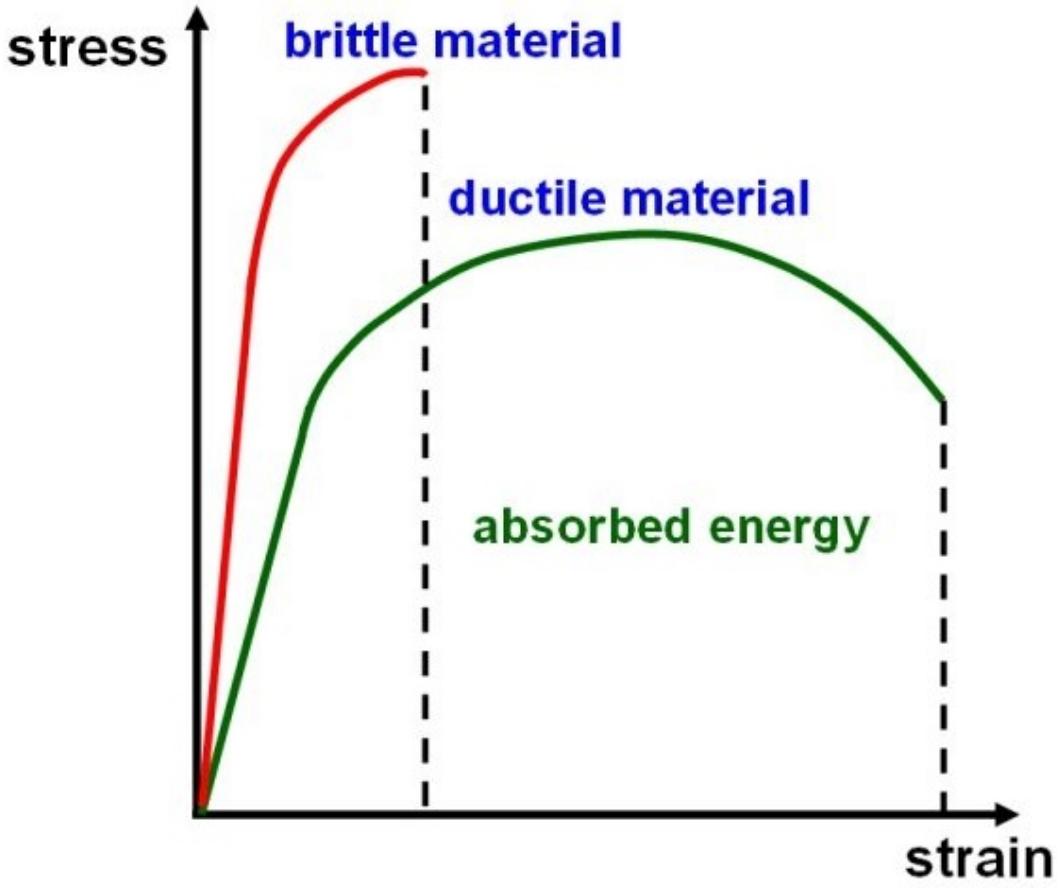

Fig. 3-1 Engineering stress-stress curve indicating the difference between ductile and brittle material. The area under the curve represents toughness or the energy absorbed to fracture the material. Taken from [13].

under the curve up to ultimate fracture point. It is usually measured in Pascals or Joules per cubic meter, which is the amount of energy per unit volume required to cause a fracture in a material.

3.2.2 Dynamic Properties

When the samples undergoes repeated small-amplitude strains in a cyclic manner, molecules perturbed store a portion of the imparted energy elastically and dissipate in the form of heat. The quantity E', Storage modulus, is a measure of the energy stored elastically, whereas E'', Loss modulus, is a measure of the energy lost as heat. Therefore

$$\frac{E''}{E'} = tan\delta \qquad \text{[Eq. 3-6]}$$

Where tan δ is called the loss tangent. δ is the angle between the in-phase and out-of-phase components in the cyclic motion. Figure 3-2 shows the dynamic mechanical behavior of an ideal polymer.

3.3 MODELS OF RUBBER ELASTICITY OF GELS

In the 1940's the statistical theories of rubber elasticity were developed that represents the network as a system of ideal chains with no interaction assuming zero volume. There are two fundamental classical theories which simplify these assumptions and are known as affine theory and phantom theory. The affine model assumes that deformation is affine or to say that the network chains move in a simple linear fashion with the macroscopic deformation. Most theories invoke a Gaussian distribution, however, non- Gaussian theories have been developed for network chains that are unusually short or stretched close to the limits of their extensibility.[6] This theory also assumes intermolecular forces are independent of the deformation, that is, the network elasticity is completely intramolecular.

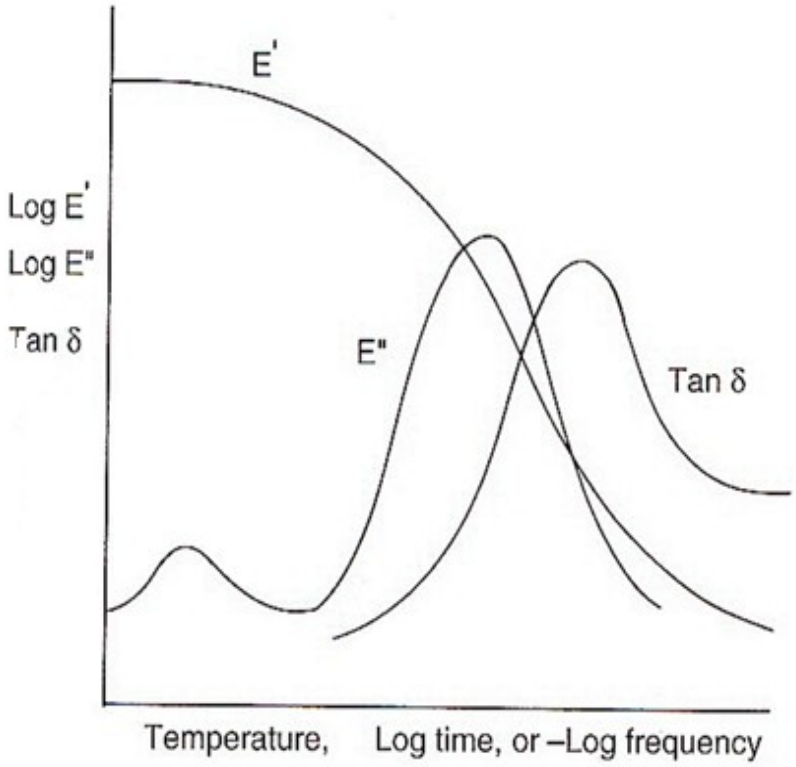

Fig. 3-2 Dynamic mechanical behavior of an ideal polymer. Where E' is the storage modulus, E" is the loss modulus and Tan δ is their ratio. Taken from [12].

An alternative to the affine model is the 'phantom chain' model where polymer chains are permitted to pass through one another as if they had zero cross-sectional area. According to this model, cross-links undergo considerable fluctuations in space which occur in an asymmetric manner so as to reduce the strain.

As shown in the Figure 3-3, at low deformations, chain junction entangling suppresses fluctuations of the junctions and the deformation is relatively close to the affine limit whereas an increase in elongation disentangles the chains which increases the magnitude of fluctuations. This causes the chains to sense a smaller deformation than that imposed, making the deformation more non-affine. Therefore, phantom theory predicts moduli that are lower than predicted by affine model. The phantom model works better with hydrogels, and hence, was used consistently throughout this study.

Based on these postulates, the relationship between stress and strain, for uni-axial extension or compression is represented by

$$\tau = {}^{F}/_{A} = G(\lambda - \lambda^{-2}) \qquad \text{[Eq. 3-7]}$$

These theories, however, are in disagreement with the Mooney-Rivlin equation where the modulus actually decreases with the increase in λ or the principal extension ratio. This equation is the correction of the discussed models.

$$f^{T} = 2\left(C_1 + \frac{C_2}{\lambda}\right)\left(\lambda - \frac{1}{\lambda^2}\right) \qquad \text{[Eq. 3-8]}$$

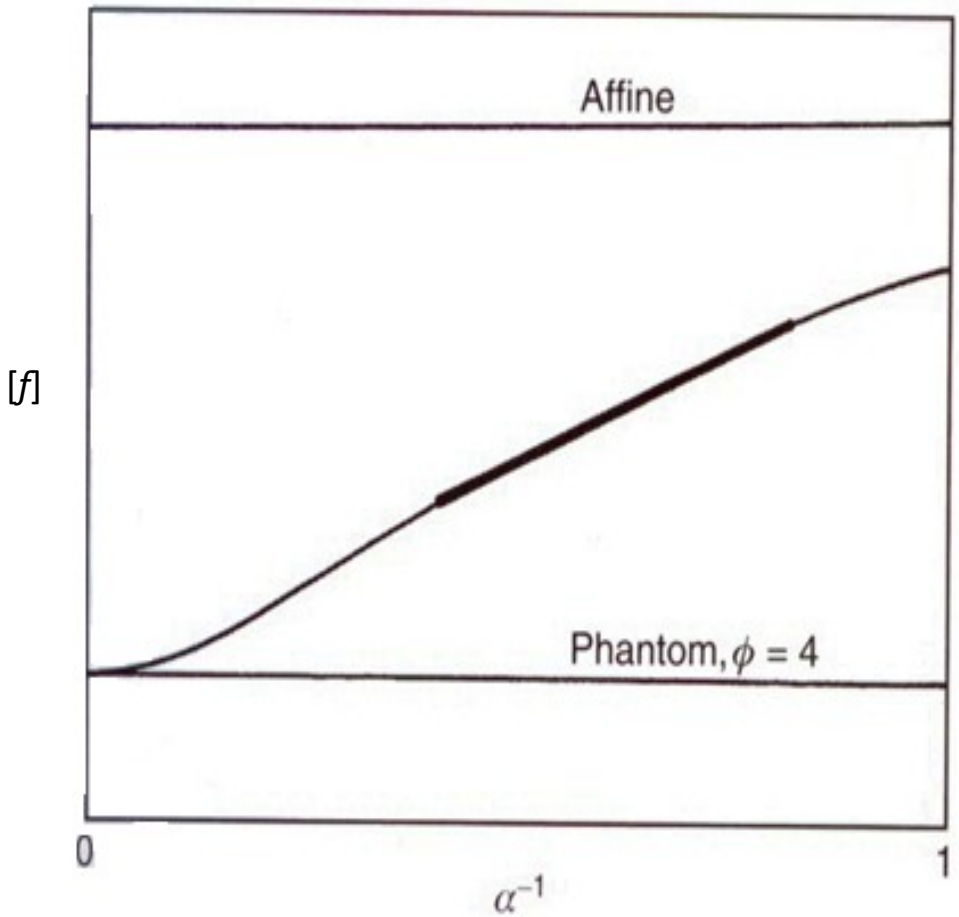

Fig. 3-3 Figure shows affine versus phantom model where the modulus is independent of the deformation. Taken from [12].

Where f^T is the nominal stress, C_1 and C_2 are empirical constants which are obtained by plotting experimental stress-strain data and are independent of the elongation .

Polymer chains in solution are free to rotate around individual bonds. There is a limit-less number of conformations or orientation in three-dimensional space for these chains. Based on the affine and phantom Gaussian rubber elasticity theories, shear modulus is described as

$$G = \rho_x \, RT \varphi_2^{1/3} \;\; \varphi_{2,r}^{2/3} \qquad \text{(Affine)} \qquad \text{[Eq. 3-9]}$$

$$G = (1 - \frac{2}{f}) \rho_x \, RT \varphi_2^{\frac{1}{3}} \;\; \varphi_{2,r}^{\frac{2}{3}} \qquad \text{(Phantom)} \qquad \text{[Eq. 3-10]}$$

Where R is universal gas constant and T is the absolute temperature. f is the crosslink functionality which is 4 for tetrafunctional polymers. According to these equations, shear modulus for an affine network is twice as large as the one for a corresponding phantom network with the same cross link density. This phenomenon has been interpreted as arising from deformation-dependent contributions from entanglements to the modulus and has been factored into more recent theories of rubber elasticity. Such dependence is particularly important for elongation experiments, but is relatively small in compression experiments involving small deformations.[9] One of the most important conclusions from these theories is the prediction that the modulus may increase due to presence of entanglements. The decreasing of the modulus with elongation can be interpreted as a result of relaxation of entanglement constraints due to the deformation.[10]

3.4 NEO-HOOKEAN ELASTIC SOLIDS

Rivlin in 1948 first discovered that stress at any point is linearly proportional to deformation and if the material is isotropic (that is, it has the same proportionality in all directions), then the extra stress due to deformation should be determined by a constant times the deformation due to the non linearity seen at higher strain. The Hookean relationship where this constant is actually the elastic modulus (E) holds only for low strain linear elastic regime where linearity is seen.[11]

$$\sigma = E\,\varepsilon \qquad \text{[Eq. 3-11]}$$

De-Long et al. did a study where they tested PLA-PEO-PLA hydrogels where no linear elastic region was seen even at low strains. Because of this nonlinear behavior, the modulus values obtained from fits using Hookean relationship were dependents on the strain range to which the data were fit. At lower strain, the slope of the fitted line was less than that of the higher strain and at higher strain the nonlinear downward sloping region led to inaccurate modulus values. They compared the modulus obtained from the Hookean model to that obtained from neo-Hookean model by converting the shear modulus to an elastic modulus by using the following equation:

$$E = 2G\,(1 + \nu) \qquad \text{[Eq. 3-12]}$$

where ν is the Poisson's ratio or the ratio of lateral tension to longitudinal compression. The Poisson's ratio for rubbers and most hydrogels and other incompressible material is reported as 0.5 which was used in this study to determine moduli.[12] Hence this equation turns to

$$E = 3G \qquad \text{[Eq. 3-13]}$$

Which is a better way of determining Young's modulus than the Hookean model which only applies to small strain regions and will be inappropriate to determine the Young's modulus on the nonlinear or higher strain region. Hence the neo-Hookean model allows for determining Young's modulus at non-linear region more accurately than the Hookean model. G can easily be calculated from plots of stress versus$(\lambda - \lambda^{-2})$ at low strains. At low strains, this yields a straight line, the slope of which is the modulus. $\lambda = {}^{1+\varepsilon}/_{100}$, where ε is the % strain calculated.

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Chapter 4

Synthesis and Characterization of Poly (N-vinyl formamide) and Polyacrylamide gels

The main motivation behind this work was to synthesize poly (N-vinyl formamide) PNVF macrogels and incorporate them within PNVF nanogels. It is hypothesized that this will improve fracture properties. PNVF nanogels have been synthesized before but a clear synthesis procedure of PNVF macrogels has not been established. Hence, one of the primary research goals of this study was to synthesize PNVF macrogels. The second main research goal was to fully characterize these gels and to compare properties of PNVF gels and the well known polyacryalmide gels which have been used in the industry for decades. Polyacryalmide gels have a wide range of applications as listed in section 2.3 but they have not been used in medical application due to their poor biocompatibility. NVF, on the other hand, is a less toxic monomer and has been claimed 'non-toxic' by certain literature including BASF.[1] In addition to lower toxicity, NVF is a liquid monomer which makes it easier to process and handle. It can also be hydrolyzed to polyvinylamine, which is a cationic polymer. It is closely related to poly (N-vinylpyrrolidone)(PNVP), which is extensively used in drug delivery applications as well as soft contact lenses and other biomedical devices.

NVF has several advantages over AAm and thus a wider range of applications due to its biocompatible nature. As these two monomers are hydrophilic isomers, if crosslinked similarly, they should have similar swelling and mechanical properties. To compare properties, these gels were fully characterized and the results are reported in this chapter. This chapter also briefs the reader on the materials, methods and equipment used to conduct this study. At the end of the chapter these results are discussed in detail and a direct comparison is made between the properties of PNVF and PAAm gels.

4.1 HYDROGEL SYNTHESIS

Poly (N-vinyl formamide) and polyacrylamide hydrogels were synthesized using the following materials and methods. Properties of hydrogels can be varied by varying monomer and crosslinker concentrations.

4.1.1 Materials

N-vinyl formamide (NVF) (Aldrich, 98% electrophoresis grade) was distilled under vacuum at 80°C and stored at –10°C prior to polymerization. 2,2'-Azobis (*N,N'*-dimethylene isobutyramidine) dihydro chloride (VA-044, Wako Pure Chemical Ind. Ltd.) was used as an initiator for the polymerization reaction of N-vinyl formamide without further purification. This initiator has a 10 hour half-life decomposition temperature, 44°C and is highly soluble in water. A non-degradable, water-soluble molecule, (N-vinylformamido) ethyl ether (NVEE), which is a derivative of N-vinyl formamide (NVF) was used as a crosslinker. This crosslinker was synthesized following the procedure reported in Appendix A.[2]

Acrylamide (Aldrich; 99 % electrophoresis grade) was used without any further purification. Ammonium persulfate (APS) and *N,N,N',N'*-tetramethylethylenediamine (TEMED) (Sigma, 98% electrophoresis grade) were purchased from Sigma Aldrich and were used to initiate polymerization reaction. *N,N'*-methylenebisacrylamide (bisacrylamide) (Sigma, 98% electrophoresis grade) was used as a crosslinker. All reagents were used as received if purification was not stated.

4.1.2 Poly (N-vinyl formamide) Gel Synthesis

NVF gels were made following T x C notations from gel electrophoresis literature where T is the ratio of total monomer (monomer + crosslinker) in solution and C is the mass of crosslinker

expressed as a percentage of the total amount of monomer plus crosslinker.[3,4] Thus, a 10x1 gel was made by dissolving 9.9g of purified PNVF monomer and 0.1g of NVEE in 100ml of deionized water. This monomer and crosslinker aqueous solution was purged at room temperature with nitrogen for 10 minutes to remove any oxygen. After purging the solution, 0.05wt% VA-44 in water was carefully added to the gel solution inside a glove box to keep any oxygen from dissolving.

It was a challenge to find a free radical initiator for this reaction. Ammonium persulfate, APS is the common initiator used frequently in combination with TEMED to synthesize PAAm chains. Hence, APS was used with NVF but it led to an orange side product[5] possibly due to formation of a conjugated structure, and that turned darker in color over time. Another free radical thermal initiator, azobisisobutyronitrile (AIBN), was also considered to synthesize PNVF gels. AIBN is partially soluble in water and requires around 80˚C for optimal performance. Such high temperatures were not ideal for the water bath setup in the lab and therefore, white crystalline powder Vazo-44 was used at about 50˚C.

The glove box was then sealed after gel solution and molds were placed inside. These silicone rubber molds were 25mm in length, 10mm in width and about 2mm in thickness as shown in Figure 4-1. A vacuum pump was left on for at least half an hour to make sure all the air was pumped out of the glove box. 200 microliters of this solution were then pipetted in 20 rectangular rubber molds under vacuum. These rubber molds filled with gel solution were carefully placed between glass sheets inside the glove box. Once these sheets were clamped shut, they were taken out of the glove box and heated in a water bath at 50°C ± 5°C for 24 hours.

Fig. 4-1 Clamped glass and rubber molds used in the lab. These molds were filled with gel solution and immersed in a water bath at 50°C for 24 hours.

Although all gel samples gelled within an hour, it was important to let the reaction proceed to completion under a fume hood as suggested by the Material Safety Data Sheets (MSDS) of N-vinyl formamide. This procedure was repeated for all different gel compositions being studied.

4.1.3 Polyvinylamine Gels

Polyvinylamine gels were synthesized by hydrolyzing PNVF gels as they cannot be polymerized directly from a monomer. These gels were synthesized following the same procedure mentioned above. After reaction reached completion, these gels were hydrolyzed by placing them in a 1M NaOH and then incubated at 80°C for 12 hours. At this point, the hydrolysis process was assumed complete and gels were tested to measure completion of the hydrolysis i.e. formamide conversion to amine. A similar procedure was followed by other research groups where 100% conversion was seen.[6] Polymers bearing primary amine functionalities are of great interest because of their high reactivity for various post-derivatization reactions as well as their cationic nature in appropriate pH regimes.[4] The hydrolysis provides a very favorable biomedical aspect because the polymer solutions are completely free of residual monomers after hydrolysis. PNVF can easily and quantitatively be hydrolyzed using basic conditions. Although acid hydrolysis of PNVF was not tested, it has been reported that PNVF exhibits limited conversions because of the electrostatic repulsion among the cationic amine groups generated during hydrolysis.[2]

4.1.4 Polyacrylamide Gel Synthesis

Polyacrylamide gels were made following the same T x C notations mentioned in Section 4.1.2. Thus, 10x1 polyacrylamide gel was made by dissolving 9.9g of AAm in 100 ml of deionized water. 0.1 g of bisacrylamide was added to the solution and was placed in a vacuum oven for 20

minutes to remove all dissolved oxygen. The freshly prepared APS solution (0.30 mg APS)/(ml monomer solution) and (0.15 mg TEMED)/(ml of monomer solution) was added to the 100ml solution. Rubber molds in glass sheets previously used were then filled with gel solution, sealed, and placed in a refrigerator at 4°C for 24 hours following the published procedure.[8] The acrylamide solution gelled within 5-10 minutes, but was left in the refrigerator for 24 hours to ensure completion of the reaction. This procedure was repeated for all of the different polyacrylamide gel compositions studied.

4.2 HYDROGEL CHARACTERIZATION METHOD

PNVF and PAAm gels were characterized by measuring swelling degree, Young's modulus, shear modulus, fracture stress and fracture strain. From this data, other network parameters like crosslink density (ρ_x), chi (χ), crosslinking efficiency and molecular weight between crosslinks (Mc) were also calculated.

4.2.1 Swelling Degree

The equilibrium swelling degree is the most important property of hydrogels: it directly influences the rate of water absorption, the permeability to drugs, and the mechanical strength of the gel. [9] After the reaction reached maximum extent, these hydrogels were placed in excess water for 24 hours to allow the water to leach unreacted materials and swell to equilibrium. Increasing the amount of crosslinker within the network increases crosslink density and thus the elastic forces, which reduces swelling in a given gel. The swelling ratio of all gel samples were calculated by dividing the mass of the swollen hydrogels known as the wet weight, by the mass of the dried hydrogels. Swelling degree was calculated by using the following equation:

$$Q = \text{Wet weight} / \text{Dry weight} \qquad \text{[Eq. 4-1]}$$

The mass of the swollen hydrogel was measured at the end of 24 hours by the gravimetric technique. These gels were blotted with blotting paper to remove any excess water which might alter the swelling ratio. Each gel sample was then weighed individually. It was important to keep track of changes in volume during the process till equilibrium swelling was reached. These swollen hydrogels were dried in a desiccator at room temperature for at least 24-48 hours.

4.2.2 Static Properties

TA Instruments RSA III was used to analyze all static mechanical properties of the test samples. The RSAIII used an advanced direct-drive linear motor to apply the strain, and a patented Force Rebalance Transducer™ to measure force. RSA III had two transducers which could bear loads up to 350N and 3500N. Low friction air bearings ensured optimal sensitivity. This device is particularly well-suited for testing soft materials, such as gels and elastomers. The tests were done in tension to determine fracture stress, fracture strain, toughness, Young's modulus (E) and shear modulus (G). To assure reproducibility of the readings, the distance of the grips was calibrated before testing each sample set.

In a typical static test, a polymer sample in the form of a dogbone is clamped at one end and pulled at a constant rate of elongation at the other clamped end. The rectangular gels were cut into a dogbone shape by using two sharp blades of equal dimensions shaped like a boat. These blades were attached to a wooden block which was pressed down on the rectangular gel sample to achieve the required dogbone shape as shown in Fig 4-2(a). The thinner portion of the static specimen encourages the sample to fail at the center of the bar, where the stress is the highest, and not at the grip sites where stress concentration may result in premature failure.[10]

This testing technique was applied to all gel samples which were precisely cut into a dogbone shape. They were held by geometries that pulled the gel apart at a constant rate of 0.5mm/sec from the center as shown in Figure 4-2(b). Three different rates, 0.25mm/sec, 0.5mm/sec and 0.6mm/sec were tested. Stress-strain curves came out the same under all these curves and therefore, 0.5mm/sec was used for all test runs. However, more recent studies carried out in the hydrogel group at KU after this thesis work was complete suggests that 0.05mm/sec yields lower moduli.

As mentioned above, gel samples were cut into a dog bone shape using two sharp bent blades. The width and height of these samples was measured by a vernier caliper and the length was measured by the instrument. Dimensions of all gel samples stayed constant throughout this study as same gel molds and dogbone blades were used for all samples. The narrow-stem part of the dogbone was about 15mm long, and it was clamped using the broader ends of the gel sample. In this experiment, it was ensured that the gels fractured from the center of the narrow part of the gel due to tension. Fracture of the gel sample from any part other than the narrow stem part of the gel was considered a failed run. Sample grips were moved in opposite directions until the gel fractured from the center. During deformation, force is measured as a function of elongation at the fixed ends by means of a force transducer. Static response was plotted as engineering stress versus engineering strain. Engineering stress is defined as the stress which is measured when load is applied to a specimen in tension, or as compression divided by the initial, unstressed cross-sectional area of the specimen. The engineering stress based on the initial cross-sectional area was calculated by TA RSA III, “Thermal Advantage” software after entering the dimensions of the gel sample. These dimensions were constant for all gel samples and are listed in Appendix B and C. Gel samples under study did not have any gaps or bubbles in them. Slipping, which

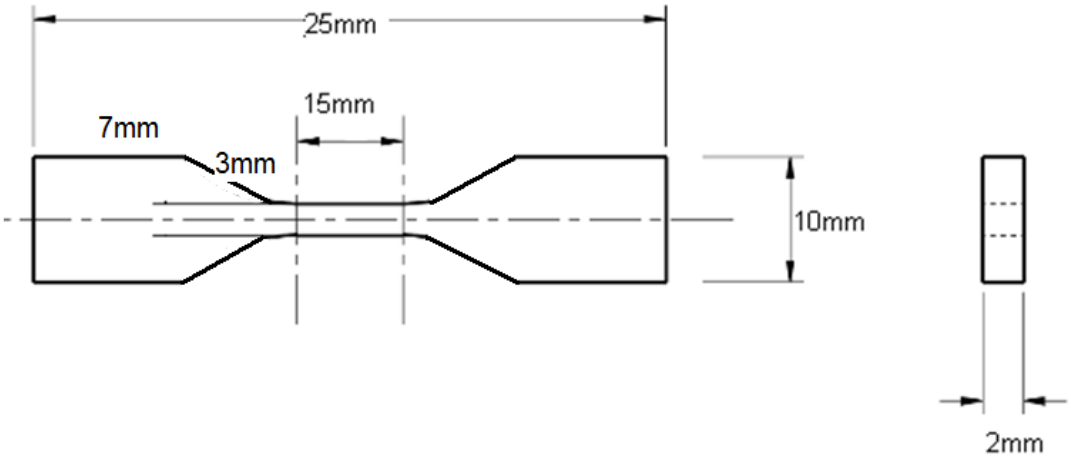

Fig. 4-2(a) Dogbone shaped gel samples for tensile testing.

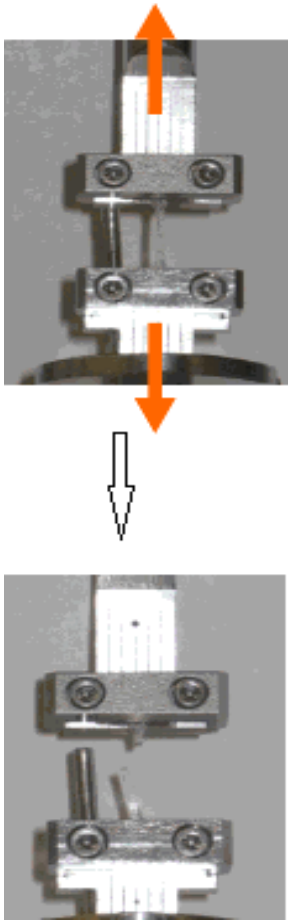

Fig. 4-2 (b) This figure shows the dog bone shape of the gels pulled apart in tension.

could visually be seen in the sample from the geometries, or fractures, due to prior deformation, or cracks in the gel, was also considered a failed run. Holding the gel sample too tight by the geometries resulted in failure at the broad part of the gel. Physical vibrations from any other lab instruments in use led to failure at an early point. Any kind of external pressure or noise was avoided during runs. Moreover, all experiments were performed at room temperature to further reduce any environmental contamination. All samples were kept hydrated while the data was being collected by immersing the gel samples in a metal well filled with water at room temperature.

4.3 RESULTS

This section includes all the experimental data collected on PNVF and PAAm single network gels. Various figures and tables have been used to illustrate the results and trends of PNVF gels and PAAm gels.

4.3.1 PNVF Gel Properties

As NVF is an isomer of AAM, to synthesize PNVF gels, conventional PAAm crosslinkers like bisacrylamide and EGDMA were used but they failed to adequately crosslink the PNVF polymer chains. Different concentrations of these crosslinkers were added to PNVF gel solution but it still did not produce a well-defined elastic gel. There were blobs or loosely crosslinked gels seen in the solution but a concrete gel failed to form. Low gel fraction was observed as compared to sol fraction because N-vinyl formamide and bisacrylamide copolymerizes following an alternating pattern rather than a random network structure resulting in polymer chains.[11] According to this hypothesis, all the crosslinker was used early in the reaction, leaving mostly homopolymer behind in the solution.

The *Q-e* scheme was used to further understand the reason behind the formation of polymer chains and goo instead of a well-defined gel. McCormick et al. calculated reactivity ratios of NVF and AAM to be 0.046 and 0.51 respectively.[11] *Q-e* values of NVF is not reported in the literature, however, NVF-AAM curve matches *Q-e* predictions for NVP-AAm seen on F_1-f_1 plot (Fig. 4-3). This similarity shows that NVP and NVF radicals with their secondary and tertiary amines attached to the propagating radicals respectively react in a similar way. Therefore, it can be concluded that NVP could be synthesized using the same procedure as NVF.

Furthermore, equations 2-3 and 2-4 were used to calculate reactivity ratios of NVP and EGDMA. r_{NVP} was calculated to be 0.0043 where r_{EGDMA} was calculated to be 3.95. As the amount of crosslinker present in the solution was limited compared to the monomer, the more available monomer reacted with the crosslinker earlier leaving mainly homopolymers in the solution. Using equation 2-5 and Q-e values of different monomers,[11] Figure 4-2 shows this trend.

In the case of NVF and AAm, Figure 4-2 shows that NVF and acrylamide with the ratio of 9-1 in the feed, copolymerizes to about only the ratio of 6-4. This figure also shows that the incorporation of the crosslinker in the network decreases towards the right to finally forming a homopolymer of the monomer. There is a difference in reactivity because there is nitrogen attached to the carbon radical as oppose to another carbon seen in acrylamide. This nitrogen next to the carbon radical is what NVF, NVEE and NVP all have in common. This is also clear by looking at the 'e' values of acrylamide versus NVP which is chemically similar in structure to NVF. Acrylamide has an electron withdrawing group which undergoes free-radical polymerization in the presence of a cation where as NVP has a negative e value. Due to difference in polarity of NVP and NVF, these monomers fail to react with each other.

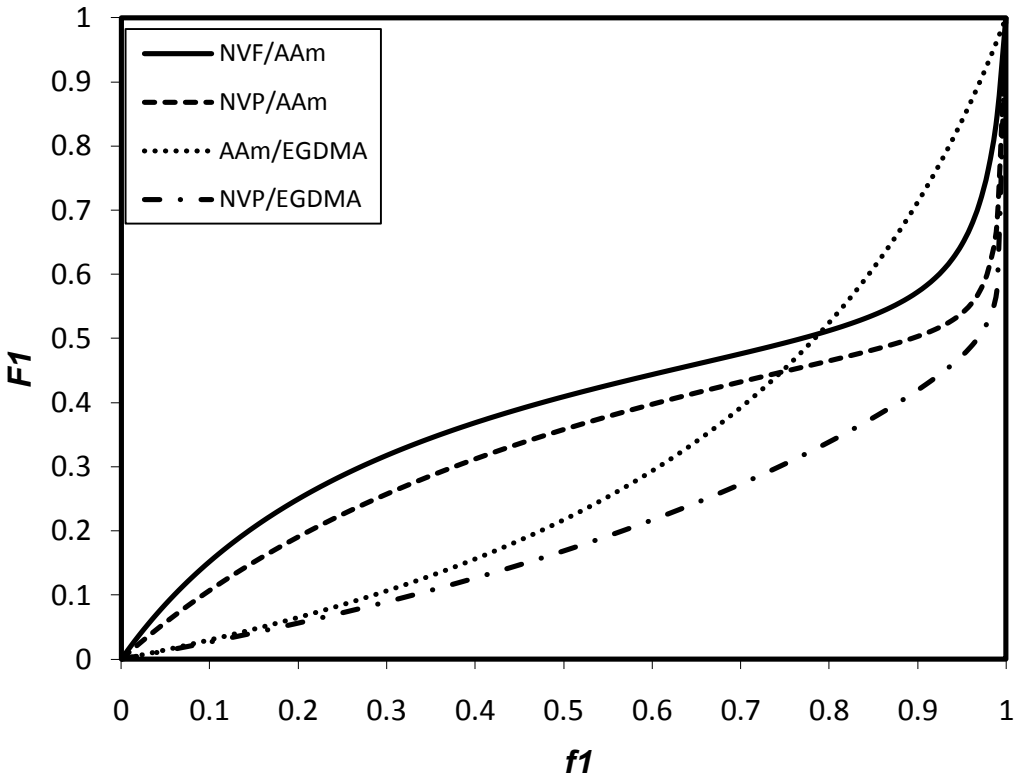

Fig. 4-3 This figure shows the trends of copolymerization between different monomers and crosslinkers where f_1 is the mole fraction of monomer and F_1 is the mole fraction of copolymer in the monomer mixture. The solid line is based on measured reactivity ratios, the others are predicted by the *Q-e* scheme. It can be seen that NVP-AAm is predicted to match NVF-AAm. Also, AAm/EGDMA is the best combination with the highest amount of monomer converting to polymer where as NVP/EGDMA seems to be the worst combination.

Thus, it can be concluded that it is important to investigate the polarity of monomer and crosslinker to establish whether they will react with each other in a network or not.

EGDMA is also a well known acrylamide crosslinker but this crosslinker also failed to crosslink PNVF chains together. EGDMA also has a positive 'e' and a carbon next to the radical which keeps it from reacting with NVF.

After establishing that EGDMA and bisacrylamide does not work well with NVF, NVEE was used as a crosslinker to achieve suitable copolymerizability with NVF. This gave the first significant success resulting in well-defined NVF hydrogels. Table 4-1 shows the % conversion of monomer into the network. These hydrogels were prepared by free radical copolymerization with various monomer concentrations of NVF and NVEE in deionized water. All gel samples were tested using RSA III and gel properties were calculated and reported. Percent monomer was calculated by using the equation 4-2.

$$\%\ monomer = \frac{dry\ \ wt}{mold\ \ wt * [\frac{T+C}{T+C+water}]} \qquad \text{[Eq. 4-2]}$$

Table 4-1 Table shows the percent conversion of the monomer with 1mass% NVEE into the network. This was calculated for a range of gel types to estimate the quality of the gel using molecular weight of NVF.

$MW_{NVF} = 71.08 g/mol$

Gel type	% Monomer Conversion
10x1	86
15x1	88
20x1	89
25x1	93

Figures 4-4 and 4-5 are static experiments that show the stress-strain response to increasing NVF concentration with fixed amount of crosslinker. All gel samples were made using either 0.02 g or 0.06g of crosslinker in 20 ml solution. In both Figures 4-4 and 4-5, the increase in monomer concentration increased the fracture stress when crosslinker was kept constant. Elasticity of gels decreased as the crosslinking efficiency increased with the increasing amount of monomer, though 10x1 and 10x3 did not follow this trend. This could be because 10% is the lowest composition to synthesize a gel and fracture properties might be misleading as the network may not be as well-developed as at higher monomer concentrations.

In addition to varying monomer concentration, increasing crosslinker concentration also increased fracture stress and fracture strain as seen in Fig. 4-4 and 4-5. All samples had a clear point of failure where there was a sharp drop in stress. Fracture stress, fracture strain, Young's

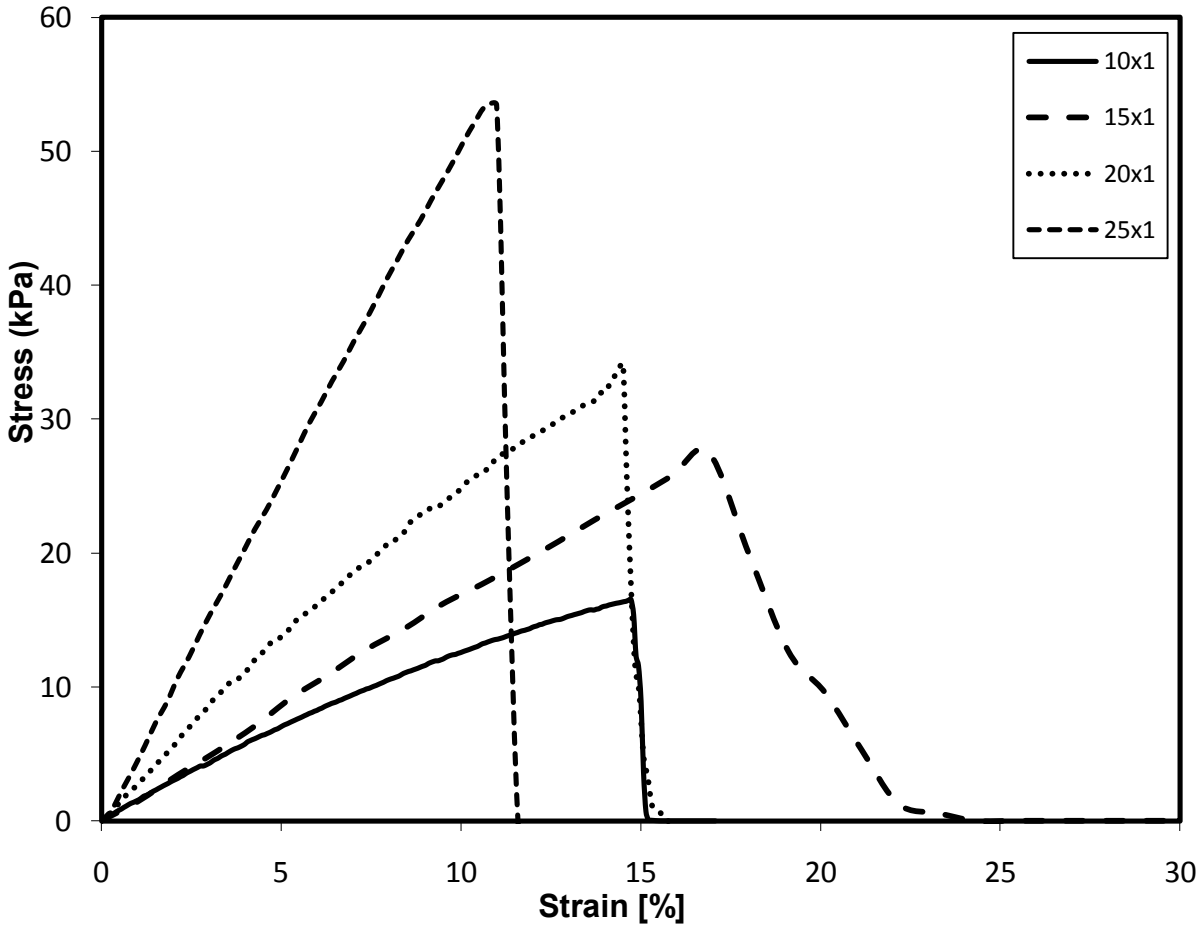

Fig. 4-4 Static stress-strain response of poly (N-vinyl formamide) gels swollen to equilibrium in water as a function of monomer concentration for 1% crosslinking ratio at a strain rate of 0.5 mm/sec. Young's modulus (E) and fracture stress increased as monomer concentration was increased, while fracture strain decreased at monomer concentration above 10%.

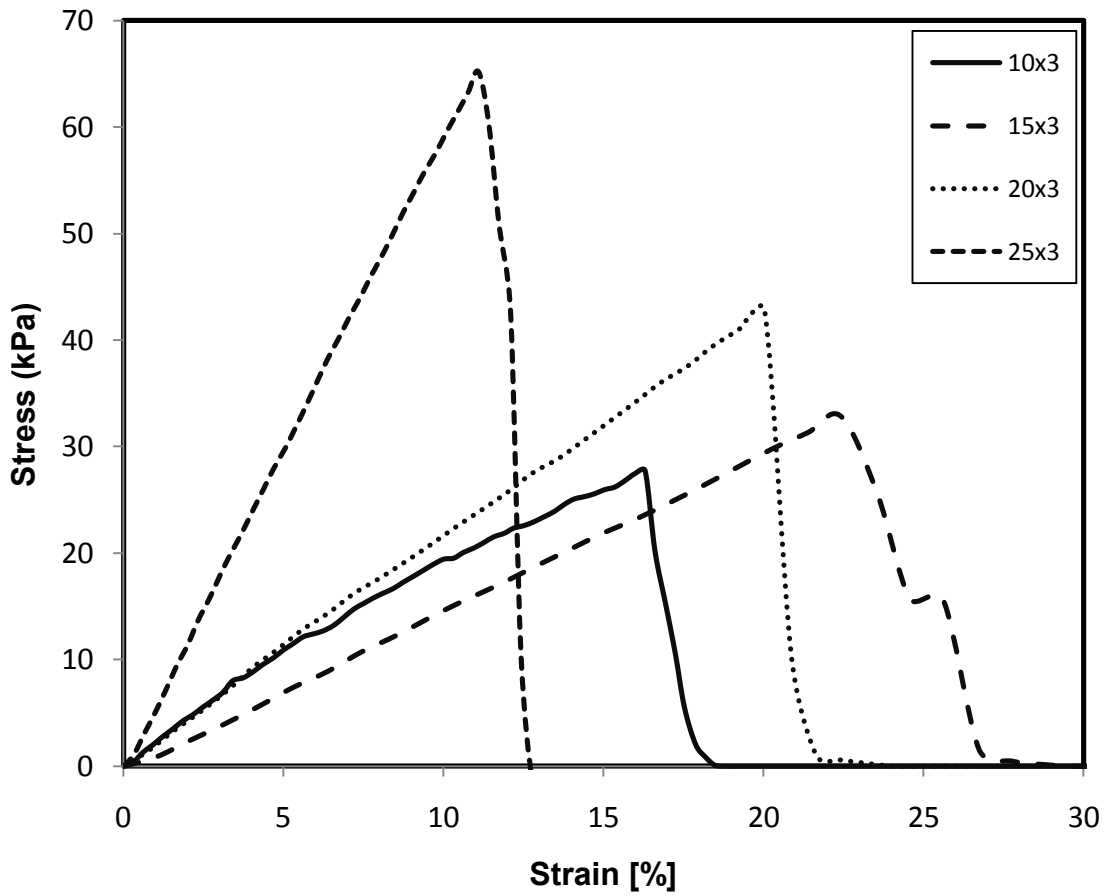

Fig. 4-5 Static stress-strain response of poly (N-vinyl formamide) gels swollen to equilibrium in water as a function of monomer concentration for 3% crosslinking ratio at a strain rate of 0.5 mm/sec. Young's modulus (E) and fracture stress increased as monomer concentration was increased, while fracture strain decreased at monomer concentration above 10%.

modulus, shear modulus and toughness were calculated and tabulated in Table 4-2. Other network parameters such as crosslink density and χ parameters were also calculated from theory as well as measured quantities and reported in Table 4-3 (a) and (b).

Figure 4-6 is an example of shear modulus calculation. Shear modulus (G) was calculated from plots of stress versus strain function$(\lambda - \lambda^{-2})$ at low strains. At strains up to 50-70%, this yields a straight line of whose slope is the modulus. $\lambda = {}^{1+\varepsilon}/_{100}$, where ε is the % strain calculated. R^2 value indicated how well the line fit the curve.

Figure 4-7 shows the trend of decrease in swelling with the increase in monomer concentration. It also represents and compares swelling with higher monomer concentration (filled circles). Swelling decreased with the increasing monomer concentration when the crosslinker was kept constant. Increasing the crosslinker concentration decreased the swelling ratio. These results followed the same general trend seen in the literature which is discussed in detail in the discussion section.

Figure 4-8 shows the trends of gel moduli with varying monomer and crosslinker concentration. Young's modulus and shear modulus is expected to increase with the increase in monomer and crosslinker concentration. Increasing the monomer increased the PNVF gels moduli but increasing the crosslinker failed to increase the modulus for every gel formulation.

The shear modulus and Young's modulus of PNVF gels were calculated and tabulated in Table 4-2. Figure 4-9 shows the trend of crosslink density with increasing monomer/crosslinker concentration. Increasing monomer and crosslinker is expected to increase crosslinking within a polymer network. The 10x1 and 10x3 formulations seem to be outliers in this case showing

unexpectedly high crosslink density. As expected crosslink density increased with increasing monomer concentration which is consistent with published literature.

4.3.2 PAAm Gel Properties

Polyacrylamide gels were synthesized using bisacrylamide their percent conversion is listed Table 4-5. tested following the same techniques used for PNVF gels. Three different gel compositions were synthesized with varying monomer and crosslinker concentration. In all cases fracture stress increased with the increase in monomer concentration. Increasing crosslinker also increased fracture stress of PAAm gels. These results were later compared with those of PNVF gels to determine the similarity of these two gel types.

Figure 4-10 is a static experiment showing the stress-strain response of increasing acrylamide concentration with varying crosslinker concentration. Fracture stress, fracture strain, Young's modulus, shear modulus and toughness were calculated and tabulated in Table 4-6. All these gel samples were made at room temperature. Other network parameters such as crosslink density and χ parameters were also calculated and reported in Table 4-7 (a) and (b).

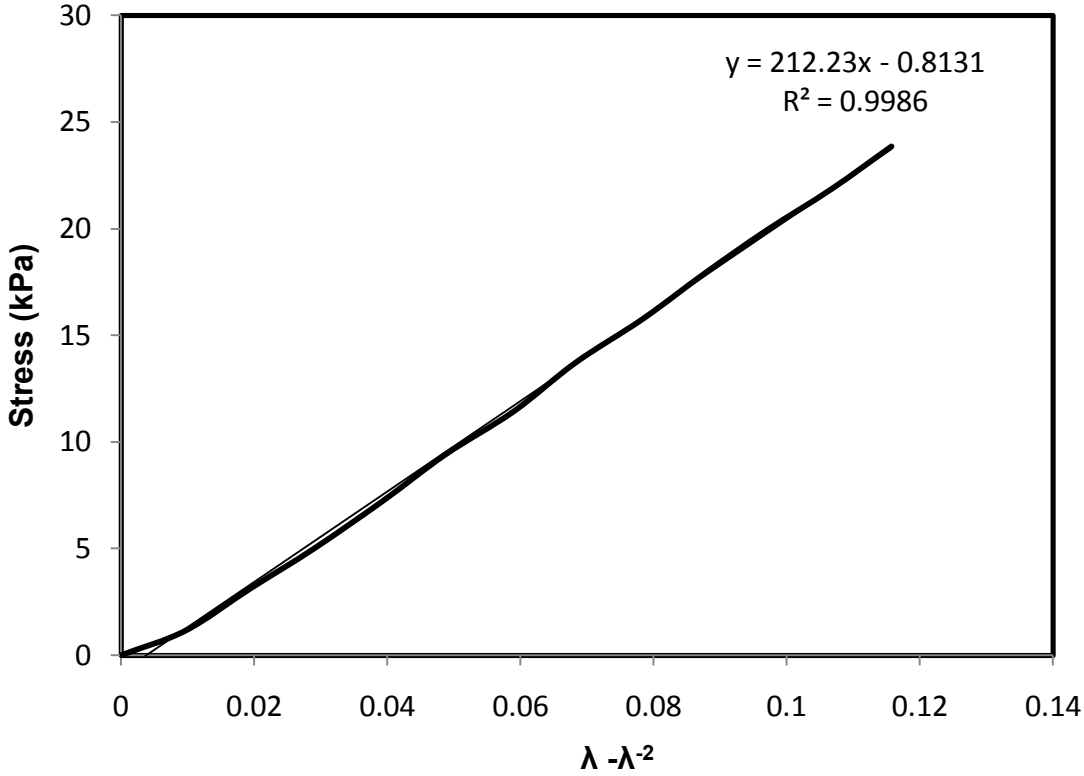

Fig. 4-6 An example of the sample calculation of shear modulus (G) of 25x3 PNVF gel from stress-strain data in Figure 4-4. A straight line is obtrained by plotting stress versus strain function shown where $\lambda = L/L_o$ as predicted by theory for rubbers. The shear modulus is found from the slope to be 212.2kPa.

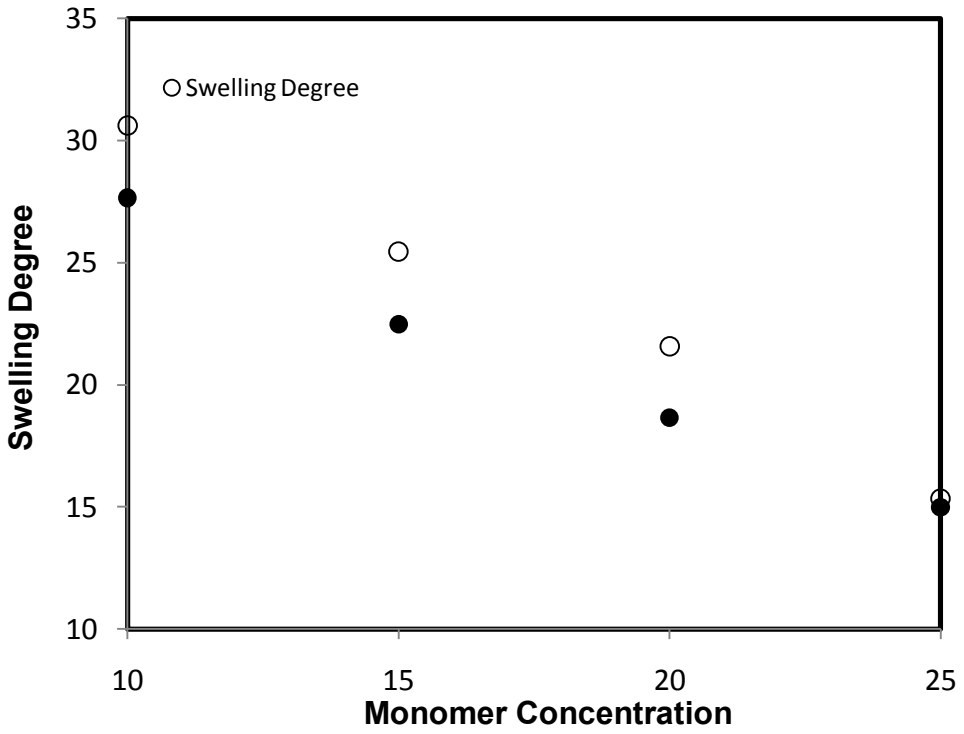

Fig. 4-7 Swelling degree of PNVF gels in water at room temperature for different compositions. Swelling ratio increases with the decrease in monomer and crosslinker concentration, consistent with expectations for gels. ○, 1% crosslinker; ●, 3% crosslinker concentration.

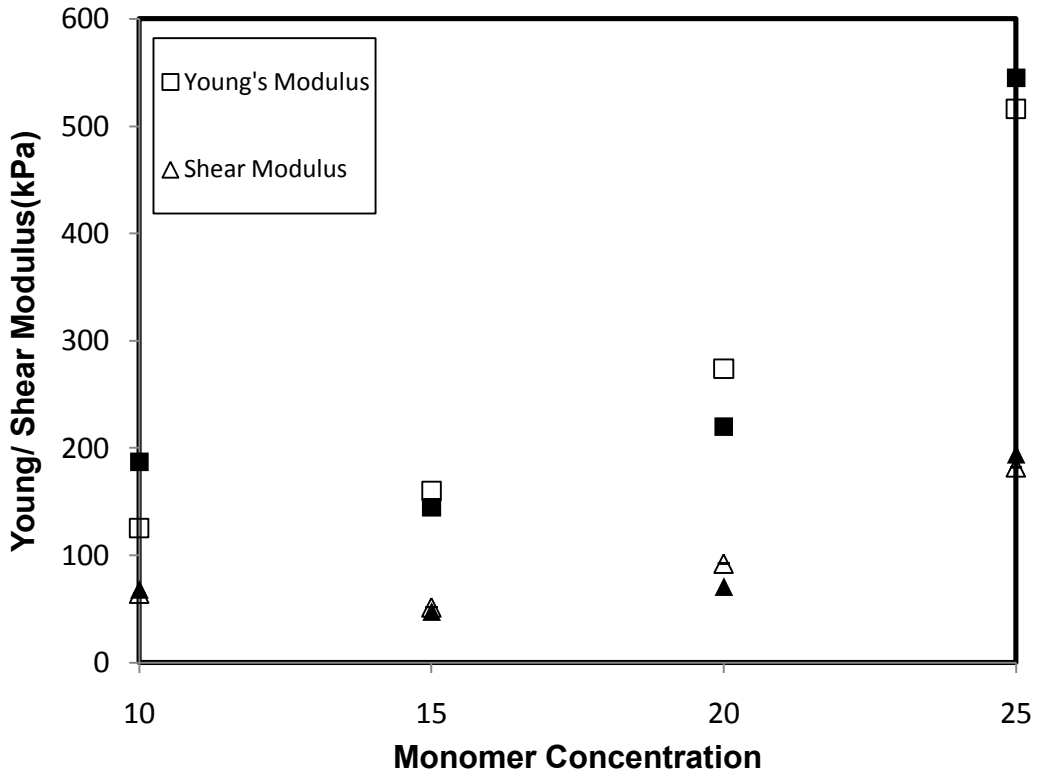

Fig. 4-8 Trend of moduli of PNVF gels with 1% and 3% crosslinker added. Increasing the monomer concentration increases the moduli while decreasing the swelling ratio. □, Young's Modulus with 1% crosslinker; ■, Young's Modulus with 3% crosslinker; Δ, Shear Modulus with 1% crosslinker; ▲, Shear Modulus with 3% crosslinker.

Table 4-2 Effect of varying monomer and crosslinker concentration on mechanical properties of PNVF gels swollen to equilibrium in water at 25°C. Standard deviations were calculated using excel STDEV function using 3 gel samples (n=3) from the same batch. Underlying data tables are tabulated in Appendix B.

Gel type	Swelling Degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]	E/G	Toughness AUC (J/m^3)
10x1	30.6±0.2	138.1±3.1	48.9±9.1	16.5±0.2	14.8±0.7	2.81±0.5	142.2±4.3
10x3	27.7±0.4	220.1±1.1	77.3±3.1	26.8±0.2	15.6±0.2	2.92±0.1	273.5±5.4
15x1	25.5±0.6	172.4±1.5	60.7±2.1	27.6±0.4	16.8±0.1	2.93±0.1	305.6±4.6
15x3	22.7±0.4	142.5±2.1	49.7±4.1	32.8±0.1	22.4±1	2.92±0.2	445.4±5.2
20x1	21.6±0.1	282.6±1.5	98.8±4.2	29.6±0.1	15.0±0.5	2.91±0.1	310.2±3.9
20x3	18.7±0.5	232.8±1.7	81.7±3.3	42.9±0.1	20.0±0.7	2.92±0.1	460.8±3.4
25x1	15.1±1.1	520.6±3.2	182.3±5.2	53.5±0.1	10.9±0.2	2.92±0.1	320.3±4.2
25x3	15.0±0.2	609.8±2.3	212.2±2.1	65.5±0.1	11.1±0.4	2.80±0.02	424.4±4.1

Table 4-3(a) Network parameters of PNVF gels of different monomer/crosslinker concentrations calculated from data in Table 4.2 using the affine model, which is the most widely used rubber elasticity theory.

Gel Type	Swelling Degree Q (g/g)	Crosslink density ρ_x (mol/cm^3)	Solubility parameter χ	M_c (g/mol)	Crosslinking efficiency (%)
10 x 1	30.6	2.90E-04	0.25	4.13E+03	31.1
10 x 3	27.7	4.44E-04	0.19	2.70E+03	47.5
15 x 1	25.5	2.59E-04	0.28	4.63E+03	18.4
15 x 3	22.7	2.00E-04	0.37	5.99E+03	14.5
20 x 1	21.6	3.29E-04	0.24	3.64E+03	17.6
20 x 3	18.7	2.59E-04	0.35	4.63E+03	14.1
25 x 1	15.1	4.66E-04	0.27	2.57E+03	19.9
25 x 3	15.0	3.57E-04	0.33	3.01E+03	21.23

Table 4-3(b) Physical properties of PNVF gels of different monomer/crosslinker concentrations calculated from data in Table 4.2 using the phantom model, which is expected to work best for highly swollen gels.

Gel Type	Swelling degree Q (g/g)	Crosslink density ρ_x (mol/cm^3)	Solubility parameter χ	M_c (g/mol)	Crosslinking efficiency (%)
10 x 1	30.6	1.45E-04	0.38	2.07E+03	15.5
10 x 3	27.7	2.22E-04	0.41	1.35E+03	23.8
15 x 1	25.5	1.30E-04	0.40	2.32E+03	9.2
15 x 3	22.7	1.00E-04	0.42	2.99E+03	7.2
20 x 1	21.6	1.65E-04	0.38	1.82E+03	8.8
20 x 3	18.7	1.30E-04	0.39	2.31E+03	6.9
25 x 1	15.1	2.33E-04	0.39	1.29E+03	9.9
25 x 3	15.0	2.45E-04	0.38	1.57E+03	11.2

Table 4-4 Basic hydrolysis of PNVF to Polyvinylamine (PVAm).

Gel Type	Swelling Degree (Q) (g/g)	Fracture Stress (kPa)	Fracture Strain [%]	Toughness (AUC) (J/m^3)
10x3	16.8±0.5	21.6±2.1	15.8±1.1	196.9±3.2
15x3	26.4±0.7	22.6±2.4	24.2±1.4	335.2±3.7
20x3	24.7±1.2	35.9±3.1	25.5±2.3	586.3±2.8
25x3	15.9±0.3	51.1±2.4	9.4±2.1	271.2±3.5

Table 4-5 This table shows the percent conversion of the acrylamide monomer into the network when bisacrylamide was used as a crosslinker. This was calculated for each gel type to estimate the quality of the gel using molecular weight of acrylamide.

$MW_{AAm} = 71.08 g/mol$

Gel Type	% Monomer Conversion
10x1	65.2
20x1	69.9
20x3	72.3

Table 4-6 Table summarizing results from polyacrylamide gels made with different monomer/crosslinker concentration.

Gel Type	Swelling degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]	E/G	Toughness AUC (J/m^3)
10x1	28.6±0.1	149.6±0.5	52.5±2.1	20.9±1.3	13.8±0.5	2.9±0.1	159.9±2.3
20x1	20.4±0.4	199.2±2.3	69.2±0.5	35.2±1.9	15.2±1.1	2.9±0.4	360.2±2.5
20x3	18.9±0.3	258.6±5.1	89.5±3.2	42.4±1.1	22.2±0.9	2.8±0.1	617.9±3.1

Table 4-7(a) Network parameters of polyacrylamide gels of different monomer/crosslinker concentrations calculated from data in Table 4.6 using the affine model. This model is most commonly used in the literature.

Gel Type	Solubility parameter χ	Swelling Degree Q (g/g)	Crosslink density ρ_x (mol/cm^3)	M_c (g/mol)	Crosslinking efficiency (%)
10 x 1	0.39	28.6	3.05E-04	3.94E+03	23.7
20 x 1	0.43	20.4	2.26E-04	5.30E+03	8.8
20 x 3	0.42	18.9	2.85E-04	4.20E+03	11.1

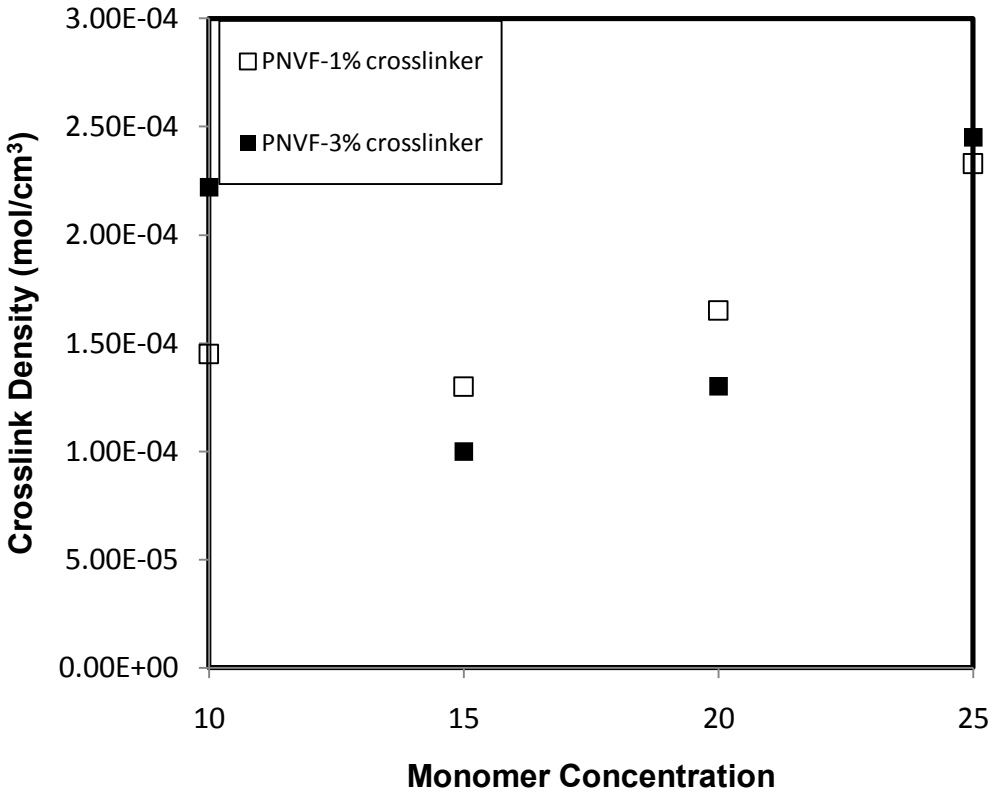

Fig. 4-9 Figure shows the effective cross-link density of PNVF gels with varying monomer/crosslink concentration. Effective crosslink density increases with the increasing monomer concentration above 20% concentration. □, 1% crosslinker;, ■, 3% crosslinker concentration.

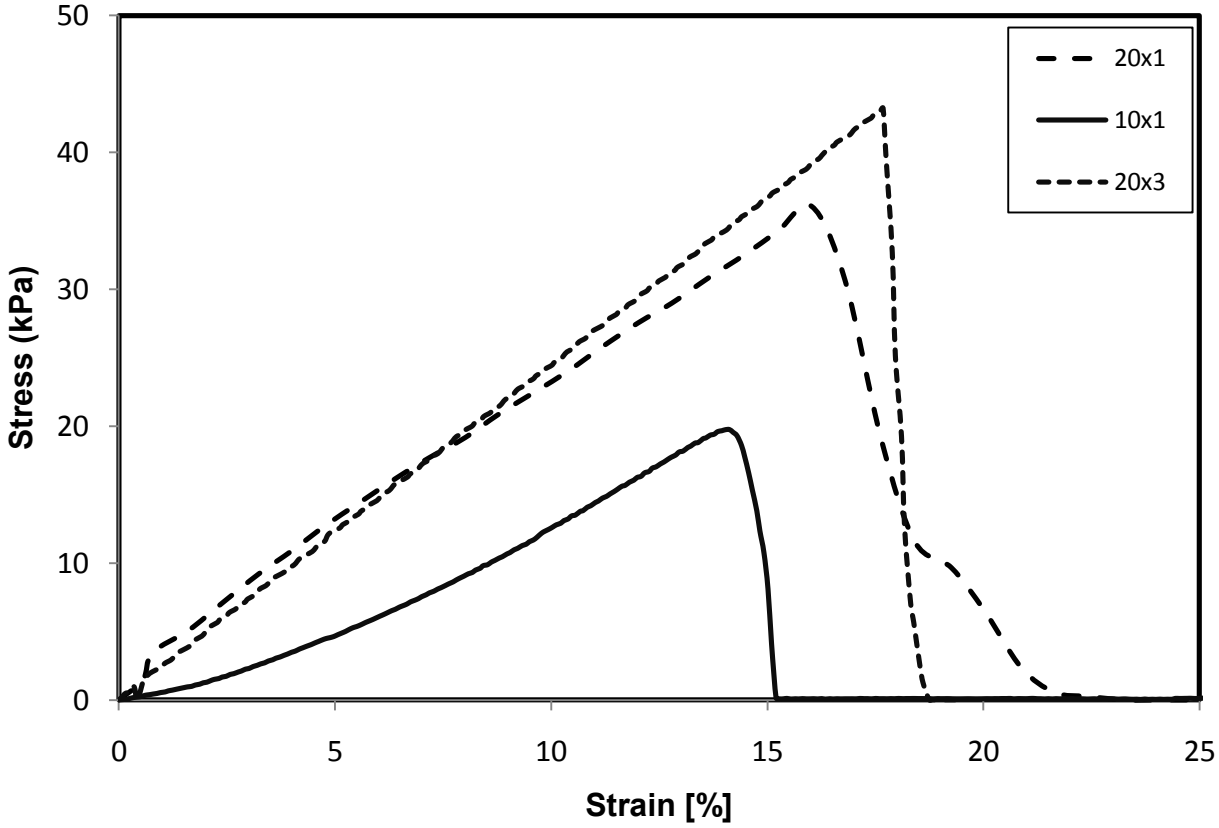

Fig. 4-10 Static stress-strain response of polyacrylamide gels swollen to equilibrium in water as a function of monomer concentration for 1% and 3% crosslinking ratio at 0.5mm/sec. Increasing crosslinker concentration increased fracture properties resulting in a better quality gel. **Standard deviations did not show on the plot due to small values.*

4.3.3 PNVF versus PAAm Gel Properties

Figures 4-11 (a, b and c) show direct comparison between the swelling and moduli of PNVF and PAAm gels. As seen in Figure 4-11(a), PNVF gels swell more than PAAm gels. Higher swelling of PNVF is observed because it is somewhat more hydrophilic than PAAm. Figure 4-11 (b and c) compares the moduli of the two gel types. Moduli of both gel types increased with increasing monomer/crosslinker concentration. PNVF have higher Young's modulus and shear modulus at 20x1 formulations when compared to PAAm gels. Figure 4-11(a, b and c) represents bar graph showing the minor differences in the moduli and swelling of PNVF versus PAAm gels. The results of these plots are tabulated in Table 4-9.

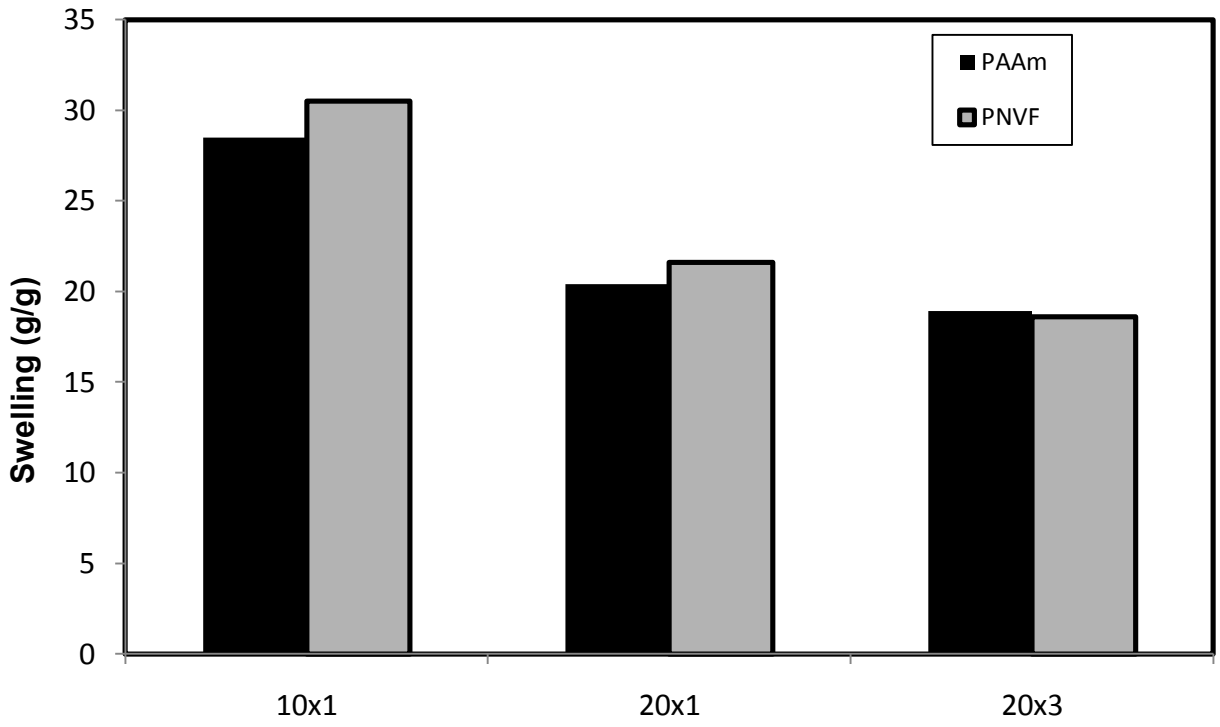

Fig. 4-11 (a) Bar graph showing the swelling ratios of PAAm versus PNVF gels of various monomer/crosslinker concentration. It can be seen from the bar graph that there is less than 5% difference between the swelling ratios of PNVF and PAAm gels at a certain gel formulation. **Standard deviations did not show on the plot due to small values.*

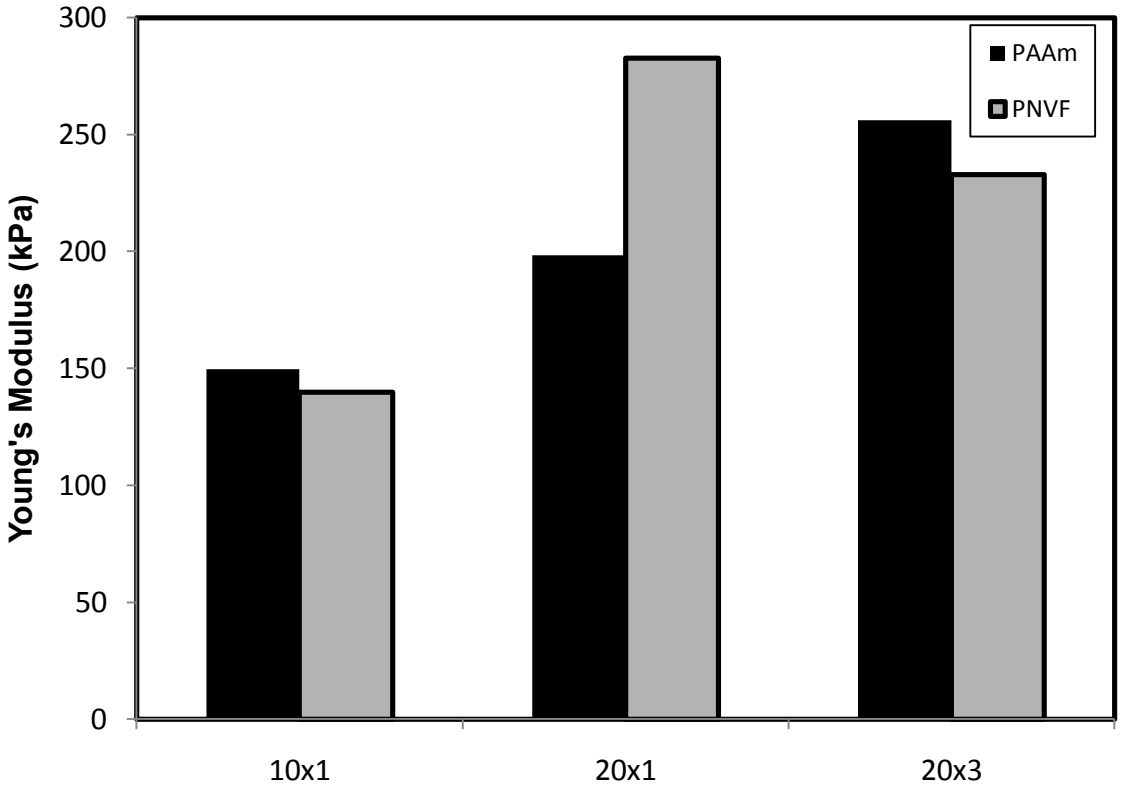

Fig. 4-11 (b) Bar graph showing the difference between the Young's modulus of PAAm versus PNVF gels. As expected, increasing the monomer/crosslinker concentration increases the moduli of the gel.

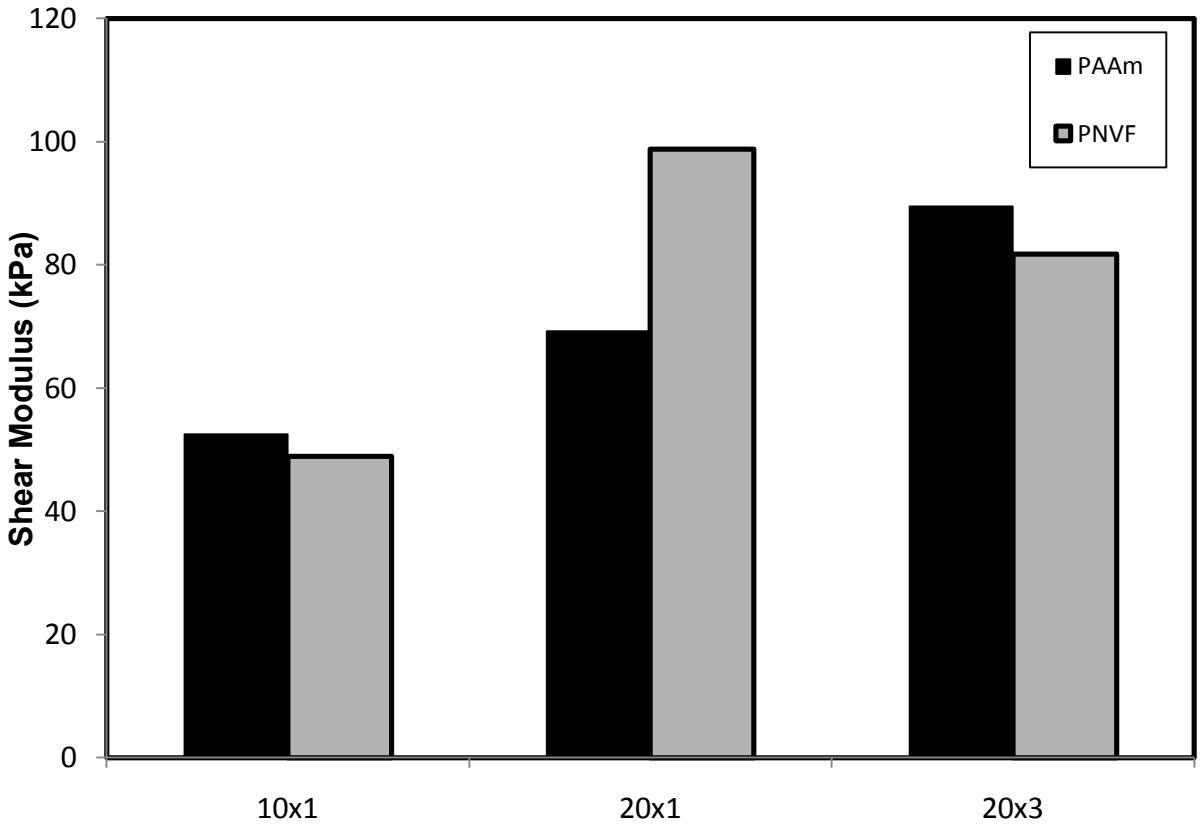

Fig. 4-11(c) Bar graph summarized the difference between shear modulus of PAAm versus PNVF gels. The modulus of both gels increased with increasing monomer/crosslinker concentration.

Table 4-7(b) Additional physical properties of polyacrylamide gels of different monomer/cross-linker concentrations calculated from data in Table 4.6 using the phantom model.

Gel Type	Solubility parameter χ	Swelling Degree Q (g/g)	Crosslink density ρ_x (mol/cm^3)	M_c (g/mol)	Crosslinking efficiency (%)
10 x 1	0.39	28.59	1.52E-04	1.97E+03	11.87
20 x 1	0.43	20.42	1.13E-04	2.65E+03	4.38
20 x 3	0.42	18.96	1.43E-04	2.10E+03	5.59

4.4 DISCUSSION

One of the research goals of this study was to synthesize PNVF gels which could later be combined with PNVF nanogels to improve mechanical properties. Successfully synthesizing PNVF gels was one of the major accomplishments as very limited data is available on this gel type in the literature. As this gel has not been extensively used before, it was essential to fully characterize and evaluate its properties. N-vinyl formamide is an isomer of acrylamide and thus it was hypothesized that their gels might have similar characteristics if crosslinked similarly. This section focuses on interpretation of the results and is largely limited to discussing the gel properties of both PNVF and PAAm hydrogels. It directly compares the mechanical properties such as fracture stress, fracture strain, Young's modulus, shear modulus and toughness of these two gel types and draws conclusions regarding their similarities and differences. It also discusses the two main models of rubber elasticity, affine and phantom, which were used to calculate

swelling parameters like χ parameters, crosslink density and average molecular weight between crosslinks (M_c). It was determined that phantom model gave more convincing results when compared to affine model as phantom model is used for highly swollen gels. A direct correlation between varying monomer/crosslinker concentration and gel properties was seen in this study which is similar to trends seen in the literature.[13]

4.4.1 Poly (N-vinyl formamide) Gel Behavior

The choice of a class of hydrogel for use in a given application strongly depends on its properties. The composition, that is amount of T and C added, may also help predict its properties. This measured quantity was used to calculate monomer reaction efficiency and the crosslinker reaction efficiency which gave an idea of how much each had been incorporated in the copolymer network. The properties of these gels, however, may differ significantly if the reaction does not go to completion, or if the monomer or crosslinker does not incorporate in the network in a random manner. There are three main extremes for incorporation of different monomers into a network formation. The first extreme is the formation of a block copolymer, where one monomer tends to react with its own kind much more strongly than with another kind of monomer or crosslinker. This happens when the reactivity ratio 'r' is greater than 1. This leads to long polymer chains which are weakly crosslinked with each other and tend to not become a part of the network. They form weak 'blob' like gels with poor quality, especially when the co-monomer is the crosslinker. As the crosslinker fails to effectively react with the monomer, this leads to low gel moduli and high swelling ratios. This particular structure will lead to low effective crosslink density; in other words, low efficiency of the crosslinker in the network structure will lead to a poor quality gel profile. Once the shear modulus was calculated using stress-strain curves, crosslink density was easily calculated using equations 3-9 or 3-10,

which shows shear modulus, is directly proportional to crosslink density. As the number of crosslinks increases in the network structure, shear modulus should also increase.

The second basic extreme for the incorporation in a network formation is the alternating pattern. In this pattern, an alternating copolymer forms when a monomeric unit has a strong preference to add a monomer of the opposite type. In hydrogel networks, the second monomer is the crosslinker, which is present in a much smaller fraction of the monomer's amount. Therefore, it will be consumed first leaving unreacted monomer, or will simply form a homopolymer which will remain in the solution. On the other hand, if monomer and crosslinker have equal reactivity towards each other, random or 'ideal' copolymers form, leading to a structure where the spacing of monomers and crosslinkers in chains is correlated with their relative abundance in the solution. This ideal network structure will have 100% conversion or incorporation of the monomer in the structure and will have a random incorporation of the crosslinker holding the polymer chains together. This can be further explained by looking at an example of a 10x1 formulation. In an ideal case, it is expected that this formulation will lead to a structure which will have a crosslink unit present after every 9 monomer units. This should give an ideal effective crosslink density where all the crosslinker will be consumed in the structure. The percent monomer conversion shows how much of the monomer ended up in the network. Table 4-2 shows that most (85-93%) PNVF became a part of the final network. Throughout this study, crosslink density was calculated from swollen gels. Reaction conditions which lead to high crosslink density and high monomer incorporation in the network leads to higher moduli. As mentioned before, crosslink density was calculated from the modulus using either affine or phantom model (equations 3-9 and 3-10). These two models of rubber elasticity have been discussed in chapter 3. The affine model works best at low extensions where chains and

crosslinks are closer together and deformations are translated to all chains as opposed to the phantom model, where it is assumed that polymer chains have no constraints in movement. This works best with hydrogels which are highly swollen with crosslinks relatively farther apart. Both models neglect dangling ends of the chain and assume Gaussian distribution of end-to-end chain length. The difference in the two model comes down to a factor of 2 for tetrafunctional crosslinkers, i.e $\rho_{x,affine} = (1/2)\ \rho_{x,phantom}$.[9]

Equilibrium swelling degree of a nonionic gel depends on a balance between swelling forces and elastic forces, which are characterized by two main parameters, the crosslink density and the solubility parameter (χ). Swelling degree (Q) increases as the average number of repeat units between the crosslinks increase. Higher number of crosslinks or crosslink density tends to decrease the number of end-to-end conformations a polymer chain can take, thus restricting overall swelling of the network. Swelling degree also depends on polymer-solvent interactions or solubility parameter. This polymer-solvent interaction parameter accounts for free energy changes caused by mixing polymer and solvent and is generally between 0 and 1. χ values are usually constant for a given polymer because it does not depend on synthesis conditions but the polymer itself. The solubility is enhanced when polymer-solvent interactions are highly favorable. This allows more water to enter the network resulting in chain expansion and increasing the swelling ratio. According to the theoretical basis that has been developed for the interaction parameters, $\chi < 0.5$ translates into polymer-solvent interactions dominate while $\chi > 0.5$ means polymer-polymer interactions dominate, discouraging dissolution. These values are significantly dependent on concentration and temperature. χ values were not measured but directly calculated using Equation 3-2 as they are related to the other parameters.

In addition to swelling parameters and shear modulus, Young's modulus of hydrogels was also studied under tension. A linear fit to the stress-strain curve was used to calculate the materials Young's modulus. These measurements were restricted to regions of low strain where stress-strain curves were accurately linear. For an elastic hydrogel, the ratio of E/G should equal 3.0. For finite strain the value of E/G should be slightly < 3 in extension and slightly > 3 when using compression.[14] Another trend that can be expected is that Young's modulus increase with the increase in crosslink density and monomer concentration. An increase in monomer concentration leads to higher reaction efficiency and better incorporation of crosslinker into elastically effective junctions and also increases the number of physical entanglements. Fracture strain, on the other hand, is observed to decrease with increased crosslink density. As the number of crosslinks increase in a network, the length of each polymer between crosslinks decreases, reducing the overall fracture strain of the structure. However, there is not a good theory for correlating fracture properties to crosslink density.[15] These stress-strain curves can also be used to calculate material toughness. Toughness is defined as the area under a stress-strain curve which equals to the work of deformation. This corresponds to the energy absorbed by the material prior to failure. Material toughness as well as other network parameters allows researchers to better understand hydrogel properties which in turn enhance the use of these gels.

In addition to calculating mechanical properties of these gels, they were also hydrolyzed to poly-vinylamine (PVAm) in 1M NaOH solution. PVAm is a cationic polymer with high charge density. Table 4-5 shows the results of these tests. These tests were carried out in the hydrolysis solution (1M, NaOH) as opposed to water. This diminishes any ionic effects, and therefore, an increase in swelling or significant changes in other mechanical properties was not

observed. This does, however, show the stability of the crosslinked structure under hydrolysis and suggests that PVAm gels can be made by this method.

4.4.2 Polyacrylamide Gel Properties

Polyacrylamide gels were prepared by free radical polymerization in aqueous solution. In general, the concentration of monomer in the reaction solution affects the properties of the resulting gel. Addition of crosslinker to the hydrogel system changes the swelling value of the hydrogel as the molecules of crosslinkers are placed between the chains of the monomers of polyacrylamide restricting their movement. Swelling degree calculated from this study was compared against literature values. A study done by Ruvalcaba et al. looked at the effect of pH and gelatin concentration on swelling of polyacrylamide gels. It can be seen that with 0% gelatin concentration in polyacrylamide gel solution, the gels swelled to 23.2 g/g.[12] The concentration of crosslinker bisacrylamide was not mentioned but the swelling range is comparable to the calculated swelling degree of polyacrylamide gels from this study. In another study, Young's modulus of PAAm gels in tension using the stress strain curves was calculated 21kPa where fracture stress was 120kPa using 10x1 gel formulation.[12] M_c of PAAm gels was reported $1.46x10^3$ at 10% crosslinker on mass basis which is similar to the range calculated from this study. [13] This study concluded that having crosslinker values greater than 5% does not have a significant effect on mechanical properties of PAAm gels.[14] Zugic et al. conducted a similar study where they looked at mechanical properties of poly(N-isopropyl acrylamide) (PNIPA). Gel formulation of 10x1crosslink density of $0.85x10^4 mol/cm^3$, however, calculated efficiency of crosslinking was much lower than calculated from this study. They also reported swelling degree of 12.2 which was lower than calculated from this study.[16]

Change in the swelling alters the mechanical properties of the hydrogel. Various monomer to crosslinker concentrations were considered in these sets of experiments to study the effect of monomer/ crosslinker concentration on the gel solution. Although PAAm gels have been studied before, it was important to conduct these experiments to compare PNVF gel properties against PAAm gel properties under the same environmental and technique-dependent conditions. It is evident from the results displayed in Table 4-7(a) that polyacrylamide gels are affected by changes in monomer and crosslinker concentration. Large amount of crosslinks in the network structure increases crosslink density so that the elastic forces outweigh the osmotic forces of polymer-solvent mixing sooner than for less-crosslinked gels. Similar trends of N-substituted acrylamide gels are seen in the literature and one such example is presented in Table 4-8.[7] Gehrke et al. conducted a study where they studied the effect of synthesis conditions on properties of poly (N-isopropylacrylamide) gels.[7] Similar to the objective of this study, Table 4-8 shows the trends of increasing monomer/crosslinker concentration and its effects on crosslink density. They observed that increasing monomer/crosslinker concentration increased the moduli as well as the effective crosslink density. They also established that, for a more concentrated monomer solution, most of the network is formed after the gel point. This study showed similar trends in PAAm and PNVF. At the same time, it was made sure that all reactions completed by leaving gel solution to react for at least 24 hours.

Table 4-8 Properties of Poly (N-isopropylacrylamide) gels. Increasing monomer concentration increases moduli. Adapted from [7].

Gel Type	Shear Modulus (kPa)	Equilibrium degree of swelling	Crosslinking density (mol/cm^3)	Crosslinking efficiency (%)
10x1	3.23±5	19.6	3.6×10^{-5}	23
16x1	8.53±6	14.8	6.5×10^{-5}	39
10x4	12.1±7	11.1	1.1×10^{-4}	18
16x4	21.5±5	7.8	1.3×10^{-4}	20

Table 4-7(a) also gives the values of the polymer-water interaction parameter. $\chi < 0.5$ translates into polymer-solvent interactions dominate while $\chi > 0.5$ means polymer-polymer interactions dominate, discouraging dissolution. $\chi = 0.5$ is a special case where the polymer-solvent interaction is cancels out.[10] These values are significantly dependent on concentration and temperature. The χ value for polyacrylamide has been reported in the literature in the range of 0.499-0.53.[15] In a similar study, Xue et al. reported χ value of poly (N-ethylacryalmide)(PEA) of 0.487 and 0.5 when crosslinker concentration was increased.[16] Figure 4-12 compares the χ values of PAAm gels and PNVF gels. Although, calculated χ values are lower than expected, this figure shows that they stay consistent throughout as they are not dependent on the network. It can also be seen that χ values of PNVF are slightly smaller than PAAm gels, indicating they are more soluble in water as compared to PAAm.

χ values calculated by Xue et al. are higher than those calculated in this polyacrylamide gel study which stayed between 0.39-0.42. The χ values using the phantom model were more

convincing than those calculated using affine model because the phantom model assumes that polymer chains are permitted to pass through one another as if they had zero cross-sectional area.

In addition to polymer-solvent parameters, effective crosslink density was also calculated and tabulated in Table 4-7(a). The effect on crosslink density did not increase as significantly as expected with the increase in crosslinker concentration. It has been pointed out by Mark[13] that gel formation in solution is often characterized by low crosslinking efficiency due to the formation of a large number of elastically ineffective dangling ends in the network.[15]

Fracture properties of polyacrylamide gels have not been studied in the past as extensively as the moduli of the gels. It is not possible to make meaningful comparison of these fracture properties with literature values because such values are scant for polyacrylamide hydrogels. There is not a good theory for fracture properties.[15] It is more common to list the moduli of the gel than to measure the failure point. It was important to study the fracture properties of PAAm gels with varying monomer/crosslinker concentration to allow comparison with PNVF gels. Fracture stress and fracture strain increased with the increase in monomer/crosslinker concentration as crosslinking efficiency and crosslink density increased as seen in Table 4-9.

Increasing the crosslinker concentration increased the number of crosslinks in the network resulting in an increase in crosslinking density. Increasing monomer/crosslinker concentration also increased the number of polymer chain entanglements within a network allowing the network to bear more load giving a higher modulus.

4.4.3 Comparison of polyacrylamide and poly (N-vinyl formamide) gels

Poly (N-vinyl formamide) gels were synthesized using a custom made crosslinker, NVEE. Although NVF is an isomer of acrylamide, it does not copolymerize with bisacrylamide. APS solution resulted in degradation and side products. It was a major challenge to find an optimal crosslinker and initiator for PNVF gels. NVEE is an ester whereas bisacrylamide is almost a near dimer of acrylamide and thus these two crosslinkers have different radical reactivity as discussed in Section 4.3.1 and seen in Figure 4-13. Although both crosslinkers are water-soluble and have vinyl groups which are the site of polymer propagation, the polarity of propagating radicals of each is different.

PNVF and NVEE system resulted in well-defined macrogels which were then fully characterized. Swelling parameters and mechanical properties calculated for this gel led to an important discovery that acrylamide and N-vinyl formamide were not only structurally similar but possessed similar properties in a swollen network. Table 4-9 (a, b) compares some of the main properties of three different formulations of these two gel types.

Swelling degrees of both types of gel were calculated following the same procedure. Table 4-9(a) shows that both these gels swell to the same amount at a given monomer/crosslinker concentration, overall swelling was higher compared to the literature value of similar polymers. This higher swelling resulted from low percentage of monomer conversion into the network leading to low crosslinking efficiency.

Table 4-9(a) Comparison between PNVF and PAAm gels. n=3 for all tests.

	Gel Type	Swelling Degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]
PAAm	10x1	28.6±0.1	149.6±0.5	52.5±2.1	20.9±1.3	13.8±0.5
PVNF	10x1	30.6±0.2	138.1±3.1	48.9±9.1	16.5±0.2	14.8±0.7
PAAm	20x1	20.4±0.4	199.2±2.3	69.2±0.5	35.2±1.9	15.2±1.1
PVNF	20x1	21.6±0.1	282.6±1.5	98.8±4.2	29.6±0.1	15.0±0.5
PAAm	20x3	18.9±0.3	258.6±5.1	89.5±3.2	42.4±1.1	22.2±0.9
PVNF	20x3	18.7±0.5	232.8±1.7	81.7±3.3	42.9±0.1	20.0±0.7

Table 4-9(b) Comparison of PNVF and PAAm network parameters calculated using the phantom model.

	Gel Type	Solubility parameter χ	Crosslink density ρ_x (mol/cm^3)
PAAm	10x1	0.39	1.52E-04
PVNF	10x1	0.38	1.45E-04
PAAm	20x1	0.43	1.13E-04
PVNF	20x1	0.38	1.65E-04
PAAm	20x3	0.42	1.43E-04
PVNF	20x3	0.39	1.30E-04

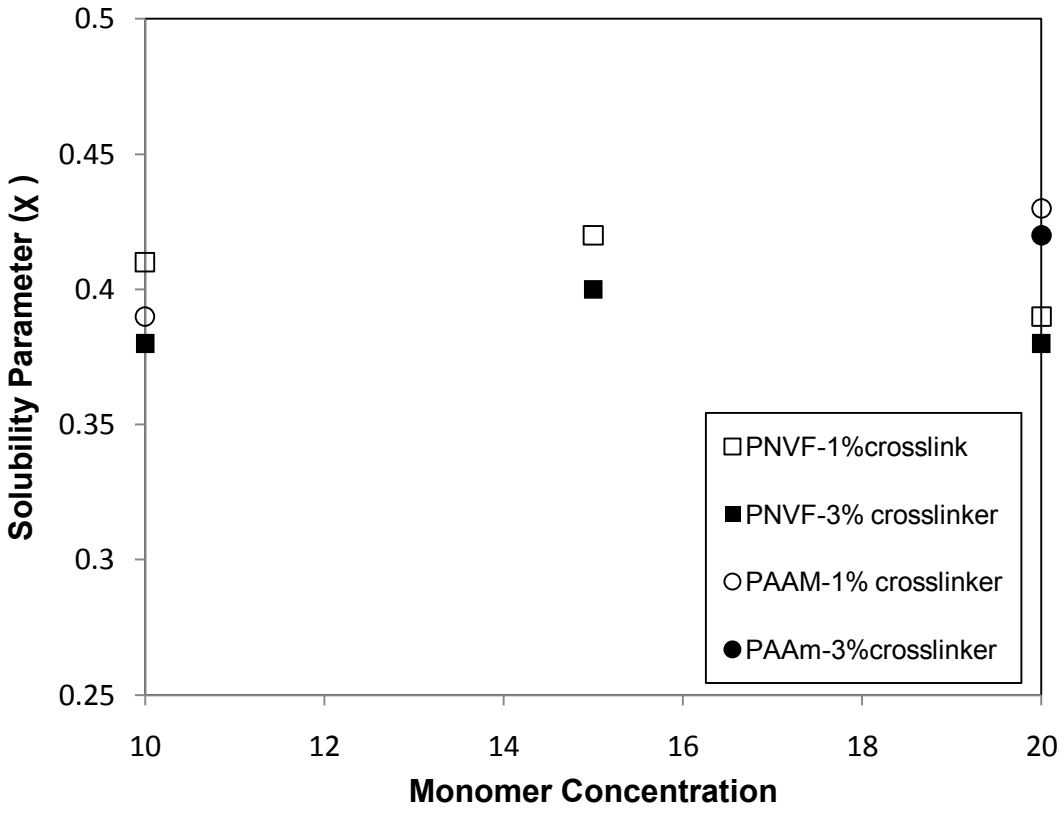

Fig. 4-12 Solubility parameter χ of PNVF and PAAm gels. χ should stay consistent for a given monomer. Similar χ values for PNVF and PAAm gels show that these two polymers have similar solvent solubility.

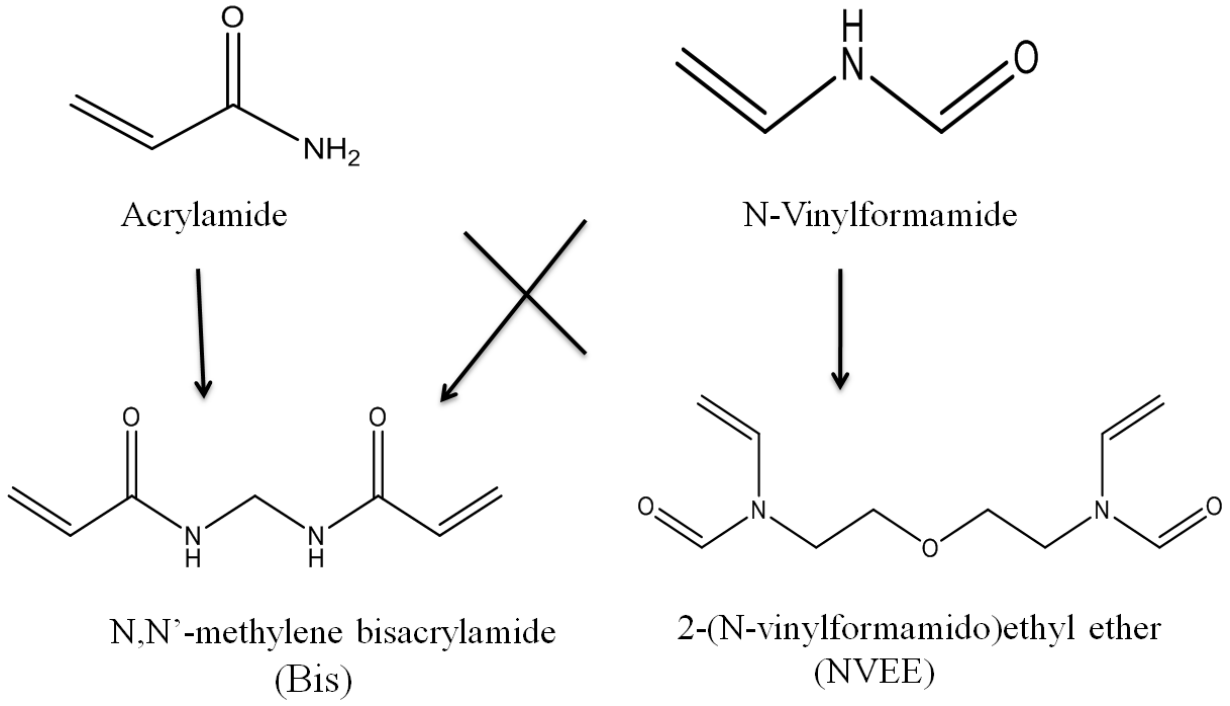

Fig. 4-13 This figure shows how NVF can crosslink with NVEE due to the presence of a nitrogen next to the propagating radical instead a carbon present in Bis.

PNVF gels swelled more than PAAm gels because they are more hydrophilic in nature. This can be seen from lower χ values of PNVF gels. This could also be seen from swelling degree of these two gel networks in water as χ influences Q ($Q = f(\rho_x, \chi)$). χ comes from polymer-solvent interaction and is not dependent on the crosslink density whereas crosslink density was measured independently from shear modulus G as ($G= f(\rho_x, Q)$) using equation 3-4. Similar ρ_x suggests that both these polymers form similar network structures, which is why their fracture properties are comparable as seen in Figure 4-6(a). Fracture stress increased with the increase in monomer/crosslinker concentration resulting in a gradual increase in the modulus of the gel sample. In addition to fracture stress, fracture strain in both cases increased with the increasing monomer concentration making the gel more ductile.

Based on these comprehensive results as well as statistical tests done on these two groups, listed in Appendix D, it is safe to say that the mean differences of PNVF gels and PAAm gels are significant at 0.05 level. Therefore, PNVF gels should be able to fulfill the same, if not a wider range of applications as it is a liquid monomer with lower toxicity than acrylamide.

4.5 CONCLUSIONS

Synthesis and characterization of PNVF gels was extensively studied by experimental methods such as swelling tests and mechanical testing. These results were compared to polyacrylamide gels because they are widely used in biomedical and pharmaceutical applications. To synthesize PNVF gels, a non-degradable water soluble crosslinker NVEE was used. In addition, a thermal initiator VAZO-44 was used at 50˚C to initiate free radical polymerization.

To characterize these gels, swelling and mechanical tests were conducted. Using a crosslinker concentration of 1 mass %, the swelling degree of the PNVF gels decreased from

30.6 g/g to 15.1g/g as the monomer concentration increased from 10% to 25% due to increased crosslinking efficiency. In addition to swelling, mechanical parameters such as fracture strain, fracture stress, Young's modulus and shear modulus were calculated. There is not a good theory for fracture stress and strain; therefore, the values were compared to fracture properties of acrylamide gels. Figure 4-9(a) shows that acrylamide gels can withstand higher stress than PNVF gels, which fracture at a smaller stress. Although fracture stresses were different, fracture strain of these two gel types were comparable. Similar fracture properties show that these gels have similar network structure. Shear modulus was calculated in the gels. It was found to be approximately two times higher than later studies done in our lab. The reason for this could be because the strain rate used for this study was 0.5mm/sec whereas later studies used 0.05mm/sec for sample testing. However, tests for this study were done in tension whereas later experiments used compression to calculate mechanical properties. This suggests the need for further work to resolve the influence of strain rate.

In addition to mechanical properties, other parameters, such as solubility of the polymer and crosslink density were calculated using an established theory as well as measured values. Solubility parameter (χ) of PNVF and PAAm gels calculated from this study stayed between 0.39-0.42 where as χ values listed for PAAm gels in the literature is 0.51. Although, χ values were lower than expected, they stayed constant through-out the study, showing that they are not dependent on the network. Crosslink density calculated from rubber elasticity theory for PAAm and PNVF gels were also comparable as seen in Fig. 4-9(b). As these parameters were calculated from the theory and not directly measured, it can be concluded that these two network types are the same.

Future work on PNVF gels is necessary due to certain discrepancies in the data. Primarily, χ values calculated from this study are different from what were found in later studies done in our lab. There should be more experiments done to examine reasons for lower χ values. Lower χ values were a result of measured shear modulus which was different by a factor of 2 when compared to later studies. This difference could be dependent on the strain rate.

From this study, it was concluded that PNVF gels can copolymerize with NVEE but not with bisacrylamide or EGDMA. Characterization of these gels show that PNVF gels behaves like a typical hydrogel and should be able to replace PAAm gels in certain applications.

Chapter 4- References

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Chapter 5

Hybrid Gels of Macrogels and Nanogels

Chapter 4 mainly focused on PNVF single network gels whereas this research chapter focuses on the significance and characterization of hybrid PNVF gels. These gels were synthesized using various PNVF nanogels concentrations which were incorporated in the network. It was hypothesized that incorporating nanogels in the network would improve mechanical properties of PNVF single network by increasing fracture strain.

5.1 REINFORCING FILLERS AND OTHER ADDITIVES

Reinforcing fillers are used to improve mechanical properties, such as modulus, static or tear strength, abrasion resistance, and fatigue strength.[1] Fillers are usually inert materials which improve processability or serve to reduce resin cost. Resin is a nonvolatile substance that is prepared by polymerization of simple molecules; i.e., the polymer. Although the disadvantage to adding fillers is that it generally increases hysteresis which is a lag in response in reacting to changes in the force. The most important example of fillers is the addition of carbon black to natural rubber and to some synthetic elastomers.[2] There are several other fillers like ellipsoidal fillers for glassy polymers, clay-like fillers and porous fillers such as zeolites[3] which threads the chains through the cavities with usually close interactions between the reinforcing phase and the host elastomeric matrix, are some examples of fillers.

However, these rubber materials are not the swollen gels which are the focus of this study. The simplest way of obtaining elastomers reinforcement in macrogels is by polymerizing the reaction in the presence of nanogels.

5.1.1 Increasing Mechanical Properties of Hydrogels

Synthetic hydrogels are seldom used in mechanical devices because of their lack of mechanical strength and volatility of the solvent. Therefore, it is essential to improve mechanical properties of these gels to widen their use.[2] This could be done by adding nanogels in the network which would work as non-covalent crosslinker. Recently, three new hydrogels with good mechanical properties have been successfully developed: a 'topological gel' using figure-of-eight crosslinkers, a 'nanocomposite gel' using clay as crosslinker, and a 'double network gel' by interpenetrating polymer network methods.[3]

One such example is the type of gels with non-covalent crosslinks known as the slide-ring gel. The slide-ring gel has mobile crosslinks which can slide along the backbone of the polymer chains.[4] Figure 5-1 shows an illustration where double or triple rings are able to slide along polymer chains.[4] These mobile crosslinks are thought to increase extensibility, degree of swelling and reversible deformability.

Similar to the concept of slide-ring gels, 'chain mail' or 'Olympic loop' theory was proposed by de Gennes in the 1970's. He explained that by introducing cyclic structures to linear chains in the polymeric network should permanently trap one or more network chains by threading through them.[1] These loops will be used as crosslinkers in the network by holding the chains together by non-covalent bonds as seen in Figure 5-2. In this figure, loops B, C and D are holding multiple polymer chains together whereas loop A is not playing any role in crosslinking. Loops B, C and D will allow polymer chains to move through them to their maximum length without breaking or fracturing the network by redistributing applied stress.

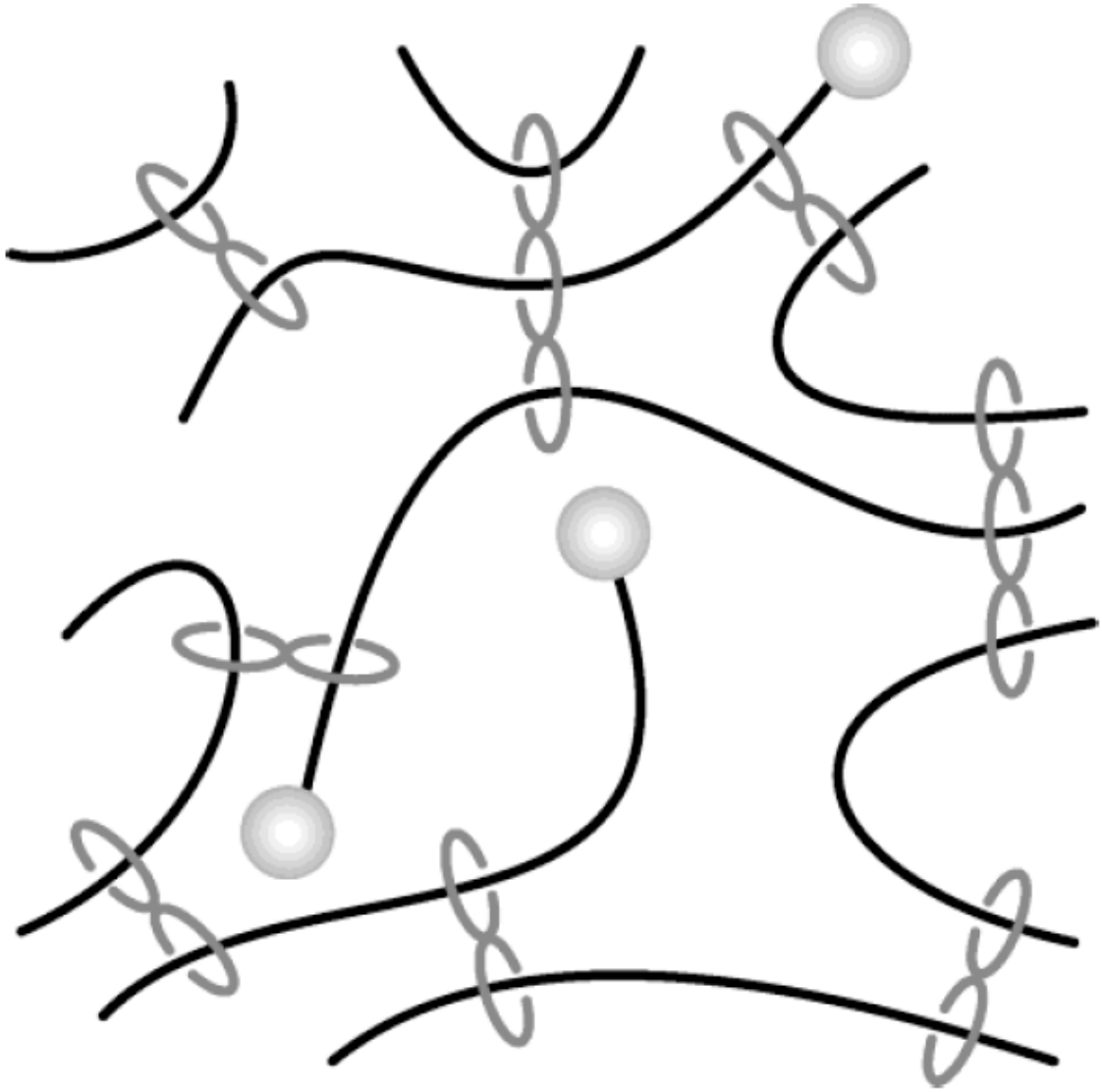

Fig. 5-1 Illustration of slide-ring gel. Polymer chains are crosslinked with double and/or triple ring crosslinkers, allowing them to redistribute applied stress. Taken from [4].

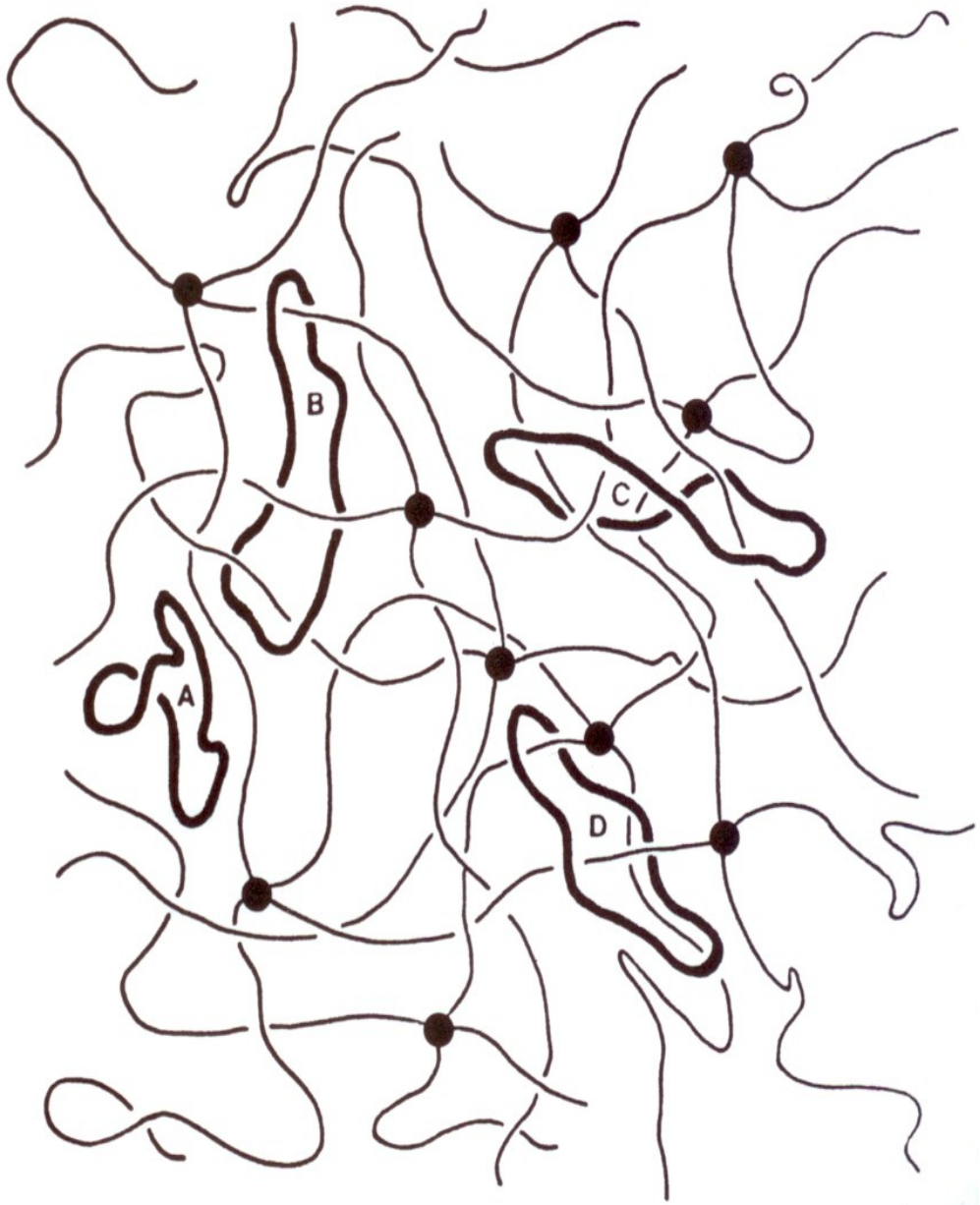

Fig. 5-2 Network chains are permanently trapped by cyclic B, C and D. “Olympic loops” are holding the chains together while giving them space to move back and forth to their maximum length as oppose to the cross-links which ties the polymer chains together at a fixed point. Taken from [1].

In this study, nanogels were incorporated into the structure similar to slide- rings and Olympic loops. It was hypothesized that nanogels will act as non-covalent crosslinker which will hold multiple polymer chains together. As these are not covalent crosslinkers, they will allow polymer chains to redistribute stress as they move back and forth through the nanogels. It is hypothesized that this phenomenon should increase elasticity of the network by increasing fracture strain.

5.2 EXPERIMENTAL WORK

PNVF hybrid gels were made by polymerizing NVF in a solution of PNVF nanogels. This study was conducted to determine whether PNVF hybrid gels can be synthesized by polymerizing it in the presence of nanogels. To determine whether PNVF nanogels had an effect on the network, they were compared to the single network PNVF gels.

5.2.1 Synthesis of Nanogels

PNVF nanogels were synthesized and purified following the same procedure previously reported.[5] Cross-linked PNVF nanogels were fabricated by inverse microemulsion polymerization of NVF with NVEE with 70 x 17 gel composition. NVF (350 µL) was added to 20 mg of initiator, VA-52, and stirred, then 50 mg of NVEE and 165 µL of deionized water were added to this solution. The resulting transparent solution was added to another vial which contained 100 ml of hexane. Surfactants Tween 80 (3 g) and Span 80 (4.1 g) were added to hexane. These surfactants did not dissolve in hexane easily at room temperature and hence the solution was placed in a sonicator to help surfactants dissolve. In the next step, the microemulsion was transferred into 150 mL jacketed three-neck reactor. Nitrogen was purged to

the gel solution for 20 minutes to remove any existing oxygen. The polymerization reaction was then carried out at 70ºC for 24 hours.

The solution was then transferred into centrifugation tubes and centrifuged at 15,000 rpm for 45 minutes at room temperature. Desiccant was discarded and precipitate was immersed in a 10ml DI water. The above step was repeated to remove any hexane from the solution. Hexane was further removed by a vacuum pump. Solution was then transferred to 10kDa dialysis bag and was dialyzed against DI water for two days. Nanogels were purified by changing water every 2-4 hours throughout the 48 hour period. After synthesis process, nanogels concentration was calculated by lyophilizing them. A known volume of dry nanogels was used to make gel solution. Dry nanogels were calculated to be ~ 110 nm in size.[5]

5.2.2 Synthesis of PNVF Hybrid gels

PNVF hybrid gels were synthesized by the same procedure mentioned in Section 4.1.2. In this case, however, the nanogels suspension was used as a solvent instead of DI water used for a single network gel. This solution was left in a sonicator to create a suspension. Any added monomer or crosslinker was vortexed to ensure a homogenous solution. This gel solution was then pipetted into rubber molds. The rubber molds were clamped shut between two glass plates or microscope slides and heated in a water bath at 50°C for 24 hours. The polymerization was carried out under a fume hood as suggested by the Material Safety Data Sheet (MSDS) of NVF. This procedure was repeated for all different gel compositions being studied. Different mechanical properties like Young's modulus, shear modulus, fracture stress and fracture strain were calculated and compared against PNVF single network gels. It was important to calculate

the same parameters as the ones calculated using PNVF single network gels to make a direct comparison.

5.3 CHARACTERIZATION

These gels were swollen according to the technique mentioned in section 4.4.1 before mechanical testing. The swelling ratio as well as other mechanical properties were recorded and compared against single network NVF gels. Nanogels were lyophilized and dissolved in water to obtain the required concentration of 0.4% and 2% incorporation of dry nanogels in the hybrid gel solution at synthesis as shows in equation 5-1. This amount was added to the macrogel composition.

$$\% \, Nanogel = \frac{g_{nanogels}}{g_{monomer} + g_{crosslinker} + g_{nanogels}} \qquad \text{[Eq. 5-1]}$$

5.3.1 Dynamic Mechanical Analysis (DMA)

DMA measures the mechanical properties of materials as a function of time, temperature, and frequency with precise stress control. A small oscillating strain is applied to the sample and the resulting stress is measured to analyze viscoelastic nature of the polymers. In elastic materials the stress in perfectly in phase with the applied strain whereas in ideal viscous material, stress is perfectly out of phase, lagging by 90°. Viscoelastic gels under study exhibit partial lagging.

The value of the complex modulus is expected to be similar to that of the Young's modulus when tan $\delta \ll 1$. Both are a measure of stiffness: the material's resistance to applied stress or strain. The in-phase component is called the storage or the elastic modulus, E' and the out-of phase component is called loss or viscous modulus (E") and it corresponds to the viscous

component.[6] The complex modulus (E^*) can be broken down into storage modulus (E') and a viscous modulus (E'')

$$E^* = E' + iE''^* \quad \text{[Eq. 5-2]}$$

$$\tan\delta = E''/E' \quad \text{[Eq. 5-3]}$$

5.4 RESULTS

This section includes all the experimental data collected on PNVF hybrid gels and analysis of these results. Constant crosslinker concentration was used in these sets of examples as single network gels were compared against hybrid gels. The effect of increasing monomer concentration and nanogels concentration was tabulated in Tables 5-1 and 5-3 and these were compared against PNVF macrogels.

These effects can be clearly seen in Figures 5-2 and 5-3. Fracture stress of hybrid gels increased with the increase in monomer concentration. Fracture strain also increased in all but 25x1 gel compositions. In addition to fracture properties seen in Fig. 5-2, Young's modulus and shear modulus was also calculated. Young's modulus and shear modulus also increased with the increase in monomer concentration.

Figure 5-3 shows representative stress-strain curves of hybrid gels synthesized with 2% nanogels concentration as opposed to 0.4% seen in Fig. 5-4. Hybrid gels behaved like a typical hydrogel even with an increase in nanogels concentration as there was an increase in fracture stress with an increase in monomer composition. Other mechanical properties such as Young's modulus and shear modulus were also calculated and tabulated in Table 5-3.

Figure 5-5 represents stress-strain curves of PNVF gels with 2%, 0.4% and without any nanogels addition to the network. There was not a significant difference seen in the fracture properties with an increase in nanogels concentration.

Table 5-1 Effect of PNVF nanogels on gel properties. All samples were made with nanogels concentration of 4.0g/L for the results in Table 5-1. Standard deviations were calculated using Excel STDEV function using 3 gel samples (n=3) from the same batch.

Gel type with 0.4% nanogel	Swelling Degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]	E/G	Toughness (AUC) (J/m^3)
10x1	28.9±3.2	128.3±2.1	39.5±2.1	18.0±3.3	15.2±2.8	3.2±0.2	104.3±2.2
15x1	23.9±3.9	155.1±4.4	49.0±1.5	28.5±2.1	16.5±3.2	3.2±0.1	212.4±4.3
20x1	20.5±3.8	191.0±3.9	60.2±1.6	35.2±1.1	18.2±2.4	3.2±0.1	327.5±2.5
25x1	16.1±2.9	280.2±3.4	75.6±2.1	50.4±2.4	16.2±2.1	3.7±0.1	341.1±3.1

Table 5-2 Network Parameters of PNVF hybrid gels of different monomer concentrations calculated from data in Table 5.1.

Gel type with 0.4% nanogel	Swelling Degree Q (g/g)	Crosslink density ρ_x (mol/cm^3)	χ	M_c (g/mol)
10x1	28.9	2.30E-04	0.39	5.21E+03
15x1	23.9	2.68E-04	0.43	4.47E+03
20x1	20.5	3.13E-04	0.42	3.83E+03
25x1	16.1	3.63E-04	0.41	3.31E+03

Table 5-3 Increasing nanogels concentration did not significantly improve fracture properties.

Gel Type with 2% nanogel	Swelling Degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]	E/G	Toughness AUC (J/m^3)
10x 1	32.3±2.3	85.6±4.3	31.6±2.9	24.2±4.5	19.7±2.5	2.7±0.3	113.8±7.3
15x1	29.7±1.9	171.1±4.7	51.6±4.1	28.6±4.2	16.2±2.7	3.3±0.3	207.3±5.2

Table 5-4 Effect of varying monomer and crosslinker concentration on mechanical properties of PNVF gels without addition of nanogels. These properties were compared against Tables 5-1 and 5-3.

Gel type with 0% nanogel	Swelling Degree Q (g/g)	Young's Modulus E (kPa)	Shear Modulus G (kPa)	Fracture Stress (kPa)	Fracture Strain [%]	E/G	Toughness AUC (J/m^3)
10x1	30.6±0.2	138.1±3.1	48.9±9.1	16.5±0.2	14.8±0.7	2.8±0.5	142.2±4.3
15x1	25.5±0.6	172.4±1.5	60.7±2.1	27.6±0.4	16.8±0.1	2.9±0.1	305.6±4.6

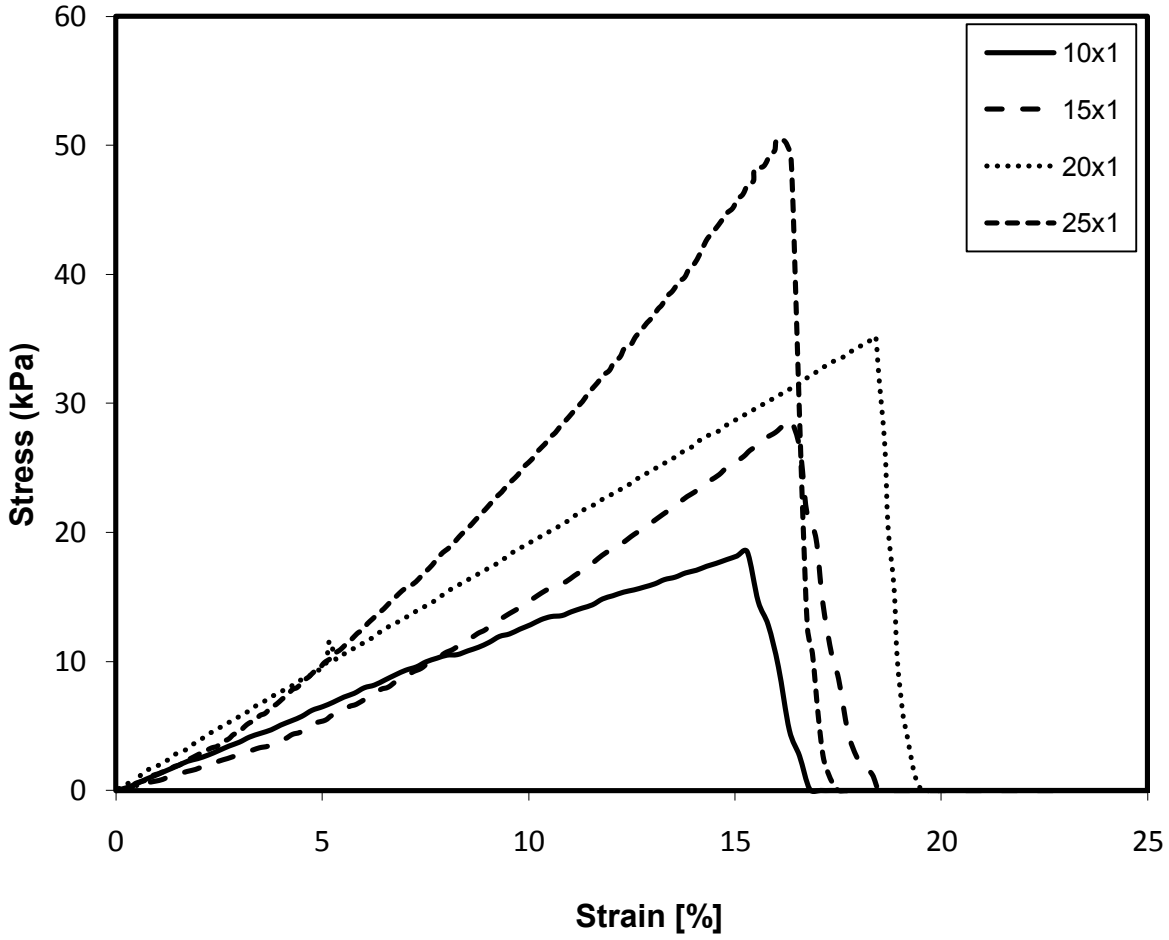

Fig. 5-3 Static stress-strain response of PNVF gels with PNVF nanogels incorporated swollen to equilibrium in water as a function of monomer concentration for 1% and 3% crosslinking ratio. All gel samples were synthesized using 0.4% PNVF nanogels.

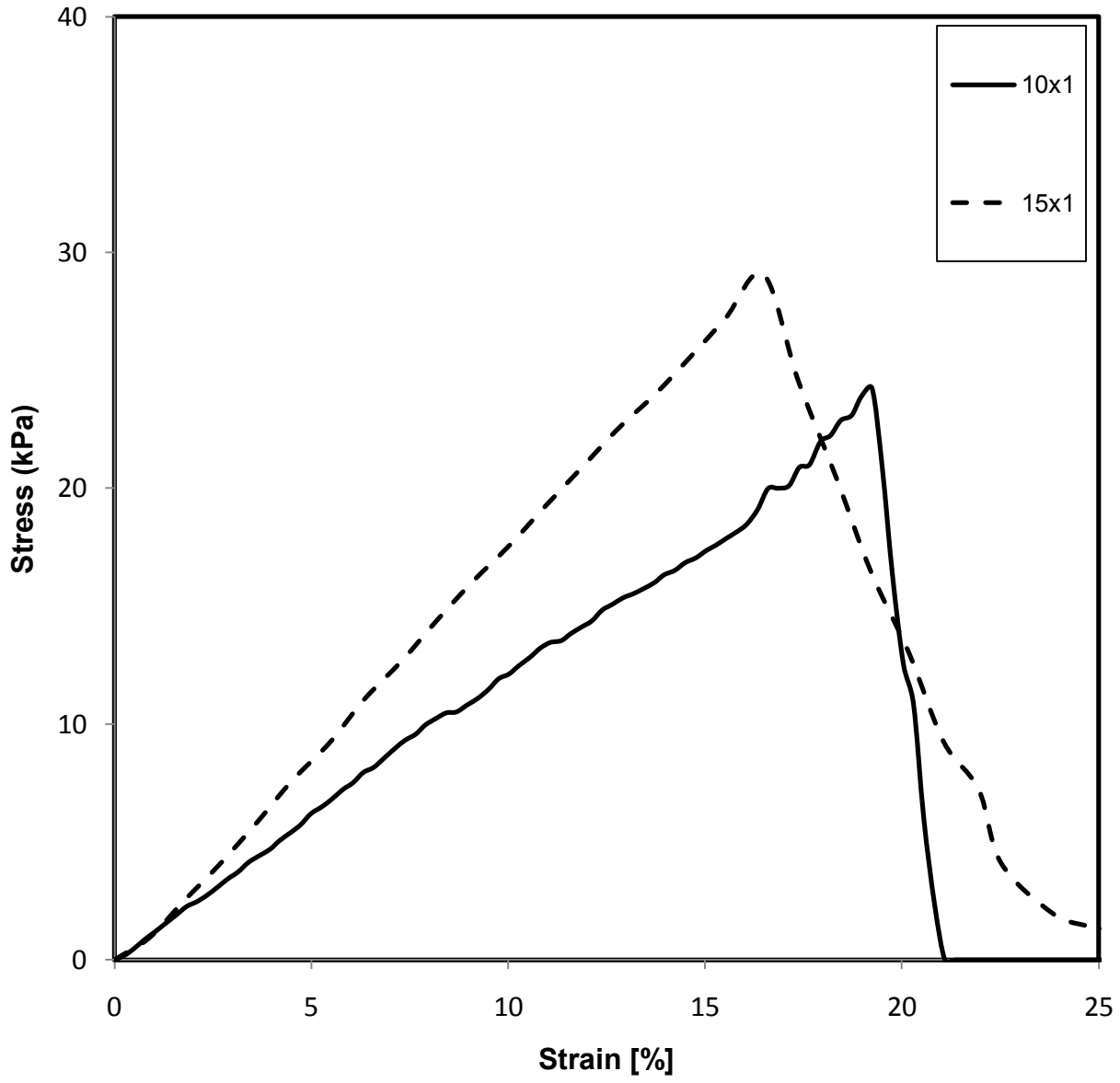

Fig. 5-4 Static properties of PNVF macrogels network with PNVF nanogels. Figure shows hybrid gels synthesized with 2% nanogels incorporation. Elongation at break increased in this gel formulation compared to the ones synthesized with 0.4% nanogels.

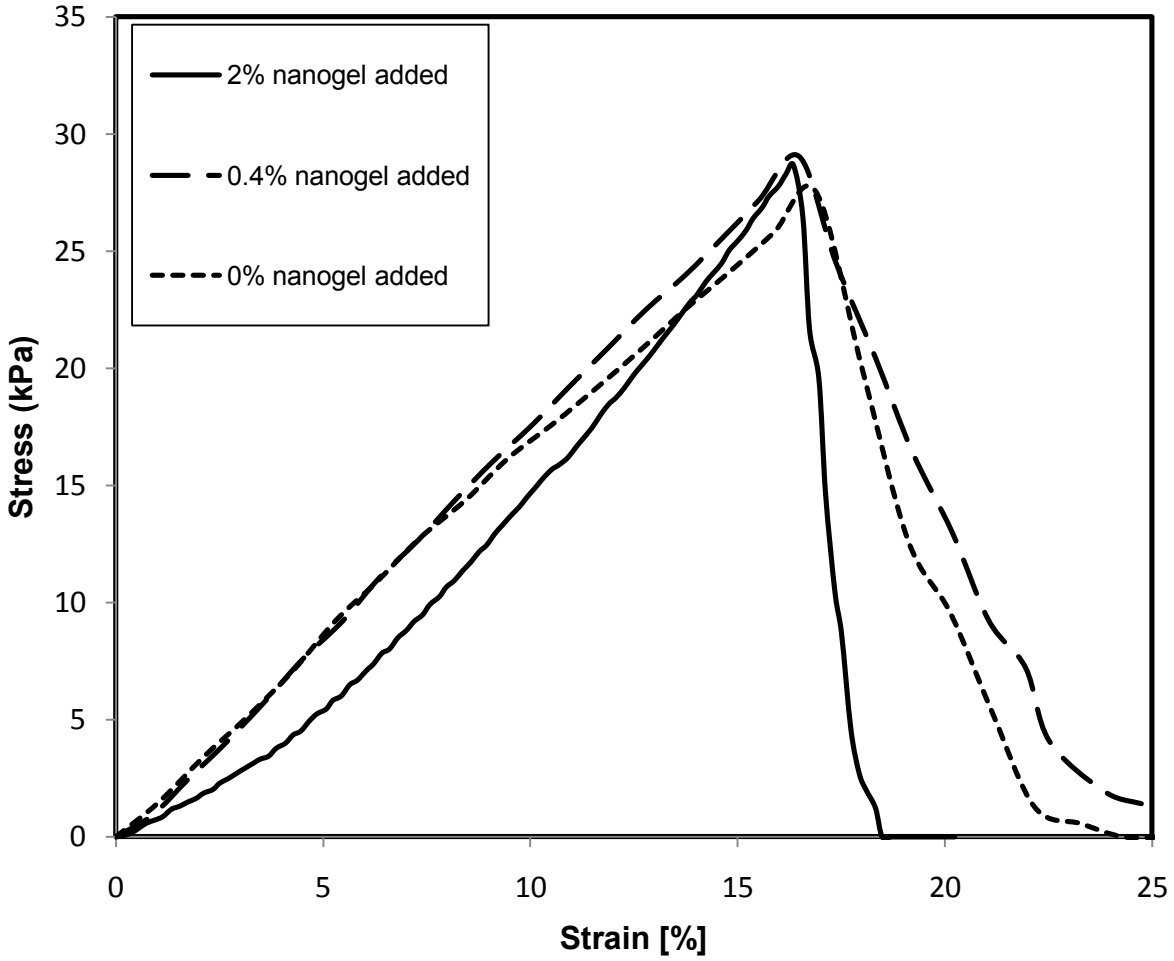

Fig. 5-5 Stress-strain curves representing PNVF gels fracture properties with 2%, 0.4% and 0% addition of nanogels in the network synthesized with 15x1 gel composition. There was not a significant change seen in fracture properties with an increase in nanogel concentration.

In addition to tensile experiments, DMA experiments were also conducted on two gel formulations to ensure the presence of nanogels in the network. Figure 5-6 is the result of DMA test done on 10x1 gel formulation. DMA test calculated loss and storage modulus against frequency range from 0.5-50 rad/sec. Figure 5-6 shows that loss modulus (E'') increased as the nanogel concentration increased proving an increase in viscous region with the addition of nanogels in the network. However, the storage modulus increased with a decrease in nanogels concentration, even though (E') calculated from DMA tests were comparable to Young's modulus calculated from stress-strain curves. Figure 5-7 is a similar test done on 20x1 gel formulation. There was an increase in loss modulus with an increase in nanogels concentration showing the presence of nanogels in the network and an increase in storage modulus with an increase in amount of nanogels in the network solution.

5.5 DISCUSSION

The last research goal of this study was to synthesize PNVF hybrid gels by incorporating nanogels in the network. It was hypothesized that these nanogels will act as non-covalent crosslinkers, resulting in an increase in the elasticity of the network. This theory is similar to slide-rings and Olympic loops described in section 5.1.1. Swollen nanogels should allow polymer chains to move through them under tension resulting in an increase in fracture strain.

Table 5-1 lists the results of all tensile and swelling tests done on hybrid gels made with 0.4% nanogels whereas tables 5-3 and 5-4 shows results of hybrid gels made 2% and 0% nanogels incorporation in the network. Swelling degree of all these gels decreased with an increase in monomer concentration which is what was expected with increased crosslinking efficiency. In addition to swelling, stress-strain curves show the fracture properties of these gels. Fracture stress increased with an increase in monomer concentration as the crosslinking

efficiency increased. Although, it was hypothesized that fracture strain would increase with an increase in amount of nanogels in the solution, there was not a significant difference seen in the fracture strain of these gels. This could be because these nanogels were made with ~17% crosslinker and 70% monomer which mitigate the effect of nanogels and results in an increase in overall amount of crosslinker present in the solution. It could be that nanogels are simply adding a filler effect to the network as oppose to the hypothesized non-covalent crosslinkers.

In addition to tensile experiments, DMA tests were carried on these gels to ensure the presence of nanogels in the network. Figures 5-6 and 5-7 show that loss modulus increased with an increase in nanogels concentration proving the increase in nanogels in the network. This is suggestive that the nanogels are embedded in the network but perhaps not interpenetrated by polymer chains, and this may be the reason for the lack of notable property improvement. As noted above, the nanogel formulation was consistent with very high crosslinking. There was not a coherent trend seen in storage modulus, though they were calculated to be comparable to the Young's modulus calculated from stress-strain curves of these gels.

It was concluded from these experiments that hybrid gels could be synthesized using the described procedure. Increasing nanogels concentration increased fracture stress but did not have a significant impact on fracture strain which could be due to the high amount of crosslinker present in the nanogels.

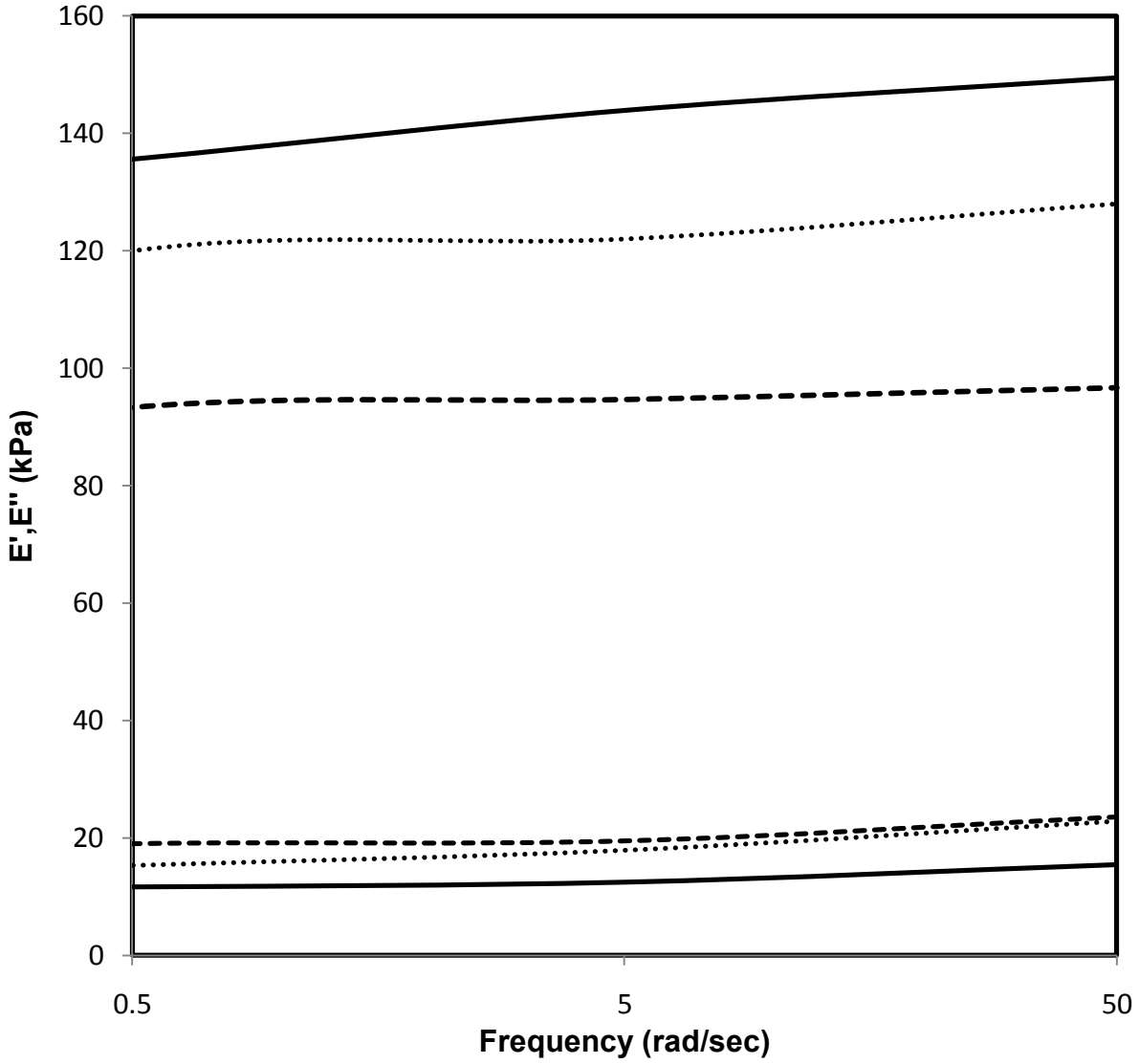

Fig. 5-6 Dynamic storage and loss moduli over a frequency range of 0.5-50 for a PNVF 10x1 hybrid gel. ___, no nanogel; ……, 0.4% nanogel; -----, 2% nanogel.

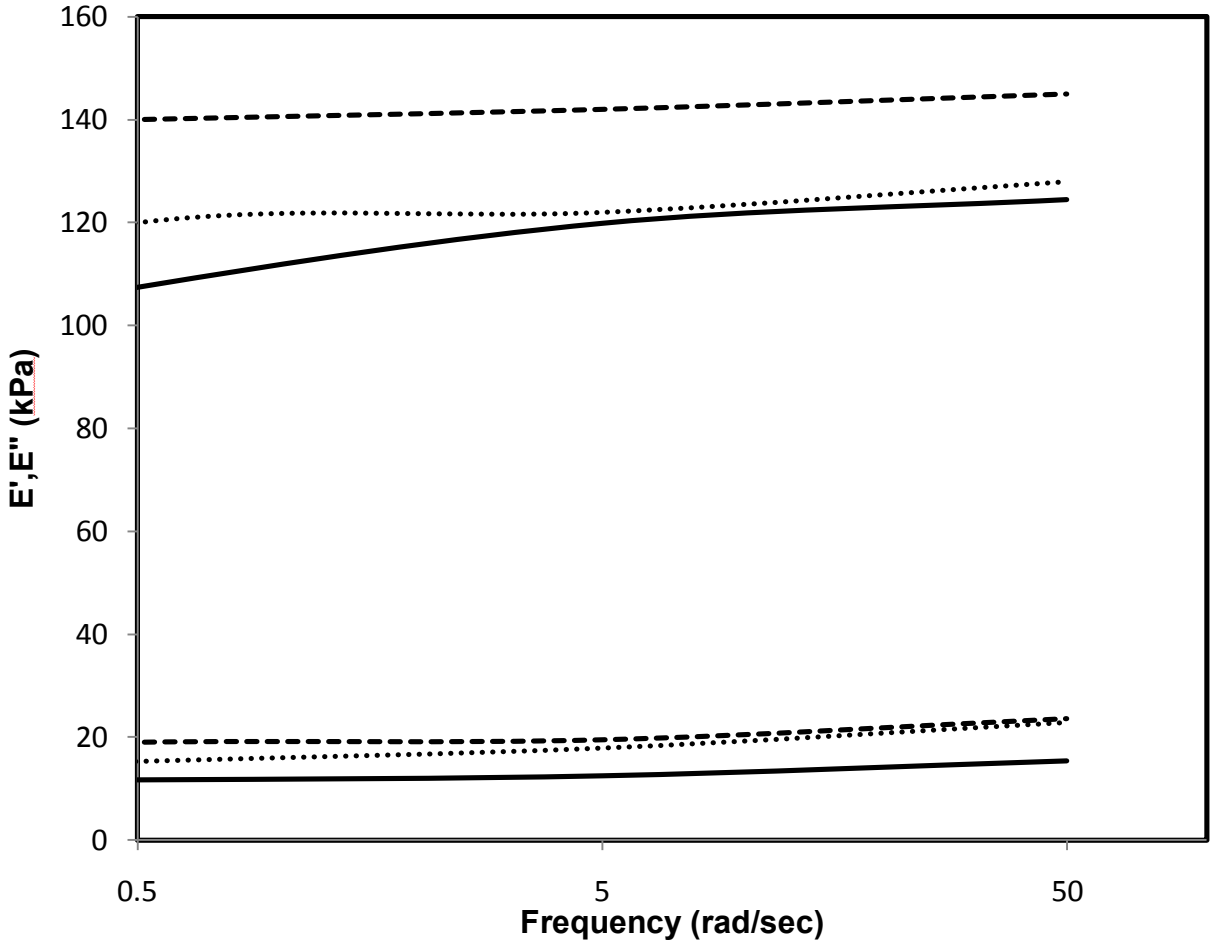

Fig. 5-7 Dynamic storage and loss moduli over a frequency range of 0.5-50 for a PNVF 20x1 hybrid gel. Loss modulus increased with an increase in nanogel concentration due to the presence of nanogels in the network. ___, no nanogel; ……, 0.4% nanogel; -----, 2% nanogel.

Chapter 5- References

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Chapter 6

Conclusions and Recommendations

The main focus of this study was to synthesize and characterize poly (N-vinyl formamide) (PNVF) gels as an alternative to the widely used gels of polyacrylamide (PAAm). Prior to this work, there had been no systematic methodology established in the literature for synthesis of these gels. N-vinyl formamide (NVF) is an isomer of acrylamide (AAm) but it was found that it does not crosslink with the conventional acrylamide crosslinkers, likely due to differences in propagating radical polarity. 2-(N-vinylformamido)ethyl ether (NVEE) and NVF have similar radical polarity and thus readily copolymerize resulting in well crosslinked gels which resulted in high (86%-93%) monomer conversion. To initiate this propagation reaction, Vazo-44, a thermal initiator was used at 50°C. A combination of Vazo-44 and NVEE synthesized transparent PNVF gels with properties comparable to the well known polyacrylamide gels, suggesting that PNVF gels could substitute for applications in which PAAm gels are used.

PNVF hydrogels were then fully characterized by swelling and mechanical tests. Tensile tests were done to determine fracture properties, Young's modulus, shear modulus and toughness of gel samples whereas established theory and measured values were used to calculate crosslink density and solubility parameters (χ). Fracture properties were calculated from tensile stress-strain curves. An increase in monomer concentration increased fracture stress due to increasing crosslinking efficiency at higher monomer concentrations. The moduli also increased with an increase in monomer concentration for the same reason. Other network parameters such as crosslink density, Flory-Huggins solubility parameters (χ) and average distance between crosslinks were also calculated using the two main models of rubber elasticity. The difference

between affine and phantom model is discussed in detail in Chapter 3. It was determined that use of the phantom model led to calculation of more reasonable χ values, consistent with previous work showing that the phantom model generally works better than the affine model for highly swollen gels.

Chi (χ) values calculated for PAAm gels were lower than literature value as well as recent work done on these gels by the hydrogel research group at KU. χ values should be independent of the gel formulation as it is a property of the polymer and not expected to be significantly affected by the crosslinking. These values are related to crosslink density which is calculated from mechanical tests for shear modulus (G). On the other hand, swelling degree yields polymer volume fraction. Swelling degree combined with crosslink density yields χ. As χ is related to crosslink density calculated from shear modulus, lower χ values could be a result of increased strain rate which led to higher shear modulus. All gels were tested at 0.5mm/sec but recent work shows that strain rate has an effect on fracture properties and thus on shear modulus. For this study, higher strain rates were used which led to higher modulus than reported in the literature and found in more recent studies done by the hydrogel group at KU.

Although moduli calculated from this study are higher than expected resulting in lower χ values, there is consistency between the results found from this study and those published in the literature. The E/G ratio came out around 3 signifying a Poisson's ratio around 0.5, which is reported for hydrogels and other incompressible materials following neo-Hookean model. Furthermore, stress-strain curves were linear despite expecting neo-Hookean behavior as high enough strain levels were not reached to observe non-linearity: crosslinking efficiency and crosslink density were comparable to literature values as well.

As shear modulus was apparently higher than expected, based on the observation of lower than expected χ values, it is strongly recommended to repeat tensile tests using lower strain rates to identify a strain rate where the modulus becomes independent of strain rate. In addition to this, it would be helpful to either synthesize pure crosslinker or test other crosslinkers with similar chemical structure available in the industry to synthesize PNVF gels. χ values should also be calculated from linear polymers to confirm those calculated from the shear modulus and gel swelling theory.

In addition to PNVF macrogels described above, PNVF hybrid gels were also synthesized by creating a network of PNVF macrogels and PNVF nanogels. Nanogels were added to the network to improve fracture properties as hydrogels are generally brittle in nature. In Chapter 5, it was hypothesized that nanogels suspension added to PNVF solution would increase elasticity of the network by adding non-covalent crosslinkers. However, there was not a significant change seen in the fracture properties of these gels. This could be because of low nanogels concentration compared to the monomer-crosslinker concentration originally used or the high monomer and crosslinker ratios used in the nanogels, which may have made them impermeable to the macrogel network. Future work should conduct more experiments on these hybrid gels with a wide range of nanogels incorporation to clearly establish the effect of these nanogels on mechanical properties of PNVF gels. Such experiments should allow a more accurate description of the nature of hybrid gels and interaction of the network with nanogels. In addition to different nanogel concentration, the crosslinker added to this network should be controlled to see the effect of nanogels only.

Appendix A

To synthesize 2-(N-vinylformamido)ethyl ether NVEE (crosslinker) a mixture of N-vinyl formamide (21 g), potassium t-butoxide (35.44 g) and dicyclohexyl-18-crown-6 (3 g) in anhydrous THF was stirred vigorously at room temperature for 45 minutes to activate secondary amine group and then cooled in an ice bath. Bis(2-bromthyl)ether (27.8 g) was added drop wise, and the mixture was stirred at room temperature for 72 hours. To purify potassium bromide, salt was removed by filtration and then the reaction mixture was concentrated under vacuum and diluted with 300 mL of water. The crude product was obtained by extraction with chloroform five times (50mL× 3). The combined organic layers were washed twice with brine and dried over anhydrous sodium sulfate. The resulting product was recovered after concentration in a vacuum distillation column and purification by chromatography on silica (ethylacetate/hexane: 8:2 v/v) [4].

Appendix B

STRESS-STRAIN PLOTS OF PNVF GELS: EXPERIMENTAL DATA

Table B.1.1 Stress-strain data for poly (N-vinyl formamide) gel.

Length (mm)	15.54	
Width (mm)	1.7	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.151796	0.324324	0.009698
3.107755	0.648648	0.019334
4.985091	0.973001	0.02891
7.22761	1.326811	0.039285
9.416599	1.651135	0.048734
11.43715	2.004946	0.058974
13.73251	2.32927	0.0683
15.78614	2.68308	0.078407
17.90448	3.007404	0.087614
19.89545	3.331757	0.096765
21.86845	3.685568	0.106683
23.87431	4.009892	0.115719
25.99407	4.363702	0.125514
27.96462	4.688026	0.134437
29.82552	5.041837	0.144111
31.77348	5.366189	0.152926
33.7936	5.690513	0.161689
36.11391	6.044324	0.171191
38.21967	6.368648	0.179848
40.19703	6.722459	0.189237
42.14332	7.046783	0.197792
44.05879	7.400593	0.207071
46.01882	7.724946	0.215527
47.8055	8.04927	0.223936
49.92577	8.40308	0.233056
51.96566	8.727404	0.241368
53.80178	9.051757	0.249636
55.68271	9.376081	0.257859
57.61629	9.75935	0.267519
59.52573	10.0837	0.275647
61.28468	10.40803	0.283731
63.24894	10.76184	0.292502
65.43177	10.91116	0.300498

Data used in Figure 4-4 (25x3)

Table B.1.2. Stress-strain data for poly (N-vinyl formamide) gel. (25x3,ii)

Length (mm)	15.62	
Width (mm)	1.67	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.12579	0.348226	0.010411
2.50715	0.753138	0.022426
4.40278	1.128356	0.033474
6.32898	1.508975	0.0446
8.22375	1.886891	0.055565
10.3101	2.294506	0.067303
12.3472	2.672427	0.078104
14.4223	3.050343	0.088828
16.5426	3.455259	0.100234
18.7068	3.833176	0.110802
20.8048	4.213808	0.121372
22.9081	4.588996	0.131718
25.0913	4.996611	0.142878
27.3183	5.377238	0.153225
29.4255	5.755146	0.163429
31.6956	6.157364	0.174214
34.0721	6.535314	0.184278
36.3468	6.913222	0.194275
38.7165	7.29113	0.204206
41.1586	7.698745	0.214846
43.6658	8.079372	0.224714
46.0778	8.45728	0.234448
48.5827	8.862218	0.24481
51.0669	9.240126	0.254417
53.5176	9.618033	0.263964
56.0739	9.998661	0.27352
58.5619	10.40356	0.28362
61.2647	10.78146	0.292987
63.8428	11.15941	0.302298
65.6298	11.56163	0.312145

Data for PNVF 25x3(ii)

Table B.1.3. Stress-strain data for poly (N-vinyl formamide) gel. (25x3,iii)

Length (mm)	15.72	
Width (mm)	1.64	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
14.90085	0.423758	0.012659
1.464716	0.916497	0.027246
4.52235	1.37639	0.040734
7.379835	1.836278	0.054101
9.41208	2.296166	0.06735
12.87555	2.792195	0.081511
15.18675	3.248803	0.094429
19.1238	3.711976	0.107421
20.097	4.204715	0.12112
22.7721	4.664608	0.133794
24.57495	5.124491	0.146362
28.55595	5.584369	0.158827
31.0617	6.077138	0.172069
34.7964	6.537016	0.184323
38.40975	7.000204	0.196567
41.75265	7.496232	0.209569
45.9528	7.952851	0.221441
50.15475	8.415988	0.233388
54.4155	8.875916	0.24516
58.05615	9.368635	0.257671
61.0398	9.828513	0.269256
64.8006	10.28844	0.280755
65.46945	10.78442	0.29306

Data for PNVF 25x3(iii)

Table B.2.1 Stress-strain data for poly (N-vinyl formamide) gel. (20x3)

Length (mm)	15.59	
Width (mm)	1.9	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.452252	0.77325	0.02302
3.43854	1.616659	0.047732
5.093424	2.389909	0.070037
7.076352	3.233386	0.093995
9.201276	4.006568	0.115626
10.908588	4.77975	0.136951
12.74544	5.553	0.15798
14.38296	6.396477	0.180589
16.1604	7.169659	0.201021
17.67204	8.013136	0.223001
19.16424	8.786318	0.242873
20.86944	9.629795	0.264261
22.50984	10.40298	0.283606
24.0378	11.17623	0.302711
25.77972	12.0197	0.323284
27.55524	12.79289	0.341903
28.9842	13.63636	0.361964
30.819	14.47984	0.381768
32.52912	15.25302	0.399703
34.2684	16.02627	0.417436
36.05484	16.79945	0.43497
37.60104	17.64293	0.453879
39.45312	18.41611	0.471015
41.0868	19.25959	0.489502
42.82468	20.79277	0.506262

Data used in Figure 4-4 (20x3)

Table B.2.2 Stress-strain data for poly (N-vinyl formamide) gel. (20x3,ii)

Length (mm)	15.82	
Width (mm)	1.79	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.0537	0.875232	0.02603
3.28345	1.782112	0.052533
5.47727	2.688992	0.078576
7.72983	3.595856	0.104175
10.001	4.502736	0.129345
12.1653	5.399072	0.153817
14.3508	6.295392	0.177897
16.544	7.212816	0.202154
18.724	8.109136	0.225483
20.8614	9.016016	0.248727
23.0289	9.922896	0.271623
25.2248	10.82976	0.29418
27.3003	11.72608	0.316153
29.4642	12.62243	0.337818
31.6067	13.52928	0.359431
33.7405	14.42563	0.380503
35.8239	15.3325	0.401535
37.9254	16.2288	0.422048
40.0436	17.13568	0.442535
42.0782	18.04256	0.462759
42.9321	18.93888	0.482498

Data for PNVF 20x3(ii)

Table B.2.3 Stress-strain data for poly (N-vinyl formamide) gel. (20x3,iii)

Length (mm)	15.77	
Width (mm)	1.64	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.902817	0.829606	0.024684
2.407863	1.702315	0.050219
4.4112	2.682788	0.078399
6.366767	3.544709	0.102742
8.322233	4.503606	0.129369
10.23937	5.365543	0.152909
12.1616	6.335212	0.178958
14.1597	7.304897	0.204567
16.03023	8.166834	0.226972
17.92583	9.147275	0.252063
19.81347	10.00923	0.273785
21.70653	10.96813	0.297593
23.5765	11.84083	0.318944
25.45353	12.8105	0.342325
27.44817	13.76942	0.365104
29.28663	14.63135	0.385299
31.15747	15.62258	0.408203
32.96207	16.48452	0.427852
34.8345	17.44348	0.449428
36.7883	18.31606	0.468808
38.5443	19.28583	0.490074
40.5833	20.25543	0.511057
42.8834	21.01748	0.529486

Data for PNVF 20x3(iii)

Table B.3.1 Stress-strain data for poly (N-vinyl formamide) gel. (15x3)

Length (mm)	15.65	
Width (mm)	1.76	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.897037	1.03194	0.030643
2.42037	2.06388	0.060673
3.88247	3.095911	0.090116
5.62898	4.221672	0.121589
7.3338	5.253612	0.149872
8.90744	6.379373	0.180134
10.6951	7.411313	0.207351
12.2945	8.537074	0.236496
13.9443	9.569014	0.262729
15.4949	10.60105	0.288522
17.0315	11.72681	0.316171
18.5937	12.75875	0.341086
20.2446	13.88451	0.367816
21.7793	14.91645	0.391921
23.2286	16.04221	0.4178
24.7457	17.07424	0.441155
26.319	18.10618	0.464168
28.1261	19.23194	0.488899
29.7661	20.26388	0.511238
31.3061	21.38964	0.535261
32.7219	22.21158	0.556973

Data used in Figure 4-4 (15x3)

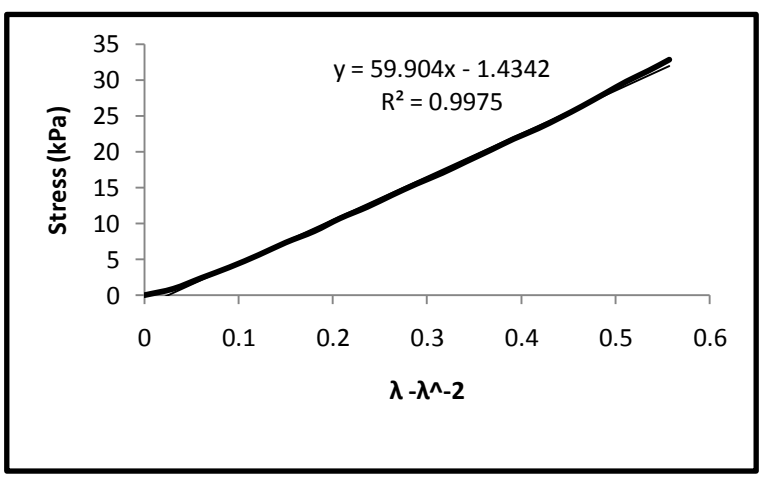

Table B.3.2 Stress-strain data for poly (N-vinyl formamide) gel. (15x3, ii)

Length (mm)	15.83	
Width (mm)	1.81	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.963553	1.36452	0.040387
2.92073	2.74839	0.080266
4.88268	4.13226	0.119113
6.70012	5.506455	0.156722
8.64391	6.9	0.193926
10.6365	8.274195	0.22974
12.5865	9.66774	0.265216
14.5484	11.04195	0.29941
16.5999	12.42581	0.333091
18.6988	13.80968	0.366053
20.748	15.1839	0.398108
22.7919	16.5774	0.429955
24.9829	17.95155	0.460741
27.0135	19.33545	0.491154
29.2144	20.7096	0.520792
31.3863	22.0935	0.550101
32.9107	23.5714	0.58083

Data for PNVF 15x3(ii)

Table B.3.3 Stress-strain data for poly (N-vinyl formamide) gel. (15x3, iii)

Length (mm)	15.93	
Width (mm)	1.88	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.28029	1.119798	0.033223
2.40037	2.4408	0.071493
4.24702	3.674326	0.106369
6.17516	4.899092	0.140216
8.10017	6.123872	0.173319
10.1724	7.427395	0.207771
12.3374	8.660908	0.239668
14.4242	9.885674	0.27069
16.5699	11.20669	0.303459
18.7793	12.43146	0.333227
20.9103	13.65628	0.362434
23.0295	14.88102	0.391099
25.2805	16.19322	0.421239
27.5294	17.41809	0.44886
29.8557	18.64283	0.476005
32.2362	19.96385	0.504775
32.7601	21.18911	0.531008

Data for PNVF 15x3(iii)

Table B.4.1 Stress-strain data for poly (N-vinyl formamide) gel. (10x3)

Length (mm)	15.73	
Width (mm)	1.87	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.315174	0.285936	0.008554
1.9794	0.901807	0.026813
2.75482	1.209731	0.03586
3.50689	1.539643	0.045492
4.81724	2.155514	0.063311
6.92798	3.101274	0.090268
8.29919	3.739133	0.108179
8.85728	4.047057	0.11675
9.51381	4.332993	0.124666
10.9074	4.99284	0.142775
11.4627	5.278753	0.150555
12.6883	6.224536	0.176007
13.1957	6.554471	0.184787
14.7449	7.19233	0.201616
15.2691	7.500254	0.209674
16.247	8.116102	0.225663
17.8318	9.061885	0.249894
18.426	9.391797	0.258256
19.5253	10.31557	0.281431
20.0673	10.62349	0.289078
21.5261	11.56928	0.312332
22.5848	12.51506	0.335239
23.3973	13.13093	0.349974
24.561	13.76877	0.357811
25.0727	14.07671	0.365089
25.305	14.40663	0.372334
25.5639	14.71455	0.380059
25.9454	14.8225	0.387234
26.1999	15.15239	0.394377
26.2807	15.26033	0.401993
26.4769	15.29024	0.409069
26.7461	15.33017	0.416615

Data for PNVF 10x3

Table B.4.2 Stress-strain data for poly (N-vinyl formamide) gel. (10x3,ii)

Length (mm)	15.86	
Width (mm)	1.89	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.394027	0.445059	0.013293
0.877503	0.962567	0.028603
1.540973	1.442123	0.042652
2.215143	1.928583	0.05677
2.878313	2.411588	0.070657
3.608535	2.932551	0.085494
4.32152	3.415561	0.09912
5.047805	3.898567	0.112623
5.78991	4.41608	0.126958
6.54738	4.899086	0.140215
7.28168	5.385561	0.153451
8.017835	5.86508	0.166384
8.781955	6.386043	0.180311
9.561405	6.872513	0.193201
10.29893	7.355508	0.205892
11.09346	7.869572	0.219282
11.92524	8.35262	0.231759
12.72138	8.835615	0.244132
13.55078	9.31861	0.256405
14.40551	9.839572	0.269534
15.28303	10.32604	0.281692
16.12723	10.80904	0.293668
17.00395	11.32658	0.306398
17.87342	11.80957	0.318184
18.73116	12.29257	0.32988
19.62587	12.77904	0.341572
20.49667	13.29652	0.353913
21.44265	13.77952	0.365343
22.34498	14.26257	0.37669
23.28855	14.77663	0.388677
24.23694	15.26305	0.399934
26.6318	15.8261	0.411034

Data for PNVF 10x3(ii)

Table B.4.3 Stress-strain data for poly (N-vinyl formamide) gel. (10x3,iii)

Length (mm)	15.85	
Width (mm)	1.92	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.426749	0.47567	1.016449
1.329797	0.968539	1.710484
2.218294	1.461409	2.296352
3.130581	1.95427	2.839692
4.050405	2.447139	3.362984
4.926947	2.934278	3.869673
5.812074	3.421409	4.370255
6.70032	3.920009	4.878697
7.58322	4.407139	5.372936
8.448867	4.900009	5.871281
9.326705	5.392878	6.36841
10.21604	5.885739	6.864648
11.05662	6.37287	7.354473
11.933	6.860017	7.843831
12.80071	7.35287	8.338537
13.6649	7.840017	8.827221
14.50868	8.332878	9.321398
15.35979	8.82	9.80963
16.21766	9.31287	10.30347
17.04167	9.805739	10.79717
17.89966	10.29287	11.28503
18.70015	10.78574	11.77854
19.54716	11.27861	12.27198
20.39588	11.77148	12.76535
21.23152	12.26435	13.25866
22.08177	12.75722	13.75193
22.93126	13.24435	14.23942
23.75078	13.73148	14.72687
24.5641	14.22435	15.22003
25.3871	14.71722	15.71317
26.18374	15.21009	16.20628
26.98545	15.63296	16.69937

Data for PNVF 10x3(iii)

Table B.5.1 Stress-strain data for poly (N-vinyl formamide) gel. (25x1)

Length (mm)	15.85	
Width (mm)	1.92	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0.00
0.897037	0.29484	0.01
2.42037	0.58968	0.02
3.88247	0.884546	0.03
5.62898	1.206192	0.04
7.3338	1.501032	0.04
8.90744	1.822678	0.05
10.6951	2.117518	0.06
12.2945	2.439164	0.07
13.9443	2.734004	0.08
15.4949	3.02887	0.09
17.0315	3.350516	0.10
18.5937	3.645356	0.11
20.2446	3.967002	0.11
21.7793	4.261842	0.12
23.2286	4.583488	0.13
24.7457	4.878354	0.14
26.319	5.173194	0.15
28.1261	5.49484	0.16
29.7661	5.78968	0.16
31.3061	6.111326	0.17
32.8219	6.406166	0.18
34.3137	6.727812	0.19
35.8402	7.022678	0.20
37.2317	7.317518	0.20
38.883	7.639164	0.21
40.4717	7.934004	0.22
41.9017	8.22887	0.23
43.3666	8.52371	0.24
44.8725	8.872136	0.25
46.3596	9.167002	0.25
47.7295	9.461842	0.26
49.2593	9.783488	0.27
50.7763	10.07833	0.28
52.1415	10.37319	0.28
53.349	10.66803	0.29

Data for Fig. 4-3 PNVF 25x1

Table B.5.2 Stress-strain data for poly (N-vinyl formamide) gel. (25x1,ii)

Length (mm)	15.83	
Width (mm)	1.91	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0.00000	0	0
0.93395	0.282256	0.011243
2.49089	0.579176	0.022968
4.56331	0.912761	0.036018
6.58631	1.206011	0.047385
8.60921	1.532256	0.059917
10.59245	1.825511	0.07108
12.58097	2.15542	0.083526
14.64797	2.485335	0.095855
16.58300	2.778591	0.106717
18.54397	3.112165	0.118963
20.49669	3.405426	0.129635
22.45503	3.73167	0.141404
24.38948	4.028591	0.152022
26.33124	4.3585	0.163719
28.39466	4.68475	0.175182
30.29652	4.978006	0.185399
32.23186	5.31525	0.197049
34.09869	5.608506	0.207094
36.01379	5.934773	0.218177
37.91379	6.231648	0.228178
39.86897	6.561591	0.239201
41.90690	6.891477	0.250128
43.76552	7.184773	0.259764
45.70345	7.518352	0.270635
46.51131	7.811591	0.280115
47.14048	8.137841	0.290578
49.46672	8.43108	0.299909
50.28110	8.764659	0.31044
50.99028	9.090909	0.320654
51.53969	9.384148	0.329765
51.33317	9.717727	0.340049
52.09300	10.01097	0.34902
52.20600	10.33722	0.358925
53.00690	10.63051	0.367763
53.66438	10.99409	0.377739

Data for PNVF 25x1(ii)

Table B.5.3 Stress-strain data for poly (N-vinyl formamide) gel. (25x1,iii)

Length (mm)	16.33	
Width (mm)	1.99	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.786343	0.309051	0.009243
2.450336	0.629277	0.01876
4.087515	0.949503	0.028218
5.76853	1.269723	0.037616
7.463433	1.589949	0.046956
9.078582	1.906452	0.05613
10.70955	2.222949	0.065249
12.34627	2.546898	0.074525
13.97313	2.863395	0.083533
15.56821	3.183621	0.092592
17.18575	3.503847	0.101597
18.82448	3.824068	0.110548
20.37336	4.140565	0.119344
21.98821	4.457073	0.128088
23.58709	4.777288	0.136883
25.17948	5.093797	0.145527
26.73425	5.414017	0.154221
28.30254	5.730508	0.162766
29.88328	6.050734	0.171362
31.40164	6.37096	0.17991
32.98261	6.687458	0.188311
34.45761	7.007684	0.196764
36.01836	7.32791	0.205169
37.58224	7.648136	0.213529
39.12201	7.968362	0.221842
40.68873	8.288588	0.230111
42.25403	8.605085	0.238239
43.7641	8.921582	0.246323
45.26276	9.241808	0.25446
46.77925	9.562034	0.262554
48.24716	9.88226	0.270604
49.71709	10.20249	0.278613
51.24045	10.52271	0.28658
52.67694	10.83921	0.294413
53.43221	11.12171	0.302207

Data for PNVF 25x1(iii)

Table B.6.1 Stress-strain data for poly (N-vinyl formamide) gel. (20x1)

Length (mm)	16.01	
Width (mm)	1.49	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.194724	0.112035	0.003357
3.732156	1.360392	0.040266
3.732156	1.360392	0.040266
5.06124	1.816518	0.053529
6.49266	2.280663	0.066906
7.9494	2.7368	0.079936
8.955768	3.080891	0.089692
11.92369	4.225232	0.042252
13.21872	4.681358	0.046814
15.52776	5.705656	0.057057
15.74472	5.817691	0.058177
16.51884	6.161793	0.061618
17.87964	6.729954	0.0673
18.6961	7.074056	0.070741
20.21796	7.762249	0.077622
21.069	8.210389	0.082104
23.93304	9.586786	0.095868
24.16848	9.706818	0.097068
25.84752	10.49905	0.10499
25.947	10.61108	0.106111
26.28012	10.73112	0.107311
27.21732	11.07524	0.110752
27.4554	11.18722	0.111872
27.54936	11.29931	0.112993
28.3878	11.75537	0.117554
28.54524	12.87549	0.118755
28.7196	13.08747	0.119875
28.88808	13.89956	0.120996
29.5932	14.88154	0.122115

Data for Fig. 4-3 PNVF 20x1

Table B.6.2 Stress-strain data for poly (N-vinyl formamide) gel. (20x1,ii)

Length (mm)	16.51	
Width (mm)	1.63	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.797814	0.467663	0.013965
1.275956	1.015709	0.030166
2.18701	1.527223	0.045131
3.061825	2.038726	0.059948
3.952306	2.550234	0.07462
4.854334	3.098286	0.090183
5.788594	3.609789	0.104564
6.710452	4.124949	0.118911
7.619701	4.669343	0.133924
8.616383	5.184509	0.147995
9.546827	5.696011	0.161837
10.54861	6.207543	0.175554
11.56985	6.7592	0.190209
12.65666	7.270743	0.203672
13.70124	7.782229	0.217016
14.83766	8.326629	0.23109
15.98693	8.841771	0.244289
17.05491	9.353314	0.257283
18.16255	9.8648	0.270166
19.29892	10.41286	0.283851
20.48225	10.92434	0.296513
21.64525	11.43954	0.309163
22.87773	11.98394	0.322417
24.04903	12.49909	0.334855
24.76683	13.01057	0.347106
24.9917	13.52577	0.359348
27.45749	14.07383	0.372266
28.66487	14.58897	0.384312
29.54098	15.04046	0.39618

Data for PNVF 20x1(ii)

Table B.6.3 Stress-strain data for poly (N-vinyl formamide) gel. (20x1,iii)

Length (mm)	16.43	
Width (mm)	1.93	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.928781	0.487144	0.014544
2.27307	0.986786	0.029315
4.043397	1.55514	0.045943
5.766984	2.054773	0.06041
7.515302	2.61062	0.076343
9.242889	3.116515	0.090698
10.94835	3.678606	0.106489
12.76356	4.240697	0.122116
14.46949	4.740339	0.135871
16.18289	5.308683	0.151367
17.88041	5.808325	0.164859
19.57781	6.370426	0.179896
19.85813	6.870068	0.193137
20.36508	7.438403	0.208059
20.47206	7.994259	0.222513
20.77238	8.493901	0.235388
21.00984	9.062236	0.249903
21.42048	9.561878	0.26255
21.49222	10.12401	0.276654
21.7454	10.62362	0.289082
23.30571	11.18567	0.302943
23.65698	11.74153	0.316529
23.9281	12.24124	0.328642
24.61286	12.80958	0.342303
24.73698	13.3092	0.354214
28.26984	13.87125	0.367504
29.436	14.37096	0.379225
29.77079	14.92301	0.392305

Data for PNVF 20x1(iii)

Table B.7.1 Stress-strain data for poly (N-vinyl formamide) gel. (15x1)

Length (mm)	16.55	
Width (mm)	1.72	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.581425	0.713066	0.021241
1.682814	1.434618	0.042433
2.76302	2.173158	0.063818
3.903397	2.894737	0.084422
5.014844	3.633276	0.105222
6.142957	4.354842	0.125269
7.318001	5.084882	0.145284
8.523406	5.814934	0.165037
9.709842	6.5365	0.18431
10.98237	7.266539	0.203562
12.29656	7.988118	0.222354
13.58168	8.718171	0.241132
14.82058	9.448211	0.259682
16.19841	10.17826	0.278008
17.57804	10.90832	0.296118
18.92708	11.62987	0.31381
20.29988	12.35992	0.331504
21.68841	13.0815	0.348797
23.10241	13.81158	0.366098
24.50438	14.53316	0.383011
25.81043	15.27171	0.400134
26.95483	15.99316	0.416681
28.19999	16.71474	0.433059

Data for Fig. 4-3 PNVF 15x1

Table B.7.2 Stress-strain data for poly (N-vinyl formamide) gel. (15x1,ii)

Length (mm)	16.23	
Width (mm)	1.76	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.449498	0.695373	0.002085
1.62124	1.424645	0.004268
2.68936	2.15393	0.006448
3.84447	2.883215	0.008625
4.92868	3.6125	0.010799
5.96664	4.333294	0.012944
7.12189	5.062579	0.015111
8.35081	5.783373	0.017251
9.47598	6.512671	0.019412
10.7957	7.241956	0.02157
12.0341	7.971241	0.023725
13.3828	8.692035	0.025852
14.7786	9.42132	0.028001
16.0286	10.14211	0.030122
17.501	10.8714	0.032265
18.8469	11.59222	0.03438
20.1653	12.32149	0.036516
21.6181	13.05078	0.03865
23.1095	13.77156	0.040756
24.6018	14.50092	0.042884
26.0411	15.23015	0.045008
27.0592	15.95938	0.04713
27.3776	16.79021	0.049224

Data for PNVF 15x1(ii)

Table B.7.3 Stress-strain data for poly (N-vinyl formamide) gel. (15x1,iii)

Length (mm)	17.03	
Width (mm)	1.83	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.717347	0.674431	0.020098
1.691183	1.399757	0.041416
2.845527	2.120848	0.062313
4.111357	2.884352	0.084127
5.452751	3.609678	0.104561
6.783188	4.330769	0.124605
8.218663	5.051854	0.144384
9.698406	5.772945	0.163908
11.22648	6.49403	0.183182
12.79316	7.215122	0.202214
14.42267	7.978632	0.222108
16.05568	8.703945	0.240769
17.64195	9.425049	0.259097
19.23239	10.14615	0.277207
20.8273	10.86719	0.295104
22.41763	11.59257	0.3129
23.9619	12.31361	0.330388
24.91373	13.07712	0.348692
25.49532	13.79822	0.365784
26.16015	14.51933	0.382689
26.39235	15.24037	0.399411
27.10642	15.96575	0.416055
27.56394	16.91679	0.432427

Data for PNVF 15x1(iii)

Table B.8.1 Stress-strain data for poly (N-vinyl formamide) gel. (10x1)

Length (mm)	16.88	
Width (mm)	1.78	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0.00	0
0.05	0.06	0.00188
1.10	0.75	0.007524
1.98	1.37	0.013697
2.10	1.44	0.014372
2.91	1.99	0.019918
3.01	2.06	0.020593
3.91	2.68	0.078224
4.00	2.75	0.080283
4.97	3.51	0.035062
5.09	3.57	0.035737
5.98	4.20	0.041958
6.06	4.26	0.042633
7.07	5.09	0.05088
7.16	5.16	0.051604
7.92	5.78	0.057777
8.02	5.85	0.0585
8.98	6.67	0.066748
9.04	6.74	0.067422
9.96	7.57	0.075718
10.04	7.64	0.076393
10.89	8.40	0.232886
11.00	8.46	0.234621
12.95	10.47	0.104655
13.02	10.53	0.105329
13.99	11.57	0.115651
14.05	11.63	0.116326
14.97	12.66	0.126646
15.01	12.74	0.12737
16.45	14.72	0.145938

Data for Fig. 4-3 PNVF 10x1

Table B.8.2 Stress-strain data for poly (N-vinyl formamide) gel. (10x1,ii)

Length (mm)	16.72	
Width (mm)	1.93	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.296667	0.511255	0.005113
0.936392	1.028594	0.010286
1.572393	1.552038	0.01552
2.207359	2.069387	0.020694
2.861044	2.598906	0.025989
3.481375	3.116245	0.031162
4.126965	3.639679	0.036397
4.798721	4.163113	0.041631
5.445857	4.680453	0.046805
6.095284	5.203896	0.052039
6.764775	5.721236	0.057212
7.437197	6.250764	0.062508
8.120943	6.768104	0.067681
8.771703	7.291538	0.072915
9.449774	7.814972	0.07815
10.13309	8.332311	0.083323
10.82297	8.855755	0.088558
11.49374	9.373094	0.093731
12.15665	9.902642	0.099026
12.81932	10.42	0.1042
13.47354	10.9434	0.109434
14.10919	11.46679	0.114668
14.7361	11.98415	0.119842
14.95334	12.50151	0.125015
15.25388	13.02491	0.130249
15.36832	13.5484	0.135484
15.87218	14.06575	0.140658
15.98505	14.58915	0.145892
16.70203	14.82651	0.151065

Data for PNVF 10x1(ii)

Table B.8.3 Stress-strain data for poly (N-vinyl formamide) gel. (10x1,iii)

Length (mm)	17.12	
Width (mm)	1.88	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.254726	0.521087	0.015552
0.821287	1.048375	0.031126
1.348618	1.581885	0.046721
1.893114	2.115385	0.062156
2.461394	2.648885	0.077434
3.015852	3.176173	0.092382
3.583668	3.715875	0.10753
4.171076	4.243173	0.122184
4.764176	4.770462	0.136696
5.376843	5.303971	0.151239
5.986806	5.837471	0.165643
6.579247	6.370962	0.17991
7.194431	6.89826	0.19388
7.821633	7.437962	0.208047
8.469837	7.96525	0.221762
9.115925	8.492548	0.235353
9.735248	9.03225	0.24914
10.35935	9.559548	0.262491
10.98772	10.09308	0.275881
11.61848	10.62038	0.289001
12.25339	11.15385	0.302161
12.89856	11.68731	0.315209
13.52894	12.20846	0.32785
14.1504	12.74817	0.340833
14.81432	13.28163	0.353559
15.44881	13.80894	0.366036
16.07554	14.33625	0.378414
16.35464	15.02196	0.390982

Data for PNVF 10x1(iii)

Table B.9.0-Standard Deviations.

PNVF	n=3			
xlink 1	**Swelling**	**E(kPa)**	**Frac. Stress(kPa)**	**Frac. Strain-%**
10	30.4	125.3	16.5	14.72
	30.85	125.7	16.7	14.83
	30.45	125.4	16.35	15.02
	30.566±0.2	**138.13±3.07**	**16.5±0.17**	**14.85±0.15**
15	25.35	171	28.2	16.68
	25.55	174.02	27.3	16.79
	25.48	172.11	27.56	16.91
	25.46±0.1	**172.38±1.53**	**27.6±0.4**	**16.8±0.11**
20	21.72	283.04	29.59	14.88
	21.45	284.11	29.54	15.04
	21.68	281.08	29.77	14.92
	21.62±0.1	**282.74±1.53**	**29.6±0.12**	**14.95±0.08**
25	15.46	518.06	53.4	10.66
	15.15	520.7	53.66	10.99
	15.38	523.02	53.45	11.12
	15.33±0.16	**520.6±2.5**	**53.5±0.13**	**10.9±0.2**
xlink 3	**Swelling**	**E(kPa)**	**Frac. Stress(kPa)**	**Frac. Strain-%**
10	27.31	220.3	26.75	15.33
	27.54	218.7	26.63	15.82
	28.1	221.5	26.99	15.63
	27.65±0.4	**220.2±1**	**26.8±0.18**	**15.6±0.2**
15	22.01	140.5	32.72	22.21
	22.7	144.3	32.91	23.57
	22.7	142.71	32.76	21.19
	22.7±0.4	**142.5±2.0**	**32.8±0.1**	**22.4±1.19**
20	18.54	232.9	42.82	20.79
	18.75	229.2	42.93	19
	18.67	235.1	42.88	21.01
	18.65±0.1	**232.4±3.1**	**42.9±0.05**	**20.03±1.1**
25	14.02	610.22	65.43	10.91
	16.33	607.32	65.63	11.56
	14.95	611.92	65.47	10.78
	15.1±1	**609.82±2.33**	**65.5±0.1**	**11.08±0.4**

Appendix C

STRESS-STRAIN PLOTS OF PAAm GELS: EXPERIMENTAL DATA

Table C.1.1 Stress-strain data for polyacrylamide gel. (10x1)

Length (mm)	18.12	
Width (mm)	1.92	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.217834	0.405248	0.012108
0.432396	0.810496	0.02412
1.084546	1.266557	0.037523
1.666255	1.671805	0.049334
2.030168	2.127615	0.062508
2.4255	2.532864	0.074124
3.299072	2.988924	0.087091
3.673628	3.444734	0.099939
4.146968	3.850233	0.111278
4.585616	4.306043	0.123922
5.215364	4.711291	0.135075
5.662048	5.217914	0.148903
6.403908	5.5726	0.158509
6.950748	6.02841	0.170765
7.589904	6.484471	0.182928

8.146152	6.889719	0.193655
8.808828	7.396342	0.20696
9.283344	7.750778	0.216198
9.6383	8.206838	0.228004
10.58047	8.612086	0.238418
11.20885	9.118709	0.251337
12.19002	9.523957	0.261593
12.63004	9.929206	0.271781
13.82192	10.38527	0.283165
14.68334	10.79051	0.293211
15.2731	11.29714	0.305677
16.29936	11.65157	0.314339
16.83522	12.10763	0.325412
18.1792	12.56344	0.336401
18.71388	12.96869	0.346106
19.91948	13.47531	0.358153
20.64272	13.83	0.366532

Data for Fig. 4-4 PAAm 10x1

Table C.1.2 Stress-strain data for polyacrylamide gel. (10x1,ii)

Length (mm)	17.92	
Width (mm)	1.67	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.217834	0.405248	0.009209
0.432396	0.810496	0.018345
1.084546	1.266557	0.036469
1.666255	1.671805	0.045457
2.030168	2.127615	0.054392
2.4255	2.532864	0.072088
3.299072	2.988924	0.080866
3.673628	3.444734	0.089578
4.146968	3.850233	0.106867
4.585616	4.306043	0.123974
5.215364	4.711291	0.13244
5.662048	5.217914	0.149246
6.403908	5.5726	0.157587
6.950748	6.02841	0.165882
7.589904	6.484471	0.182321
8.146152	6.889719	0.198584
8.808828	7.396342	0.206658

9.283344	7.750778	0.214674
9.6383	8.206838	0.23061
10.58047	8.612086	0.238501
11.20885	9.118709	0.246365
12.19002	9.523957	0.254176
12.63004	9.929206	0.269705
13.82192	10.38527	0.277397
14.68334	10.79051	0.292679
15.2731	11.29714	0.300271
16.29936	11.65157	0.315328
16.83522	12.10763	0.322808
18.1792	12.56344	0.337648
18.71388	12.96869	0.345021
19.91948	13.47531	0.35965
20.64272	13.83	0.36692

Data for PAAm 10x1(ii)

Table C.1.3 Stress-strain data for polyacrylamide gel. (10x1,iii)

Length (mm)	16.82	
Width (mm)	1.72	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.360784	0.455403	0.0136
1.13877	0.916227	0.027238
1.912225	1.382487	0.040912
2.684424	1.843319	0.054305
3.479386	2.314992	0.06789
4.233785	2.775815	0.081046
5.018903	3.242067	0.09424
5.835842	3.708319	0.107319
6.62284	4.169143	0.120135
7.412624	4.635403	0.132993
8.226808	5.096227	0.145593
9.044557	5.567908	0.158382
9.876078	6.028731	0.170773
10.66748	6.494983	0.183207
11.4921	6.961235	0.19554
12.3231	7.422059	0.207632
13.16208	7.888319	0.219769
13.97782	8.349143	0.231669

14.784	8.82084	0.243755
15.58989	9.281681	0.25547
16.3855	9.747899	0.267232
17.15853	10.21412	0.278903
17.92094	10.67496	0.290353
18.18513	11.1358	0.301718
18.55061	11.60202	0.31313
18.86608	12.06832	0.324461
19.19332	12.52916	0.335578
19.51408	12.99538	0.346743
20.19133	13.45622	0.357701
21.00509	13.92244	0.368709

Data for PAAm 10x1(iii)

Table C.2.1 Stress-strain data for polyacrylamide gel. (20x1)

Length (mm)	17.25	
Width (mm)	1.68	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.62169	0.42	0.012547
1.677434	0.84	0.024991
2.690741	1.260037	0.037333
3.901157	1.718222	0.050681
5.082681	2.138222	0.062813
6.17329	2.596407	0.075938
7.412225	3.016407	0.087868
8.520688	3.474593	0.100777
9.664079	3.894593	0.112513
10.73872	4.31463	0.124159
11.80366	4.772815	0.136761
12.88634	5.192815	0.148221
14.03049	5.651	0.160624
15.09412	6.071	0.171905
16.09855	6.529185	0.184116
17.14998	6.949222	0.195224
18.24035	7.369222	0.20625
19.49276	7.827407	0.218188

20.62936	8.247407	0.22905
21.69665	8.705593	0.240811
22.74718	9.125593	0.251512
23.85983	9.583778	0.263102
24.96114	10.00381	0.273649
26.06246	10.42381	0.284124
27.16377	10.882	0.295469
28.26508	11.302	0.305796
29.36639	11.72204	0.316055
30.19537	12.14204	0.326245
30.67205	12.63837	0.3382
32.33691	13.05841	0.348246
32.65124	13.47841	0.358227
35.1367	13.93659	0.369042

Data for Fig. 4-4 PAAm 20x1

Table C.2.2 Stress-strain data for polyacrylamide gel. (20x1,ii)

Length (mm)	17.85	
Width (mm)	1.84	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.937542	0.475447	0.014196
2.219845	0.994033	0.029528
3.288201	1.46948	0.043449
4.568336	1.988108	0.058488
5.940139	2.463513	0.072143
7.042342	2.938918	0.085674
8.228173	3.414366	0.099086
9.28532	3.932993	0.113581
10.4328	4.408399	0.126747
11.40868	4.927026	0.140979
12.37201	5.402432	0.153908
13.47285	5.921059	0.167887
14.53185	6.396465	0.180589
15.51827	6.871912	0.193185
16.64281	7.390539	0.206808
17.78905	7.865945	0.219188
18.71156	8.384572	0.23258
19.89606	8.9032	0.245855

21.00008	9.378605	0.257923
22.12292	9.854053	0.269897
23.2762	10.32946	0.281777
24.2744	10.84809	0.294632
25.47006	11.32349	0.306323
26.52473	11.84212	0.318975
27.67894	12.31752	0.330482
28.59135	12.83619	0.34294
29.47321	13.3116	0.354271
30.91478	13.787	0.365519
31.29632	14.30563	0.377698
32.58748	14.78104	0.388779
33.36217	15.29971	0.400779
34.29632	15.77507	0.411697

Data for PAAm 20x1(ii)

Table C.2.3 Stress-strain data for polyacrylamide gel. (20x1,iii)

Length (mm)	18.85	
Width (mm)	1.89	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
0.883	0.3567	0.010663
2.07392	0.71277	0.021232
3.20778	1.42554	0.042168
4.29178	1.78224	0.052537
0.00369	2.13894	0.062834
6.60488	2.85171	0.083201
7.74549	3.20842	0.093291
8.88611	3.56449	0.103296
10.0267	4.27726	0.123126
11.1673	4.99066	0.142716
12.3079	5.34673	0.152399
13.4486	6.0595	0.171597
14.5892	6.4162	0.181114
15.7298	6.7729	0.190571
16.8704	7.48567	0.209293
18.011	8.19844	0.227788
19.1516	8.55514	0.236959
20.2922	8.91121	0.246059

21.4328	9.62462	0.26413
22.5734	9.98069	0.27307
23.7141	10.3374	0.281974
24.8547	10.6935	0.290811
25.9953	11.4069	0.308363
27.1359	11.7629	0.31705
28.2765	12.4757	0.334292
29.4171	12.8324	0.342849
30.5577	13.5452	0.359807
31.6983	13.9019	0.368225
32.8389	14.6146	0.38491
33.9796	14.9713	0.393193
35.1202	15.6841	0.409614
36.2608	16.0408	0.417768

Data for PAAm 20x1(iii)

Table C.3.1 Stress-strain data for polyacrylamide gel. (20x3)

Length (mm)	18.51	
Width (mm)	1.65	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.452252	0.77325	0.02302
3.43854	1.616659	0.047732
5.093424	2.389909	0.070037
7.076352	3.233386	0.093995
9.201276	4.006568	0.115626
10.90859	4.77975	0.136951
12.74544	5.553	0.15798
14.38296	6.396477	0.180589
16.1604	7.169659	0.201021
17.67204	8.013136	0.223001
19.16424	8.786318	0.242873
20.86944	9.629795	0.264261
22.50984	10.40298	0.283606
24.0378	11.17623	0.302711
25.77972	12.0197	0.323284
27.55524	12.79289	0.341903
28.9842	13.63636	0.361964
30.819	14.47984	0.381768

32.52912	15.25302	0.399703
34.5968	16.02627	0.417436
35.6896	16.79945	0.43497
37.258	17.64293	0.453879
39.3545	18.41611	0.471015
40.2569	19.25959	0.489502
42.1452	20.03277	0.506262
42.555	21.87632	0.545536

Data for Fig. 4-4 PAAm 20x3

Table C.3.2 Stress-strain data for polyacrylamide gel. (20x3,ii)

Length (mm)	18.57	
Width (mm)	1.73	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.452252	0.873235	0.025971
3.43854	1.825702	0.053795
5.093424	2.698937	0.078859
7.076352	3.651481	0.105731
9.201276	4.524639	0.129948
10.90859	5.397798	0.153782
12.74544	6.271033	0.177248
14.38296	7.223577	0.202436
16.1604	8.096735	0.225162
17.67204	9.049279	0.249573
19.16424	9.922437	0.271611
20.86944	10.87498	0.295296
22.50984	11.74814	0.31669
24.0378	12.62137	0.337792
25.77972	13.57392	0.360487
27.55524	14.44708	0.381003
28.9842	15.39962	0.40308

30.819	16.35216	0.424851
32.52912	17.22532	0.444545
33.12	18.09856	0.463999
32.2565	18.97172	0.483217
34.6587	19.92426	0.503921
36.4587	20.79742	0.522668
39.2565	21.74996	0.542875
40.1125	22.62312	0.56118
41.2808	23.57574	0.58092

Data for PAAm 20x3(ii)

Table C.3.3 Stress-strain data for polyacrylamide gel. (20x3,iii)

Length (mm)	17.07	
Width (mm)	1.82	
Thickness (mm)	2	
Stress (kPa)	**Strain**	**Strain Function (λ-λ^{-2})**
0	0	0
1.452252	0.77325	0.02302
3.43854	1.616659	0.047732
5.093424	2.389909	0.070037
7.076352	3.233386	0.093995
9.201276	4.006568	0.115626
10.908588	4.77975	0.136951
12.74544	5.553	0.15798
14.38296	6.396477	0.180589
16.1604	7.169659	0.201021
17.67204	8.013136	0.223001
19.16424	8.786318	0.242873
20.86944	9.629795	0.264261
22.50984	10.40298	0.283606
24.0378	11.17623	0.302711
25.77972	12.0197	0.323284
27.55524	12.79289	0.341903
28.9842	13.63636	0.361964
30.819	14.47984	0.381768

32.52912	15.25302	0.399703
33.5968	16.02627	0.417436
35.0125	16.79945	0.43497
36.2656	17.64293	0.453879
38.2365	18.41611	0.471015
40.6589	19.25959	0.489502
41.2563	20.03277	0.506262
42.1125	20.87632	0.524351
43.2867	21.6595	0.540967

Data for PAAm 20x3(iii)

Table C.4-PAAm gels Standard Deviations.

Acrylamide				
10x1	28.64	149.23	20.04	13.21
	28.63	148.96	20.55	14.01
	28.49	148.22	22.04	14.12
	28.58±0.08	**149.6±0.5**	**20.97±1.03**	**13.8±0.5**
20x1	20.39	198.55	34.11	13.9
	20.41	201.82	36.29	15.77
	20.48	197.35	35.26	16.01
	20.42±0.4	**199.24±2.31**	**35.22±1.09**	**15.2±1.2**
20x3	18.89	256.11	42.55	21.1
	19.28	264.52	41.28	23.55
	18.73	254.04	43.28	21.66
	18.96±0.28	**258.6±5.6**	**42.37±1.01**	**22.15±0.98**

Appendix D

```
ONEWAY  VAR00002  VAR00003  VAR00004  VAR00005  VAR00006  BY  VAR00001
   /STATISTICS  DESCRIPTIVES
   /MISSING  ANALYSIS
   /POSTHOC=TUKEY  ALPHA(0.05).
```

Oneway

```
Set 1= 10x1 PAAm
Set 2= 20x1 PAAm
Set 3= 20x3 PAAm
Set 4= 10x1 PNVF
Set 5= 20x1 PNVF
Set 6= 20x3 PNVF
```

[DataSet0]

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean	
						Lower Bound	Upper Bound
Swelling	1	3	28.57000	.072111	.041633	28.39087	28.74913
	2	3	20.40667	.015275	.008819	20.36872	20.44461
	3	3	18.99000	.276225	.159478	18.30382	19.67618
	4	3	30.62000	.206640	.119304	30.10668	31.13332
	5	3	21.58333	.119304	.068880	21.28697	21.87970
	6	3	18.69000	.052915	.030551	18.55855	18.82145
	Total	18	23.14333	4.834597	1.139525	20.73914	25.54752
E	1	3	148.95000	.725052	.418609	147.14887	150.75113

		Minimum	Maximum
Swelling	1	28.490	28.630
	2	20.390	20.420
	3	18.730	19.280
	4	30.450	30.850
	5	21.450	21.680
	6	18.650	18.750
	Total	18.650	30.850
E	1	148.220	149.670

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean	
						Lower Bound	Upper Bound
E	2	3	224.07867	.548968	.316947	222.71495	225.44238
	3	3	234.64333	.351615	.203005	233.76987	235.51679
	4	3	125.36667	.351188	.202759	124.49427	126.23907
	5	3	274.06333	.945110	.545660	271.71555	276.41112
	6	3	219.91333	.674191	.389244	218.23855	221.58812
	Total	18	204.50256	52.666667	12.413652	178.31204	230.69307
G	1	3	56.37000	1.974411	1.139927	51.46529	61.27471
	2	3	79.54667	.548847	.316877	78.18325	80.91008
	3	3	86.88667	3.285443	1.896851	78.72517	95.04816
	4	3	52.84000	10.118755	5.842066	27.70362	77.97638
	5	3	92.39333	3.558881	2.054721	83.55258	101.23408
	6	3	73.14667	2.408741	1.390687	67.16302	79.13031
	Total	18	73.53056	15.626233	3.683138	65.75981	81.30130
Stress	1	3	21.18667	.768266	.443559	19.27819	23.09515
	2	3	34.95667	.285715	.164958	34.24691	35.66642
	3	3	42.31000	1.001349	.578129	39.82251	44.79749
	4	3	16.51667	.175594	.101379	16.08047	16.95287
	5	3	29.57667	.032146	.018559	29.49681	29.65652
	6	3	42.88333	.056862	.032830	42.74208	43.02459
	Total	18	31.23833	10.235563	2.412545	26.14831	36.32836
Strain	1	3	13.98667	.146401	.084525	13.62299	14.35035
	2	3	15.29667	.433167	.250089	14.22062	16.37271
	3	3	22.45333	.980833	.566284	20.01681	24.88986
	4	3	14.88333	.119304	.068880	14.58697	15.17970
	5	3	14.95667	.080208	.046308	14.75742	15.15591
	6	3	19.94000	.898387	.518684	17.70828	22.17172
	Total	18	16.91944	3.262436	.768964	15.29707	18.54182

Descriptives

		Minimum	Maximum
E	2	223.508	224.603
	3	234.370	235.040
	4	125.000	125.700
	5	273.110	275.000
	6	219.200	220.540
	Total	125.000	275.000
G	1	54.980	58.630
	2	78.950	80.030
	3	84.660	90.660
	4	46.360	64.500
	5	88.400	95.230
	6	70.900	75.690
	Total	46.360	95.230
Stress	1	20.550	22.040
	2	34.660	35.230
	3	41.280	43.280
	4	16.350	16.700
	5	29.540	29.600
	6	42.820	42.930
	Total	16.350	43.280
Strain	1	13.830	14.120
	2	14.920	15.770
	3	21.660	23.550
	4	14.800	15.020
	5	14.880	15.040
	6	19.000	20.790
	Total	13.830	23.550

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Swelling	Between Groups	397.064	5	79.413	3368.118	.000
	Within Groups	.283	12	.024		
	Total	397.347	17			
E	Between Groups	47149.379	5	9429.876	23362.484	.000
	Within Groups	4.844	12	.404		
	Total	47154.223	17			
G	Between Groups	3879.344	5	775.869	34.267	.000
	Within Groups	271.701	12	22.642		

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
G	Total	4151.045	17			
Stress	Between Groups	1777.615	5	355.523	1247.693	.000
	Within Groups	3.419	12	.285		
	Total	1781.035	17			
Strain	Between Groups	176.942	5	35.388	106.225	.000
	Within Groups	3.998	12	.333		
	Total	180.939	17			

Post Hoc Tests

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Swelling	1	2	8.163333*	.125374	.000	7.74221	8.58445
		3	9.580000*	.125374	.000	9.15888	10.00112
		4	-2.050000*	.125374	.000	-2.47112	-1.62888
		5	6.986667*	.125374	.000	6.56555	7.40779
		6	9.880000*	.125374	.000	9.45888	10.30112
	2	1	-8.163333*	.125374	.000	-8.58445	-7.74221
		3	1.416667*	.125374	.000	.99555	1.83779
		4	-10.213333*	.125374	.000	-10.63445	-9.79221
		5	-1.176667*	.125374	.000	-1.59779	-.75555
		6	1.716667*	.125374	.000	1.29555	2.13779
	3	1	-9.580000*	.125374	.000	-10.00112	-9.15888
		2	-1.416667*	.125374	.000	-1.83779	-.99555
		4	-11.630000*	.125374	.000	-12.05112	-11.20888
		5	-2.593333*	.125374	.000	-3.01445	-2.17221
		6	.300000	.125374	.232	-.12112	.72112
	4	1	2.050000*	.125374	.000	1.62888	2.47112
		2	10.213333*	.125374	.000	9.79221	10.63445
		3	11.630000*	.125374	.000	11.20888	12.05112
		5	9.036667*	.125374	.000	8.61555	9.45779
		6	11.930000*	.125374	.000	11.50888	12.35112
	5	1	-6.986667*	.125374	.000	-7.40779	-6.56555
		2	1.176667*	.125374	.000	.75555	1.59779

*. The mean difference is significant at the 0.05 level.

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Swelling	5	3	2.593333*	.125374	.000	2.17221	3.01445
		4	-9.036667*	.125374	.000	-9.45779	-8.61555
		6	2.893333*	.125374	.000	2.47221	3.31445
	6	1	-9.880000*	.125374	.000	-10.30112	-9.45888
		2	-1.716667*	.125374	.000	-2.13779	-1.29555
		3	-.300000	.125374	.232	-.72112	.12112
		4	-11.930000*	.125374	.000	-12.35112	-11.50888
		5	-2.893333*	.125374	.000	-3.31445	-2.47221
E	1	2	-75.128667*	.518738	.000	-76.87107	-73.38627
		3	-85.693333*	.518738	.000	-87.43573	-83.95093
		4	23.583333*	.518738	.000	21.84093	25.32573
		5	-125.113333*	.518738	.000	-126.85573	-123.37093
		6	-70.963333*	.518738	.000	-72.70573	-69.22093
	2	1	75.128667*	.518738	.000	73.38627	76.87107
		3	-10.564667*	.518738	.000	-12.30707	-8.82227
		4	98.712000*	.518738	.000	96.96960	100.45440
		5	-49.984667*	.518738	.000	-51.72707	-48.24227
		6	4.165333*	.518738	.000	2.42293	5.90773
	3	1	85.693333*	.518738	.000	83.95093	87.43573
		2	10.564667*	.518738	.000	8.82227	12.30707
		4	109.276667*	.518738	.000	107.53427	111.01907
		5	-39.420000*	.518738	.000	-41.16240	-37.67760
		6	14.730000*	.518738	.000	12.98760	16.47240
	4	1	-23.583333*	.518738	.000	-25.32573	-21.84093
		2	-98.712000*	.518738	.000	-100.45440	-96.96960
		3	-109.276667*	.518738	.000	-111.01907	-107.53427
		5	-148.696667*	.518738	.000	-150.43907	-146.95427
		6	-94.546667*	.518738	.000	-96.28907	-92.80427
	5	1	125.113333*	.518738	.000	123.37093	126.85573
		2	49.984667*	.518738	.000	48.24227	51.72707
		3	39.420000*	.518738	.000	37.67760	41.16240
		4	148.696667*	.518738	.000	146.95427	150.43907
		6	54.150000*	.518738	.000	52.40760	55.89240
	6	1	70.963333*	.518738	.000	69.22093	72.70573
		2	-4.165333*	.518738	.000	-5.90773	-2.42293
		3	-14.730000*	.518738	.000	-16.47240	-12.98760
		4	94.546667*	.518738	.000	92.80427	96.28907
		5	-54.150000*	.518738	.000	-55.89240	-52.40760

*. The mean difference is significant at the 0.05 level.

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
G	1	2	-23.176667*	3.885165	.001	-36.22663	-10.12671
		3	-30.516667*	3.885165	.000	-43.56663	-17.46671
		4	3.530000	3.885165	.937	-9.51996	16.57996
		5	-36.023333*	3.885165	.000	-49.07329	-22.97337
		6	-16.776667*	3.885165	.010	-29.82663	-3.72671
	2	1	23.176667*	3.885165	.001	10.12671	36.22663
		3	-7.340000	3.885165	.452	-20.38996	5.70996
		4	26.706667*	3.885165	.000	13.65671	39.75663
		5	-12.846667	3.885165	.055	-25.89663	.20329
		6	6.400000	3.885165	.586	-6.64996	19.44996
	3	1	30.516667*	3.885165	.000	17.46671	43.56663
		2	7.340000	3.885165	.452	-5.70996	20.38996
		4	34.046667*	3.885165	.000	20.99671	47.09663
		5	-5.506667	3.885165	.717	-18.55663	7.54329
		6	13.740000*	3.885165	.037	.69004	26.78996
	4	1	-3.530000	3.885165	.937	-16.57996	9.51996
		2	-26.706667*	3.885165	.000	-39.75663	-13.65671
		3	-34.046667*	3.885165	.000	-47.09663	-20.99671
		5	-39.553333*	3.885165	.000	-52.60329	-26.50337
		6	-20.306667*	3.885165	.002	-33.35663	-7.25671
	5	1	36.023333*	3.885165	.000	22.97337	49.07329
		2	12.846667	3.885165	.055	-.20329	25.89663
		3	5.506667	3.885165	.717	-7.54329	18.55663
		4	39.553333*	3.885165	.000	26.50337	52.60329
		6	19.246667*	3.885165	.003	6.19671	32.29663
	6	1	16.776667*	3.885165	.010	3.72671	29.82663
		2	-6.400000	3.885165	.586	-19.44996	6.64996
		3	-13.740000*	3.885165	.037	-26.78996	-.69004
		4	20.306667*	3.885165	.002	7.25671	33.35663
		5	-19.246667*	3.885165	.003	-32.29663	-6.19671
Stress	1	2	-13.770000*	.435847	.000	-15.23398	-12.30602
		3	-21.123333*	.435847	.000	-22.58731	-19.65936
		4	4.670000*	.435847	.000	3.20602	6.13398
		5	-8.390000*	.435847	.000	-9.85398	-6.92602
		6	-21.696667*	.435847	.000	-23.16064	-20.23269
	2	1	13.770000*	.435847	.000	12.30602	15.23398
		3	-7.353333*	.435847	.000	-8.81731	-5.88936
		4	18.440000*	.435847	.000	16.97602	19.90398

*. The mean difference is significant at the 0.05 level.

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
Stress	2	5	5.380000*	.435847	.000	3.91602	6.84398
		6	-7.926667*	.435847	.000	-9.39064	-6.46269
	3	1	21.123333*	.435847	.000	19.65936	22.58731
		2	7.353333*	.435847	.000	5.88936	8.81731
		4	25.793333*	.435847	.000	24.32936	27.25731
		5	12.733333*	.435847	.000	11.26936	14.19731
		6	-.573333	.435847	.772	-2.03731	.89064
	4	1	-4.670000*	.435847	.000	-6.13398	-3.20602
		2	-18.440000*	.435847	.000	-19.90398	-16.97602
		3	-25.793333*	.435847	.000	-27.25731	-24.32936
		5	-13.060000*	.435847	.000	-14.52398	-11.59602
		6	-26.366667*	.435847	.000	-27.83064	-24.90269
	5	1	8.390000*	.435847	.000	6.92602	9.85398
		2	-5.380000*	.435847	.000	-6.84398	-3.91602
		3	-12.733333*	.435847	.000	-14.19731	-11.26936
		4	13.060000*	.435847	.000	11.59602	14.52398
		6	-13.306667*	.435847	.000	-14.77064	-11.84269
	6	1	21.696667*	.435847	.000	20.23269	23.16064
		2	7.926667*	.435847	.000	6.46269	9.39064
		3	.573333	.435847	.772	-.89064	2.03731
		4	26.366667*	.435847	.000	24.90269	27.83064
		5	13.306667*	.435847	.000	11.84269	14.77064
Strain	1	2	-1.310000	.471271	.129	-2.89296	.27296
		3	-8.466667*	.471271	.000	-10.04963	-6.88370
		4	-.896667	.471271	.445	-2.47963	.68630
		5	-.970000	.471271	.367	-2.55296	.61296
		6	-5.953333*	.471271	.000	-7.53630	-4.37037
	2	1	1.310000	.471271	.129	-.27296	2.89296
		3	-7.156667*	.471271	.000	-8.73963	-5.57370
		4	.413333	.471271	.945	-1.16963	1.99630
		5	.340000	.471271	.975	-1.24296	1.92296
		6	-4.643333*	.471271	.000	-6.22630	-3.06037
	3	1	8.466667*	.471271	.000	6.88370	10.04963
		2	7.156667*	.471271	.000	5.57370	8.73963
		4	7.570000*	.471271	.000	5.98704	9.15296
		5	7.496667*	.471271	.000	5.91370	9.07963
		6	2.513333*	.471271	.002	.93037	4.09630
	4	1	.896667	.471271	.445	-.68630	2.47963

*. The mean difference is significant at the 0.05 level.

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
Strain	4	2	-.413333	.471271	.945	-1.99630	1.16963
		3	-7.570000*	.471271	.000	-9.15296	-5.98704
		5	-.073333	.471271	1.000	-1.65630	1.50963
		6	-5.056667*	.471271	.000	-6.63963	-3.47370
	5	1	.970000	.471271	.367	-.61296	2.55296
		2	-.340000	.471271	.975	-1.92296	1.24296
		3	-7.496667*	.471271	.000	-9.07963	-5.91370
		4	.073333	.471271	1.000	-1.50963	1.65630
		6	-4.983333*	.471271	.000	-6.56630	-3.40037
	6	1	5.953333*	.471271	.000	4.37037	7.53630
		2	4.643333*	.471271	.000	3.06037	6.22630
		3	-2.513333*	.471271	.002	-4.09630	-.93037
		4	5.056667*	.471271	.000	3.47370	6.63963
		5	4.983333*	.471271	.000	3.40037	6.56630

*. The mean difference is significant at the 0.05 level.

Homogeneous Subsets

Swelling

Tukey HSD

Group	N	Subset for alpha = 0.05				
		1	2	3	4	5
6	3	18.69000				
3	3	18.99000				
2	3		20.40667			
5	3			21.58333		
1	3				28.57000	
4	3					30.62000
Sig.		.232	1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

E

Tukey HSD

Group	N	Subset for alpha = 0.05					
		1	2	3	4	5	6
4	3	125.36667					
1	3		148.95000				
6	3			219.91333			
2	3				224.07867		
3	3					234.64333	
5	3						274.06333
Sig.		1.000	1.000	1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

G

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
4	3	52.84000		
1	3	56.37000		
6	3		73.14667	
2	3		79.54667	79.54667
3	3			86.88667
5	3			92.39333
Sig.		.937	.586	.055

Means for groups in homogeneous subsets are displayed.

Stress

Tukey HSD

Group	N	Subset for alpha = 0.05				
		1	2	3	4	5
4	3	16.51667				
1	3		21.18667			
5	3			29.57667		
2	3				34.95667	
3	3					42.31000
6	3					42.88333
Sig.		1.000	1.000	1.000	1.000	.772

Means for groups in homogeneous subsets are displayed.

Strain

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
1	3	13.98667		
4	3	14.88333		
5	3	14.95667		
2	3	15.29667		
6	3		19.94000	
3	3			22.45333
Sig.		.129	1.000	1.000

Means for groups in homogeneous subsets are displayed.