Proc. Indian Acad. Sci. (Chem. Sci.), Vol. 109, No. 1, February 1997, pp. 39–48.

A three-dimensional phase boundary model for diffusion processes involved in reactions of crosslinked polymeric amines with low molecular weight esters

DESY P KORUTHU, P M MADHUSUDANAN and
V N RAJASEKHARAN PILLAI*
School of Chemical Sciences, Mahatma Gandhi University, Kottayam 686 560, India

MS received 4 March 1994; revised 8 November 1996

Abstract. The reactivity of amino groups attached to N–N′–methylene-*bis*-acrylamide (NNMBA)-and triethyleneglycol dimethacrylate (TEGDMA)-crosslinked polyacrylamide gels (2–20 mol% crosslinked) has been investigated by following the aminolysis of *p*-nitrophenyl ester of benzoyl glycine in a mixed solvent. A differential method was employed for following the rate of aminolysis reaction at definite intervals of time. The extent of aminolysis was found to be maximum between 25 and 35 min after initiation for these resins. The rate constants were calculated using the equation for three-dimensional phase boundary model with spherical symmetry and three-dimensional diffusion model. The reaction appears to be a phase boundary process up to 40 min after initiation and thereafter is fully controlled by three-dimensional diffusion. The rate constants do not show any dispersion when this model is used.

Keywords. Crosslinked polyacrylamide support; aminolysis of polymeric esters; phase boundary process; three-dimensional diffusion.

1. Introduction

The use of crosslinked polymer supports for the stepwise synthesis of polypeptides was first reported by Merrifield (Merrifield 1963; Marshall and Merrifield 1971; Atherton and Sheppard 1977). Sheppard (1971) suggested the use of polar matrices as supports for peptide synthesis, as polymeric reagents and for the collection of metal ions (Atherton *et al* 1981; George and Pillai 1988). It was observed that reactivity of a functional group attached to a polymer remains more or less unaltered (Flory 1939; Morawetz 1988). In a heterogeneous reaction involving a crosslinked polymer supported reagent and a low molecular weight substrate, the substrate in the solution phase may be attracted to the functionalised polymer or repelled by it. This is observed when the polymer-bound reagent is activated or inhibited by a neighbouring group or when the local polarity of the bulk solvent is changed (Morawetz 1979). It has also been reported that in the kinetics, there is dispersion in rate constants (Morawetz 1979). When the solvent is poor, free molecules diffuse slowly into the interior of gel-type and macroporous resins by a molecular process involving motion of the polymer chains (Guyot and Bartholin 1982).

*For correspondence

This paper describes the effect of the relative polarity and chemical nature of the crosslinking agent on the kinetics and mechanism of the aminolysis of *p*-nitro phenyl ester of benzoyl glycine with different amino functionalised crosslinked polyacrylamides (Bodanszky *et al* 1979; Devaky *et al* 1990). Though the overall structures and properties of the matrices are nonuniform, an attempt is made here to formulate a mathematical mode describing the reactions involving functional groups in crosslinked polymer matrices.

2. Experimental

2.1 *General*

All the reagents were of certified ACS reagent grade.

2.2 *Preparation of crosslinked polyacrylamides*

Ammonium per sulphate (100 mg) was dissolved in water (70 ml) at 70 °C. Acrylamide and N,N′-methylene-*bis*-acrylamide were added to this with stirring. Heating and stirring was continued till the polymer precipitated. Water (50 ml) was then added and the contents were heated at 80 °C for 20 min. The polymer was filtered, washed and dried. Acrylamide–TEGDMA copolymers were prepared by solution polymerisation of monomers from ethanolic solution at 70 °C. The compositions of the monomer feeds used for preparation of different crosslinked polyacrylamide resins are given in table 1.

2.3 *Preparation of amino polyacrylamides*

2.3a *General procedure*: Ethylene diamine (100 ml) was added gradually to crosslinked polyacrylamide (10 g) taken in a round-bottomed flask and the mixture was heated at 90°C for 9 h. The reaction mixture was then poured into water (500 ml) containing crushed ice. The resin was filtered and washed with sodium chloride (0.1 M) solution until the filtrate was free from ethylene diamine. The gel was washed with deionised water till free from chloride ions. It was finally washed with methanol, filtered and dried.

2.4 *Preparation of p-nitrophenyl ester of benzoyl glycine*

Benzoyl glycine (1·79 g, 10 mmol) was dissolved in tetrahydrofuran (20 ml). Dicyclohexyl carbodiimide (2·06 g, 10 mmol) and *p*-nitrophenol (1·39 g, 10 mmol) were

Table 1. Preparation of crosslinked poly(acrylamide)s.

	Weight of monomers			
Crosslinking agent	Extent of crosslinking (mol%)	Acrylamide (g)	Crosslinking agent (g)	Yield (g)
NNMBA	2	14·08	0·61	11
	8	13·10	2·50	12·5
	20	11·50	6·17	12
TEGDMA	5	13·5	2·6	14·3
	10	13	5·2	12
	20	12	10·5	17·5

added to this. The mixture was kept at 0 °C and stirred for 1 h. The precipitated dicyclohexyl urea was filtered. The filtrate was concentrated and the residue was washed with ethanol. The residue was dissolved in dichloromethane and recrystallised from petroleum ether, and dried. Yield: 1.5 g.

2.5 *Kinetics of aminolysis of p-nitrophenyl ester of benzoyl glycine by the polymeric amines*

A standard curve was obtained from a plot of concentration vs absorbance at various concentrations of *p*-nitrophenol solutions. The absorbance of the solutions was observed by UV-Vis spectroscopy. From this, concentrations of the *p*-nitrophenol liberated at different time intervals were obtained.

2.6 *General procedure for kinetics*

p-Nitrophenyl ester of benzoyl glycine (50 mg, 0·16 mmol) was dissolved in 1:1 dioxane–water mixture (50 ml). The amino resins derived from the crosslinked polyacrylamides (0·308 mmol) were added to this solution and the contents were stirred. One millilitre of the reaction mixture was withdrawn at intervals of 10 min up to 80 min. It was diluted to 10 ml with 1:1 dioxane–water mixture. The concentrations of *p*-nitrophenol in these solutions were measured by recording the UV-Vis spectra. The reaction was also carried out using swollen wet resin, where the resin was first allowed to swell for 48 h. Aminolysis was carried out as in the case of the dry resin.

3. Results and discussion

3.1 *Preparation of crosslinked polyacrylamides*

Crosslinked polyacrylamides of varying mol% of crosslinking agents were prepared using N,N′-methylene-*bis*-acrylamide and triethyleneglycol dimethacrylate (scheme 1).

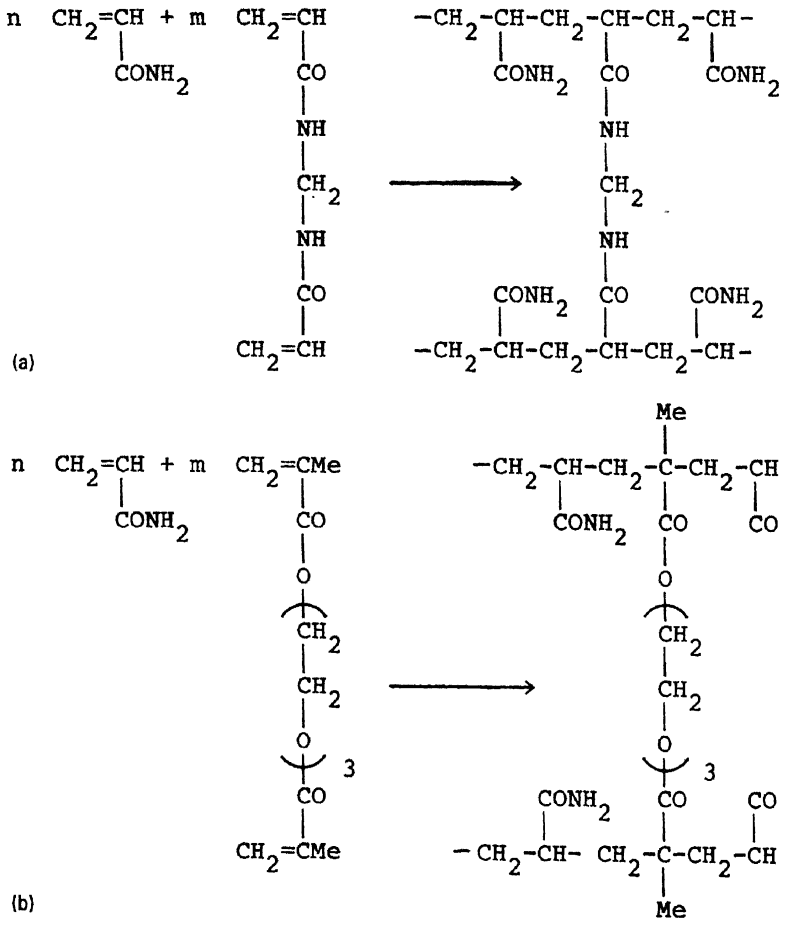
3.2 *Preparation of poly(N-2-aminoethyl acrylamide)s*

Different crosslinked polymers were functionalised to poly(N-2-amino acrylamide)s by treating with excess ethylene diamine (scheme 2). The resulting resins developed a blue colour on heating with ninhydrin reagent indicating the presence of amino groups (Spackman *et al* 1958).

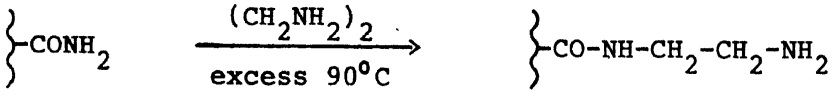
Capacities of the amino groups in these different crosslinked resins were determined by treating with excess hydrochloric acid and back titration of the excess acid. Capacities varied depending on the nature and extent of crosslinking in the copolymer (table 2). In the case of resins containing NNMBA crosslinks the capacity varied from 3·50 to 1·06 mmol/g for 2 to 20% crosslinked polymer. In triethyleneglycol dimethacrylate resins the amino capacity varied from 3·88 to 3·19 mmol/g for the 5 to 20% crosslinked resins.

3.3 *Kinetics of aminolysis of p-nitrophenyl ester of benzoyl glycine by polymeric amines*

The reactivity of amino groups in different crosslinked polyacrylamides was investigated by following the aminolysis of the *p*-nitrophenyl ester of benzoyl glycine in a hydrophilic–hydrophobic solvent mixture (scheme 3). The concentrations of resin and ester were kept constant. The reaction was followed by measuring the concentration of the liberated *p*-nitrophenol at different intervals by UV-Vis spectroscopy.



Scheme 1. Preparation of **(a)** NNMBA-crosslinked, and **(b)** TEGDMA-crosslinked polyacrylamides.



Scheme 2. Transamidation of crosslinked polyacrylamides with ethylenediamine.

Table 2. Rate constants of aminolysis of *p*-nitrophenyl ester of benzoylglycine with crosslinked poly (N-2-amino ethyl acrylamide) resins.

Cross-linking agent	Cross-linking (mol%)	Capacity (mmol/g)	Rate constant ($\times 10^{-3}$) (min^{-1})
NNMBA	2	3·51	1·3
	8	1·98	1·4
	20	1·06	1·9
TEGDMA	5	3·88	0·17
	10	3·23	3·51
	20	3·19	3·63

The reaction was carried out for NNMBA and TEGDMA-crosslinked resins with varying mol% of crosslinking.

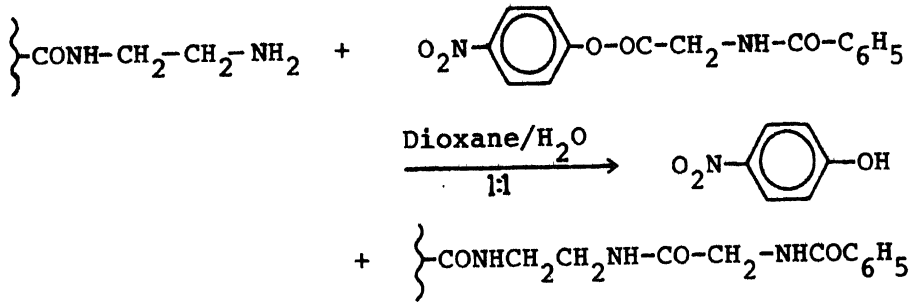
Concentration–time curves for the aminolysis of *p*-nitrophenyl ester of benzoyl glycine with N-N′-methylene-*bis*-acrylamide and triethyleneglycol dimethacrylate crosslinked acrylamide resins are given in figures 1a and 1b. A differential method was employed for calculating the slope of the concentration–time curve. Cubic spline method (de Boor 1978) is generally used to determine the slope of a curve at desired points from the experimental data. This method gives accurate values for the slopes; hence this was adopted to check whether the experimental data will follow the usual kinetics order for the reaction. The finding is that the data do not fit into the usual equations used for determining the order of the reaction. For these reactions the general nature of the curve is found to be sigmoid.

A typical sigmoidal curve will have induction, acceleration and deceleration periods. The three periods will be observed only in an ideal reaction. There are reactions in which either acceleration period or deceleration period is absent (Hannay 1974). In this case the concentration of one of the products formed during this reaction is measured. The rate of formation of the product is not uniform. The deceleratory period is not observable as the mechanism changes during the course of the reaction.

3.4 *Mechanistic model*

Here the polymer-supported reagent is in the solid phase and the low molecular weight ester is in the solution phase. The reaction occurs at the solid–liquid interface with the liberation of *p*-nitrophenol which can be regarded as a phase boundary reaction. The reactivity of functional groups attached to the polymer support and a substrate in the solution is being investigated here. In the case of lightly crosslinked gels, swelling has an important role in the reactivity of functional groups. But for highly crosslinked gels, swelling will only partially influence the reactivity of functional groups. These resins have a discrete and accessible internal surface and surface area even in the dry state and hence reaction can occur quickly.

For the earlier part of the aminolysis reaction of *p*-nitrophenyl ester of benzoyl glycine with crosslinked resins, a three-dimensional phase boundary model with spherical symmetry is assumed. According to this model, the reaction occurs at the exterior of the surface of the polymer. The radial rates of the reaction everywhere at the



Scheme 3. Aminolysis of *p*-nitrophenyl ester of benzoyl glycine with amino polyacrylamides.

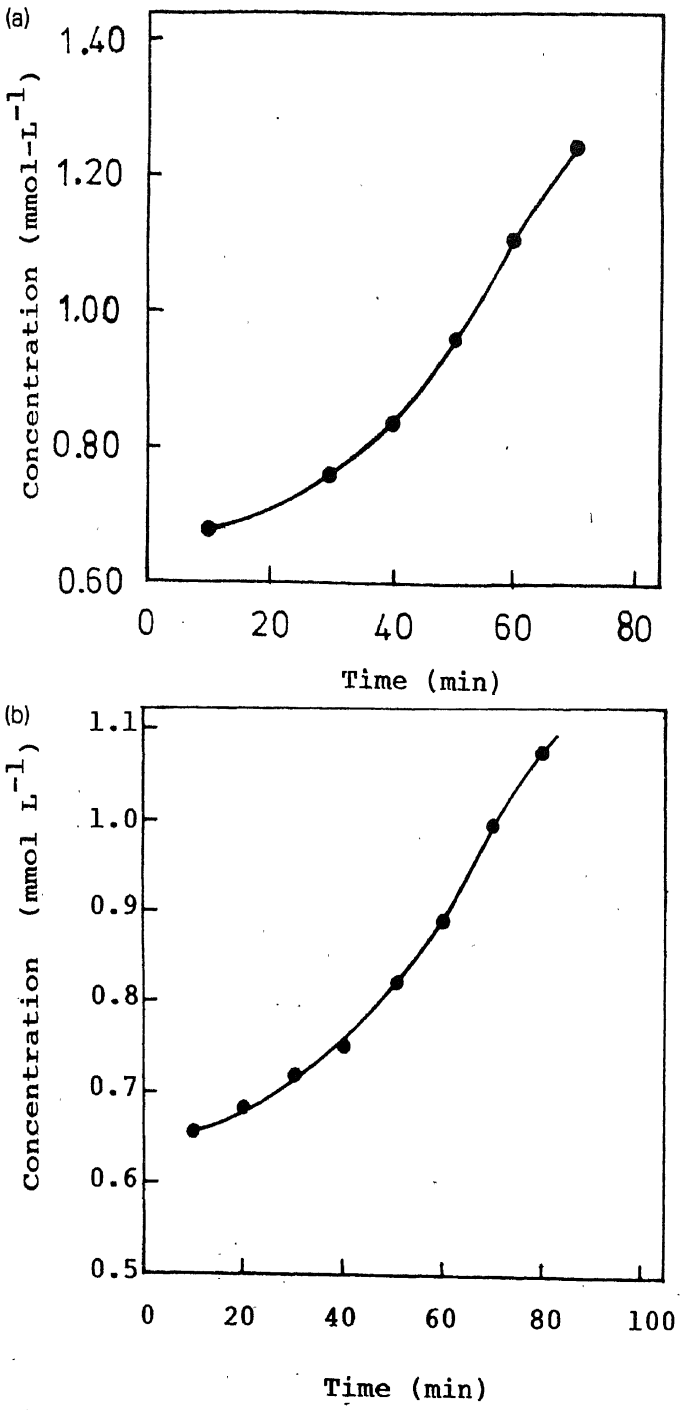


Figure 1. Concentration-time plot of 20% TEGDMA-crosslinked polyacrylamides **(a)** and 20% NNMBA-crosslinked polyacrylamides **(b)**.

surface should be same. An equation for this three-dimensional phase boundary model is (Skvara and Satava 1970; Hannay 1974),

$$[1-(1-\alpha)^{1/3}] = Kt,$$

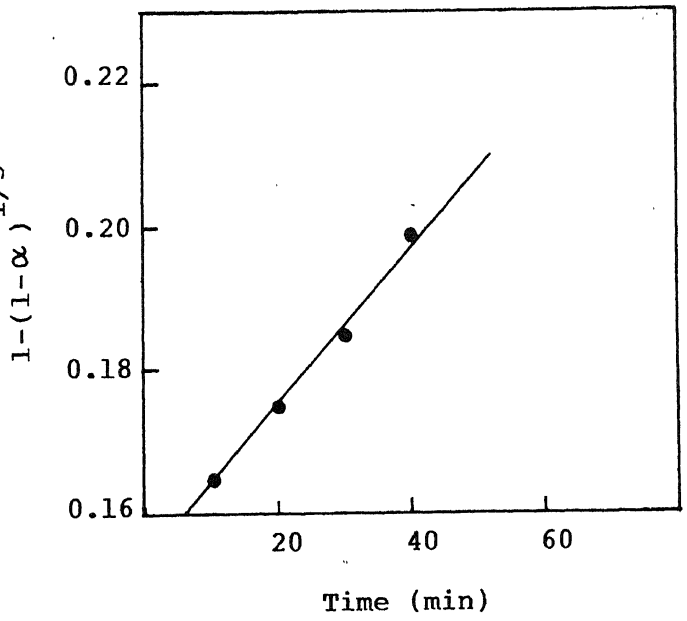


Figure 2. $[1-(1-\alpha)^{1/3}]$-time plot of NNMBA-crosslinked polyacrylamides.

where α - fraction reacted;
k - rate constant;
t - time.

The data were found to fit the above equations giving a straight line with very high correlation coefficient (γ) values for most of the resins (figure 2). The overall rate constants for different crosslinked resins are given in table 2.

In these resins, small deviations are observed after 50 min, which are due to change in the local polarity and differences in pore size, both leading to inaccessibility of the functional groups (Geising and Hornle 1973) to the reactive sites at the surface. In such cases, the ester has to penetrate into the polymer matrix in order to reach the reaction sites. This necessitates a three-dimensional diffusion. Hence the reaction follows a phase boundary process up to 40–50 min and it follows the kinetic equation $[1-(1-\alpha)^{1/3}] = Kt$, (figure 3a). Thereafter the reaction is fully controlled by three-dimensional diffusion. For such a reaction the kinetic equation is $[1-(1-\alpha)^{1/3}]^2 = Kt$, (figure 3b). The rate constants for the phase boundary part and the diffusion part of the reaction calculated separately are given in table 3.

In the case of swollen wet resins it was found that the rate of the reaction decreases (table 4) as compared to the simultaneous swelling and aminolysis of the low molecular *p*-nitrophenyl ester of benzoyl glycine by the resin.

4. Conclusion

A phase boundary reaction is possible for a reaction involving a reagent in the solid phase and the reactant in the liquid phase. For such reactions, initially the reaction occurs on the surface followed by diffusion in the later stage. A three-dimensional phase boundary model with spherical symmetry is proposed for aminolysis of *p*-nitrophenyl

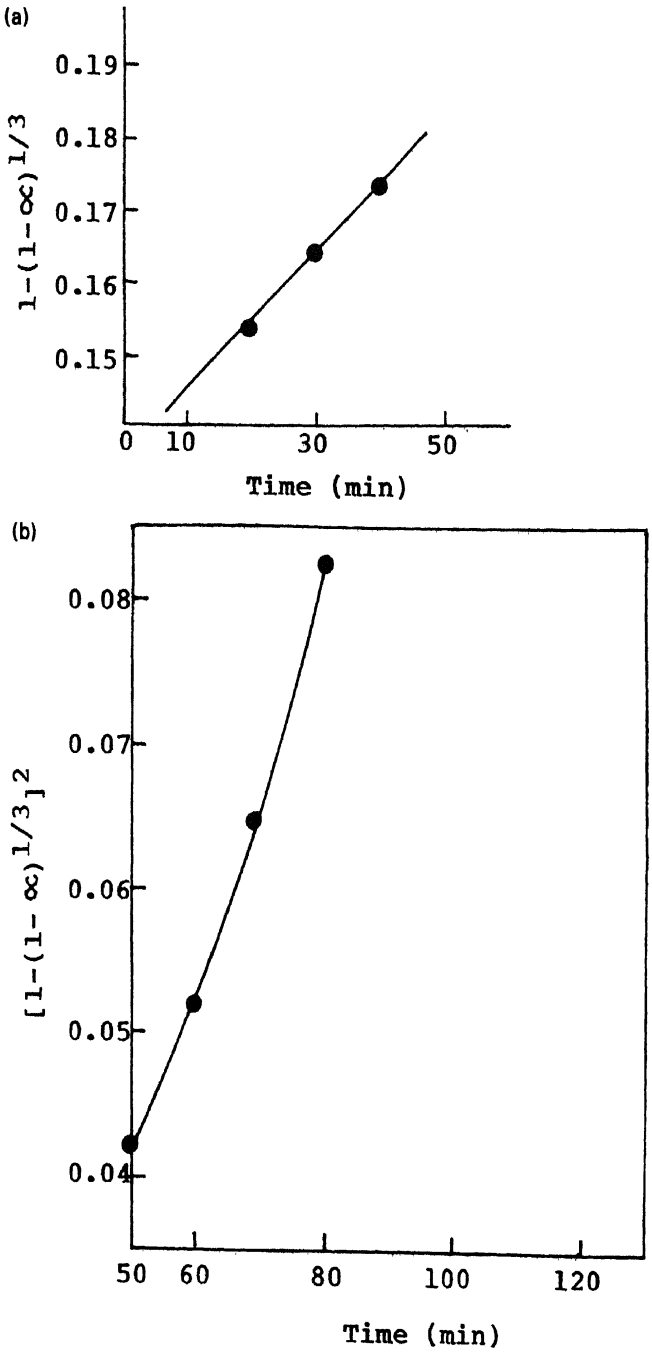


Figure 3. (a) $[1-(1-\alpha)^{1/3}]$-time plot (for three-dimensional phase boundary mechanism), and **(b)** $[1-(1-\alpha)^{1/3}]^2$-time plot (for three-dimensional diffusion mechanism), of NNMBA-crosslinked polyacrylamides.

ester of benzoyl glycine with different crosslinked resins. The mechanism of the aminolysis reaction depends on the polarity of the medium and the chemical nature of the crosslinking agents. The mechanism proposed for the reaction involves two steps,

Table 3. Rate constants for phase boundary and diffusion processes.

Crosslinking agent	Rate constants ($\times 10^{-3}$) Phase boundary (min^{-1})	Diffusion (min^{-1})	Correlation coefficient
NNMBA	0·96	1·49	0·99
TEGDMA	1·6	3·20	0·99

Table 4. Rate constants for 10% acrylamide TEGDMA crosslinked swollen wet resin and dry resin.

Nature of the resin	Aminolysis (%)	Rate constant ($\times 10^{-3}$)(min^{-1})
Swollen wet resin	44	0·21
Dry resin	76	3·51

(i) an initial phase boundary process which follows the kinetic equation,

$$[1-(1-\alpha)^{1/3}] = Kt,$$

and (ii) a change in the mechanism to three-dimensional diffusion which follows the kinetic equation,

$$[1-(1-\alpha)^{1/3}]^2 = Kt,$$

The proposed model does not show any dispersion in rate constants and therefore this model is more suitable for calculating rate constants.

References

Atherton E and Sheppard R C 1977 In *Peptides* (eds) M Goodman and J Meienhofer (New York: Wiley) p. 503

Atherton E, Logan C J and Sheppard R C 1981 *J. Chem. Soc., Perkin Trans. 1* 538

Bodanszky M, Gross E and Meinhofer J (eds) 1979 *The peptides* (New York: Academic Press) 1 105

de Boor C 1978 *A practical guide to spline* (New York: Springer-Verlag)

Devaky K S, George B K and Pillai V N R 1990 *Indian. J. Chem.* **A29** 1045

Flory P J 1939 *J. Am. Chem. Soc.* **61** 3334

Geising W and Hornle S 1973 *Proceedings of the 11th European polypeptides symposium* (ed.) H Nesvatba (Amsterdam: North Holland) p. 146

George B K and Pillai V N R 1988 *Macromolecules* **21** 1867

Guyot A and Bartholin M 1982 *Prog. Polym. Sci.* **8** 277

Hannay N B 1974 *Treatise on solid state chemistry* (Oxford: Pergamon) p. 4

Marshall G R and Merrifield R B 1971 In *Biochemical aspects of reactions on solid supports* (ed.) G R Sterk (New York: Academic Press) p. 111

Merrifield R B 1963 *J. Am. Chem. Soc.* **85** 2149

Morawetz H 1979a *Israel J. Chem.* **17** 287

Morawetz H 1979b *J. Macromol. Sci. Chem.* **A13** 311

Morawetz H 1988 *Chemical reactions on polymers* (Washington, DC: Am. Chem. Soc.) p. 317
Sheppard R C 1971 *Peptides* (Amsterdam: North Holland) p. 111
Skvara F and Satava V 1970 *J. Therm. Anal.* **2** 330
Spackman D H, Stein W H and Moore S C 1958 *Anal. Chem.* **30** 1190