

Polyoxyethylene–Polyoxypropylene Block Copolymers: a Novel Phase Transition in Aqueous Solutions of Pluronic F87 (Poloxamer 237)

Neil Mitchard,^a Anthony Beezer,^a Nicolas Rees,^b John Mitchell,^b Stephen Leharne,^c Babur Chowdhry,^c and Graham Buckton^d

^a Department of Chemistry, Royal Holloway and Bedford New College (London University), Egham Hill, Egham, Surrey TW20 0EX, UK

^b School of Chemistry, Thames Polytechnic, Woolwich, London SE18 6PF, UK

^c School of Biological Sciences, Thames Polytechnic, Woolwich, London SE18 6PF, UK

^d School of Pharmacy, University of London, Brunswick Square, London WC1N 1AX, UK

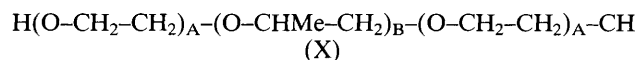
An unexpected phase transition in dilute aqueous solution of the nonionic surfactant poloxamer P237 (Pluronic F87), with an excess specific heat capacity maximum at 307.7 K in the concentration range 1–10 mg/ml, has been observed using high sensitivity differential scanning calorimetry (HSDSC); the thermodynamic parameters associated with this transition are reported.

The study of conformational transitions in dilute solutions of proteins is well documented,^{1,2} as is the theory and practice of the recording technique of differential scanning calorimetry (DSC) including the assignment of both ΔH_{cal} , the calorimetric enthalpy, and ΔH_{VH} , the van't Hoff enthalpy.³ Such DSC data for proteins have been accounted^{1,2} for in terms of, *e.g.*, domain melting and native \rightarrow denatured (folded \rightarrow unfolded) transitions.

DSC has also been widely used to investigate the thermal properties of polymers. The emphasis, normally, is on investigations of amorphous solids or crystalline materials to investigate phenomena such as melting, fusion, crystallization, glass transitions, degree of crystallinity *etc.*⁴ and such studies usually employ large sample sizes (>10 mg). In addition gel \rightarrow sol transitions, hydration/dehydration phenomena, as well the effect of solvents (*e.g.* plasticizers) on thermodynamic parameters have also been examined by DSC. The sample used for such investigations may be in suspension or semi-solid (gel).⁵ Thermodynamic parameters of these systems are more often measured using low-sensitivity DSC instruments³ at relatively high concentrations of polymer (>>10% w/w) and high scan rates (>2 K min⁻¹; normally 5–8 K min⁻¹).

There are, however, almost no data on thermodynamic characteristics for polymer transitions in dilute organic or aqueous media. For polymeric surfactants, solution-phase properties are of crucial importance since changes in conformation/association/hydration properties as a function of temperature are not only of fundamental interest but may also be of importance in, *e.g.*, affecting detergent properties in solution or pharmaceutical stability in formulations containing these polymers *etc.* We report here what we believe to be the first example of a phase transition in dilute aqueous solution (0.1–1%, w/w) of a synthetic organic block copolymer,

pluronic F87 (P237). The pluronic group of surfactants (poloxamers are a series of propylene oxide/ethylene oxide (ABA), block copolymers with the general formula (X).



Hydrophobicity is conferred on the polymer by the propylene oxide block. Hydrophilicity is provided by the ethylene oxide block. The poloxamers are used extensively in industry for a variety of applications especially in cosmetic formulations and drug delivery systems. In these areas they are used in the production of aqueous preparations of non-water soluble compounds. The P237 (supplied by Ciba-Geigy) gave a single elution peak on gel permeation chromatography. Both P237 and P407 consist of 70% ethylene oxide (EO) and 30% propylene oxide (PO) (P237; *av. M_r* = 7700, *M_r* for EO = 5390, *M_r* for PO = 2310; P407, *av. M_r* = 12500, *M_r* for EO = 8750, *M_r* for PO = 3750). The physical and chemical properties of block copolymers have been well reviewed.^{6–8} Gilbert and co-workers⁷ have used fluorescent probes to study gelation in Pluronic F127 (P407) and demonstrated the existence of some critical temperature in dilute solution (5–10% concentration, w/w) at which the diffusion coefficient of small molecules through the hydrophobic portions becomes reduced. For the hydrophilic segments at this temperature there is an increase in microenvironmental polarity possibly due to dehydration. It is the critical temperature behaviour in even more dilute solution which we believe we are capable of investigating by the use of HSDSC. It has also been reported that poloxamers similar to P237 such as P407 at a concentration range of 15–30% (w/w) in aqueous solutions (under conditions which are temperature and concentration dependent) form micelles which undergo a thermally induced swelling together with desolvation.⁹

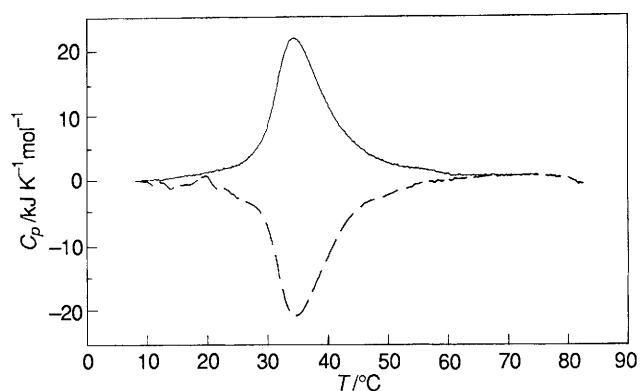


Figure 1. Temperature dependence of the partial specific heat capacity of P237 at a scan rate of 0.5 K min^{-1} in phosphate buffer, pH 7.2 (0.067 M). Poloxamer concentration was 6.47 mg ml^{-1} . The thermodynamic parameters of the transition were identical in double distilled H_2O compared to that in phosphate buffer. The HSDSC recording shows the heating (A) and subsequent cooling scan (B) of P237 (30 and -30 K h^{-1} respectively). The experiments were conducted using a Microcal Mc-2 microcalorimeter (Microcal Amherst, Mass., USA).

Table 1. Microcalorimetrically recorded thermodynamic parameters for P237 at a scan rate of 0.5 K min^{-1} in phosphate buffer (0.067 M; pH 7.2) at a concentration of P237 of 6.47 mg ml^{-1} .

Thermodynamic parameter	Value
T_m	$307.6 \text{ K (} 34.35^\circ\text{C)}$
ΔH_{cal}	$204.4 \text{ kJ mol}^{-1}$
ΔH_{VH}	$305.3 \text{ kJ mol}^{-1}$
$\Delta H_{\text{VH}}/\Delta H_{\text{cal}}$	1.5
$\Delta T_{1/2}$	8.8°C
C_p^{max}	$19.9 \text{ kJ K}^{-1} \text{ mol}^{-1}$

Further it is hypothesized by Lenaerts *et al.*⁹ on the basis of rheological, DSC, and X-ray diffraction studies that as the temperature of a P407 solution is increased a cross-linked network with no preferential three-dimensional arrangement is generated.

The phase transitions exhibited by P237 are shown in Figure 1, and the associated thermodynamic parameters are given in Table 1. Interestingly the transition is reversible in the sense that the heating endotherm and cooling exotherm scans are, within experimental error, identical. This reversibility indicates that the process, or processes, occurring during the transition are fast in relation to the minimum time course of the experiments. The first and second heating scans of the same sample were also identical. The heating and cooling transitions do not show any hysteresis and the transition is not kinetically limited (in the concentration range examined) as indicated by the identity of the T_m (the temperature at the maximum value of the excess specific heat capacity) with varying scan rates between 10 and 60 K h^{-1} . As a macroscopic technique HSDSC does not usually give any direct information as to the molecular processes occurring during the transition. However, initial HSDSC observations indicate that the processes occurring involve water and possibly include desolvation during the heating scan. This hypothesis is supported by the fact that at a scan rate of 10 K h^{-1} different values for the T_m of the transition and the calorimetric enthalpy (ΔH_{cal}) are obtained in H_2O and D_2O . Furthermore the T_m is increased and the ΔH_{cal} of the transition decreased in 0.2 M sodium perchlorate compared to the value of these parameters in H_2O . However, because the ΔH_{cal} is only slightly reduced in D_2O and sodium perchlorate solutions

compared to that in H_2O it is probable that a conformational change is also taking place in the poloxamer during the transition.

Many of the thermodynamic parameters (*e.g.* ΔH_{cal} , ΔH_{VH}), and $T_{1/2}$ (the transition width in $^\circ\text{C}$ at $1/2 C_p^{\text{max}}$) are of a similar order of magnitude to those found in the native \rightarrow denatured (folded \rightarrow unfolded) transition of globular proteins.¹ The value of 1.5 for $\Delta H_{\text{VH}}/\Delta H_{\text{cal}}$ of P237 can be explained since for reversible processes that are subject solely to thermodynamic limitation during their observation by HSDSC $\Delta H_{\text{cal}} = \Delta H_{\text{VH}}$ for a strictly two-state process, carried out under essentially equilibrium conditions such as the denaturation of globular proteins, *e.g.* ribonuclease.¹ If $\Delta H_{\text{VH}} < \Delta H_{\text{cal}}$ it can be concluded that one or more intermediate states are of significance in the overall process, whereas if $\Delta H_{\text{VH}} > \Delta H_{\text{cal}}$ intermolecular co-operation is clearly indicated.² The highly reproducible value of 1.5 for $\Delta H_{\text{VH}}/\Delta H_{\text{cal}}$ in the case of P237 at a scan rate of 0.5 K min^{-1} means that the process under study is not a two-state process.

A number of exciting facets emerge concerning the presently reported work. It is, as far as we know, the first instance in which HSDSC has been used to study the dilute aqueous solution properties of a synthetic polymer. Thus HSDSC provides a practicable direct route for accurate measurements of thermodynamic transitions in dilute solution. As for the phase transition of P237 we believe it consists of two processes, one of which involves some degree of dehydration coupled with the other process of conformational change. Initial NMR results obtained in our laboratory certainly indicate the latter process to be occurring. We finally believe, based upon our initial observations of the phase transition of P407 (the polymeric surfactant investigated by Gilbert *et al.*⁷), that the transition in P237 is the same as that which Gilbert *et al.*⁷ alluded to in their low-concentration work.

Further studies in our laboratories are in progress in order to elucidate the molecular basis of the phase transition phenomenon observed in P237, using techniques such as T_1 NMR and X-ray scattering in solution with blockcopolymer surfactants based upon Merazapol and Poloxamine. The interest will, in part, relate to the importance of the fraction of blocks A and B present in the polymer and the relationship of the experimentally derived data, ΔH_{cal} , ΔH_{VH} , $\Delta T_{1/2}$, C_p *etc.*, to the structure of the polymers in solution. As a necessity comparisons will be made with the behaviour of the solid phases and with more concentrated solutions.

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