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Effects of formulation variables on characteristics of poly (ethylcyanoacrylate) nanocapsules prepared from w/o microemulsions

S. Watnasirichaikul, T. Rades *, I.G. Tucker, N.M. Davies

Formulation and Drug Delivery Group, School of Pharmacy, University of Otago, PO Box 913, Dunedin, New Zealand

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Abstract

The effect of several formulation variables on some of the physico-chemical characteristics of poly (ethyl cyanoacrylate) (PECA) nanocapsules prepared by the interfacial polymerisation of biocompatible water-in-oil microemulsions was investigated. In all cases, yields were high (> 90%) and the polydispersity in size of nanocapsules was narrow. The molecular weight of the nanocapsules formed was influenced by the pH of the aqueous component of the microemulsion, increasing with increasing pH. The size of the nanocapsules formed (ranging from around 130 to 180 nm) was a function of the ratio of the mass of monomer used to the water weight fraction of the microemulsion, increasing as this ratio was increased. This is due to the formation of a thicker polymer wall resulting from the increased mass of monomer available per unit interfacial area as this ratio is increased. The rate of release of insulin from nanocapsules was also influenced by this ratio, in agreement with its effect on wall thickness. This study demonstrates that many pharmaceutically relevant physico-chemical properties of poly (alkyl cyanoacrylate) (PACA) nanocapsules prepared by interfacial polymerisation of microemulsions can readily be manipulated by changing either the pH of the aqueous component, the water weight fraction of the microemulsion or the mass of monomer used for polymerisation.

Keywords: Insulin; Interfacial polymerisation; Microemulsions; Nanocapsules; Peptides; Poly (alkyl cyanoacrylate)

1. Introduction

Aqueous cored biodegradable poly(alkyl cyanoacrylate) (PACA) nanocapsules suitable for the encapsulation of proteins and peptides can be prepared by interfacial polymerisation of water-in-oil microemulsions (Watnasirichaikul et al., 2000). The use of microemulsions offers advantages over the use of size-reduced kinetically stabilised emulsions as a template for the preparation of nanocapsules by interfacial polymerisation. Microemulsions are thermodynamically stable and form spontaneously so require minimal input of energy for their formation. They have a small and uniform droplet size, and they

* Corresponding author. Tel.: + 64-3-479-5410; fax: + 64-3-479-7034.

E-mail address: thomas.rades@stonebow.otago.ac.nz (T. Rades).

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form reproducibly. All these properties render them amenable to industrial scale-up. Further, if biocompatible oils and surfactants are used for the formation of the microemulsions, the necessity of isolating the nanocapsules from the reaction matrix following polymerisation is removed. Nanocapsules can therefore be prepared in situ in a microemulsion matrix.

The dispersion of bioerodible PACA nanocapsules in a biocompatible microemulsion matrix may prove beneficial for the oral delivery of proteins or peptides. Such a formulation may exploit the benefits of both nanoparticles, in their ability to translocate the intestinal epithelium and protect entrapped bioactive (Aprahamian et al., 1987; Lowe and Temple, 1994; Damgé et al., 1997; Lambert et al., 2000) and microemulsions which can affect membrane permeability and promote uptake by lymph (Ritschel, 1991; Constantinides et al., 1994; Porter et al., 1996). However, to achieve effective delivery following oral administration, such a formulation must have suitable release characteristics. The formulation must protect the protein or peptide whilst in the hostile conditions of the stomach and the intestine either by entrapment of the bioactive within the nanocapsules or its dispersion in the oily matrix. The formulation must then release the bioactive at an appropriate rate either at the site of absorption or following uptake of the nanocapsules such that the bioactive is able to exert its effect.

The aim of this work was to investigate the effect of various formulation variables on the preparation of nanocapsules by interfacial polymerisation of biocompatible microemulsions to establish whether the characteristics of the nanocapsules formed (including the release rate of insulin) could be manipulated.

2. Materials and methods

2.1. Materials

Caprylic/capric triglycerides (Crodamol GTCC™), polysorbate 80 (Crillet 4™) and sorbitan mono-oleate (Crill 4™) were supplied by Croda Surfactants NZ (Auckland, NZ). Caprylic/capric mono-/diglycerides (Capmul MCM™) was a gift from Abitec Corp. (Columbus, OH). Ethyl 2-cyanoacrylate was purchased from Sigma (St. Louis, MO). Human insulin (Humulin R™) was obtained from Eli Lilly (Auckland, NZ). Acetonitrile (HPLC grade) and chloroform (AR grade) were purchased from BDH (Poole, Dorset, UK). Distilled water was used throughout.

2.2. Preparation of nanocapsules

Microemulsions were prepared at 4 °C from a surfactant blend (polysorbate 80 and sorbitan mono-oleate, 3:2 weight ratio), an oil mixture (caprylic/capric triglycerides and caprylic/capric mono-/diglycerides, 3:1 weight ratio) and water as previously described (Watnasirichaikul et al., 2000). All microemulsions were prepared to contain 14% w/w of the surfactant blend. A solution of ethyl 2-cyanoacrylate monomer (50, 100 or 150 mg) in 300 mg of chloroform was slowly added to 10 g of microemulsion under mechanical stirring. The system was left for at least 4 h at 4 °C for polymerisation. Poly(ethyl 2-cyanoacrylate) (PECA) nanocapsules were collected for characterisation by centrifugation at $51\,500 \times g$ for 60 min at 25 °C (Beckman J2/MC Centrifuge, JA20.1 rotor). The pH of the aqueous phase was adjusted using hydrochloric acid and sodium hydroxide (pH 2.5 and 5.0) or a phosphate buffer (pH 7.4, 0.011 M).

For the preparation of PECA nanocapsules containing insulin, an aqueous solution of insulin having a concentration of 100 U/ml and a pH of 7.4 was used as the aqueous component of microemulsions.

2.3. Characterisation of nanocapsules

The particle size and distribution of the PECA nanocapsules was determined by photon correlation spectroscopy (Zetasizer 3000, Malvern Instruments Ltd.). Prior to analysis, residual oil and surfactant was removed by repeated washing (at least two times) in ethanol and centrifugation ($18\,500 \times g$ for 10 min at 25 °C). For particle size analysis, insulin-free nanocapsules were dispersed in 0.2% w/w polysorbate 80 in ethanol while

nanocapsules containing insulin were dispersed in water containing 0.2% w/w polysorbate 20. Measurements were carried out at 25 °C.

For the determination of insulin-free PECA nanocapsule yield and resultant polymer molecular weight, samples were processed as for particle size determination to remove residual oil and surfactant. Nanocapsules were then dried to constant weight (60 °C for 4 h and stored in a dessicator overnight). Yield was estimated from the mass of recovered dried nanocapsules as compared with the mass of monomer added to the microemulsion. It was noted that about 3 mg of polymer was constantly lost as a result of the washing process. Estimates for yield were thus compensated for this loss. Molecular weight of the PECA resulting from the interfacial polymerisation of the various systems studied was determined by gel permeation chromatography based on the method of Das et al. (1995) using a Waters Associates Chromatography system fitted with five Ultrastyragel™ columns (10^6–10^2 Å). Tetrahydrofuran was used as the mobile phase at a flow rate of 1.0 ml/min. Solutions of the PECA were prepared (2% w/v) in tetrahydrofuran and filtered through poly(tetrafluoroethylene) membrane filters with a pore size of 0.45 μm (Millipore) prior to analysis. Eighty microliter of sample was injected and the eluent monitored by a differential refractometer (Water 410, sensitivity range 256–512). The columns were calibrated using a series of 12 monodisperse polystyrene standards in the molecular weight range of $900–2 \times 10^6$ g/mol. The number average molecular weight (Mn), weight average molecular weight (Mw) and peak molecular weight (Mp) were determined for all samples. The polymer molecular weight distribution was estimated by calculating the polydispersity coefficient, d (where $d = \mathrm{Mw}/\mathrm{Mn}$). These molecular weight parameters were also determined, as described, for PECA nanocapsules containing insulin.

Statistical analysis of the effect of water weight fraction of the microemulsion, pH of the aqueous disperse phase and mass of ethyl 2-cyanoacrylate monomer on yield, size of nanocapsules and molecular weight of the polymer was carried out using a general linear model analysis of variance with a level of significance of $P < 0.05$ (Minitab™).

2.4. Release of insulin from nanocapsules

Polymerised microemulsions (4 g) containing insulin were diluted to 25 ml with water (adjusted to pH 2.5) and transferred into a water-jacketed beaker incubated at 45 °C and mechanically stirred (200 rpm). These conditions were chosen to obtain a time-release profile suitable for the investigation of PECA nanocapsules of different wall thickness. At various times, 300 μl of the release medium was sampled and released insulin estimated by HPLC following dilution of the samples with an equal amount of 80% methanol according to the previously described method (Watnasirichaikul et al., 2000). The poorly water-soluble components of the system (nanocapsules and oil) were separated from the methanolic aqueous phase prior to HPLC analysis by centrifugation ($12\,000 \times g$ for 12 min at room temperature). The zero-order release rates of insulin were estimated from the linear portion of the release rate versus time profiles obtained for the various systems.

3. Results and discussion

3.1. Factorial study analysis

A factorial study was used to investigate the effects of the aqueous weight fraction of the microemulsion, the pH of the aqueous disperse phase and the mass of monomer used on some of the physico-chemical characteristics of PECA nanocapsules prepared by the interfacial polymerisation of a microemulsion. Analysis of variance demonstrated that all three variables significantly affected the size of the nanocapsules formed, mass of monomer affected size polydispersity, pH had a significant effect on the resulting molecular weight of the polymer, while yield was unaffected by the parameters investigated. Significant first-order interactions were observed between all three variables in terms of the size of the nanocapsules formed. No significant second-order interactions were observed.

3.2. Effects on size of nanocapsules

A significant increase in particle size was observed when the dispersed aqueous phase of the microemulsion was buffered to pH 7.4 using a 0.011 M phosphate buffer compared with when the aqueous phase was adjusted to either pH 5 or 2.5. Further investigations varying the concentration of the buffer salts used revealed that this increase in size is a result of the presence of buffer ions rather than a pH effect. A significant increase in size was observed when the buffer concentration was increased three-fold while reducing the concentration of salt resulted in a significant decrease in the size of the resulting nanocapsules which tended towards that measured at the two lower pH levels (Fig. 1).

Both monomer mass and aqueous weight fraction had a significant effect on particle size at all pH values (Fig. 2). Increasing the mass of the monomer added to a microemulsion having a particular aqueous weight fraction resulted in nanoparticles having a larger size which may be due to the formation of a thicker polymer wall resulting from the increased mass of monomer available per unit interfacial area of the dispersed phase. This is in agreement with that reported by El-Samaligy et al. (1986) where the size of poly (methyl cyanoacrylate) nanocapsules formed by

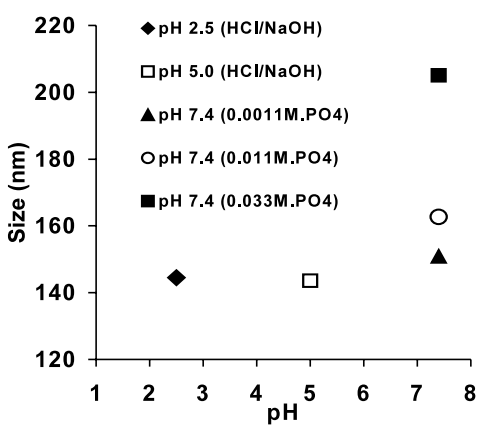

Fig. 1. Effect of pH and phosphate buffer concentrations on particle size of nanocapsules prepared from a microemulsion having a water weight fraction of 10% using 100 mg ethyl 2-cyanoacrylate. Values represent means, $n = 2$.

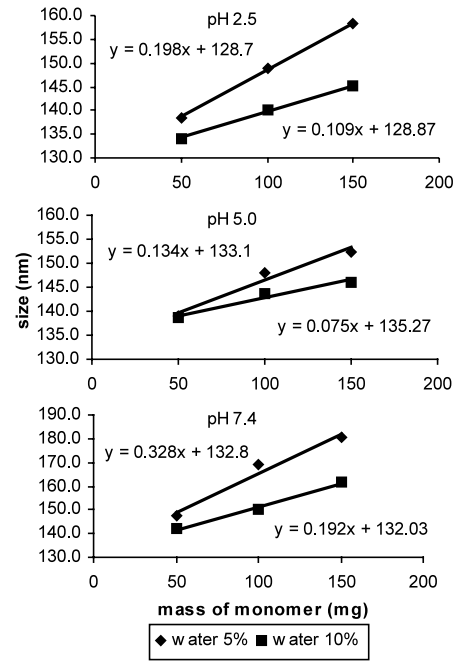

Fig. 2. Effect of ethyl 2-cyanoacrylate mass and water weight fraction of microemulsions on particle size of nanocapsules. Values represent means, $n = 2$.

the interfacial polymerisation of kinetically stabilised water-in-oil sub-micron emulsions also increased with increasing mass of monomer. Addition of an equivalent mass of monomer to microemulsions having an increasing aqueous weight fraction resulted in the formation of smaller nanocapsules. This is likely to result from a decrease in the mass of monomer available per unit interfacial area yielding nanocapsules having a thinner polymer wall and hence a smaller size. Fig. 3 is a typical freeze-fracture transmission electron micrograph of PECA nanocapsules prepared by the interfacial polymerisation of the water-in-oil microemulsion showing a central cavity surrounded by a polymer wall.

Extrapolation to zero of the near linear relationships noted between monomer mass and size of nanocapsules produced from a microemulsion having a particular water weight fraction and pH (Fig. 2) suggests that a microemulsion to which no monomer is added has a particle size of between 129 and 135 nm. However, when the parti-

cle size of the microemulsions was estimated by photon correlation spectroscopy, a size of around 20 nm was obtained which is in agreement with that reported in the literature (Constantinides et al., 1994). This discrepancy in size between that measured for the microemulsion and that estimated from the plot of monomer mass versus size of nanocapsules proposes some kind of structural collapse of the microemulsion occurs upon polymerisation resulting in a larger droplet size of the disperse phase.

The difference between the size of the nanocapsules formed and the estimated size of the collapsed microemulsion allows for an estimation of polymer wall thickness of the capsules. Wall thickness increased in a near linear fashion with increasing monomer mass and was dependent on the water weight fraction (Table 1). Thus, like nanocapsule size, wall thickness of the nanocapsules formed by the interfacial polymerisation of water-in-oil microemulsions appears to be a function of the ratio of monomer mass to the water weight fraction of the microemulsion.

The polydispersity of particle size (a parameter indicating the width of the size distribution and equivalent to the variance of the log normal model, in which a value of less than 0.08 is regarded as being nearly monodisperse) was narrow in all cases, being less than 0.2 and typically less than 0.1 (Fig. 4). This distribution is considerably narrower than that reported for PACA nanoparticles prepared by interfacial polymerisation of kinetically stabilised emulsions (El-Samaligy et al., 1986; Krause et al., 1986; Lambert et al., 2000). This may be a reflection of the uniform size distribution of the swollen reverse micelles present in the water-in-oil microemulsion compared with that of thermodynamically unstable sub-micron emulsions, despite a possible structural collapse. The polydispersity noted in the present study is also smaller than that reported for PACA nanoparticles prepared by micellar polymerisation in which large agglomerates have been reported (Krause et al., 1986; Fontana et al., 1998). Thus, interfacial polymerisation of microe-

Table 1
Estimated wall thickness (nm) of nanocapsules prepared from microemulsions of different aqueous weight fractions and pH

pH	Water (%w/w)	Monomer mass (mg)		
		50	100	150
2.5	5	4.9	10.2	14.8
	10	2.7	5.6	8.1
5.0	5	3.0	7.4	9.7
	10	1.7	4.2	5.5
7.4	5	7.4	18.1	23.7
	10	5.1	9.1	14.7

Microemulsions were polymerised using different masses of ethyl 2-cyanoacrylate. Values represent means, $n = 2$.

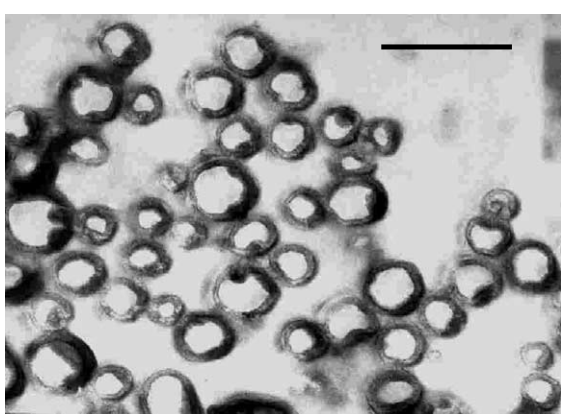

Fig. 3. Freeze-fracture transmission electron micrograph of poly (ethyl 2-cyanocarylate) nanocapsules (bar = 400 nm).

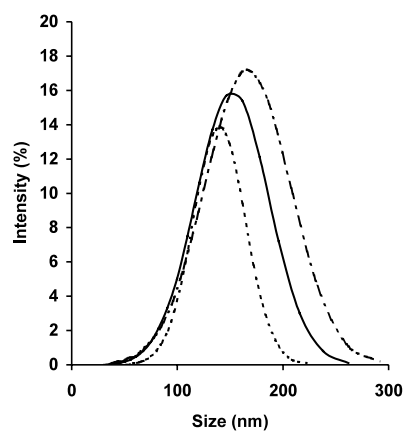

Fig. 4. Size distribution of nanocapsules prepared using 50 mg (---), 100 mg (—) and 150 mg (- - -) of ethyl 2-cyanoacrylate added to 10 g of microemulsions having an aqueous component of pH 2.5 and water weight fraction of 10%.

mulsions would seem the preferred method for the preparation of nanocapsules having a narrow size distribution. The polydispersity index was, however, significantly affected by monomer mass ($P < 0.05$). The average polydispersity index of all formulations ($3 \times$ pH and $2 \times$ water weight fraction) prepared at 50, 100 and 150 mg of ethyl 2-cyanoacrylate were 0.041, 0.055 and 0.082, respectively. Neither pH nor water weight fraction influenced the polydispersity of the resulting nanocapsules.

3.3. Effects on yield of nanocapsules

A consequence of the narrow size distribution of the nanocapsules formed by the interfacial polymerisation of water-in-oil microemulsions is the high yield obtained, as no loss occurs upon removal of aggregates by filtration. In all cases, yield was greater than 90% and was not affected by any of the formulation variables studied. The yields obtained following interfacial polymerisation of water-in-oil microemulsions are greater than those reported following interfacial polymerisation of kinetically stabilised sub-micron emulsions (Krause et al., 1986) and micellar polymerisation (Krause et al., 1986; Das et al., 1995). A loss of around 3 mg was observed for all formulations upon repeated washing. However, since the nanocapsules are prepared from a microemulsion formulated using biocompatible oils and surfactants, and are intended to be administered in such a vehicle, this loss (albeit small) would not occur upon usual preparation.

3.4. Effects on molecular weight of polymer

The molecular weight elution profile of the resultant polymer was monomodal in all cases (e.g. Fig. 5A) except for nanocapsules prepared using 100 and 150 mg of ethyl 2-cyanoacrylate and microemulsions in which the aqueous weight fraction was 10% w/w and pH 7.4. Under these circumstances, a bimodal distribution was observed (e.g. Fig. 5B) with a peak corresponding to a molecular weight of between 15 000 and 80 000 g/mol in addition to a symmetrical peak corresponding to a molecular weight of between 2000

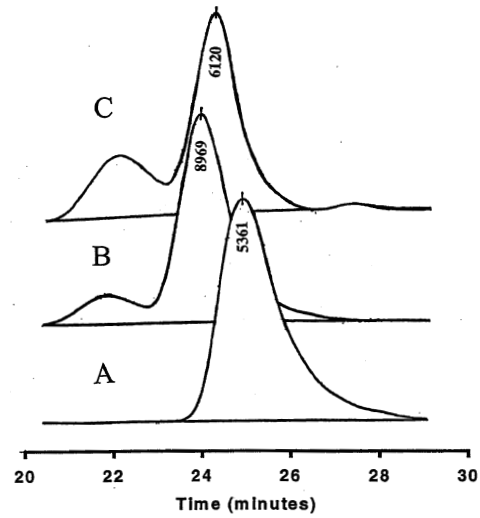

Fig. 5. Gel permeation chromatograms of the PECA polymer resulting from interfacial polymerisation when 100 mg of ethyl 2-cyanoacrylate is added to 10 g of microemulsions having 10% aqueous weight fraction where the aqueous compartment is either water pH 5.0 (A), water pH 7.4 (B) or a proprietary insulin solution of pH 7.4 (C).

and 10 000 g/mol which was observed in all samples. The molecular weight distribution profile observed at pH 7.4 was not, however, influenced by the buffer salt concentration used in the aqueous, which is in contrast to that noted on its effect on particle size.

Such a bimodal distribution in molecular weight has previously been reported by Vansnick et al. (1985) following micellar polymerisation of *iso*-butyl cyanoacrylate using polyoxyethylene-polyoxypropylene as surfactant. Under such conditions, however, the higher molecular weight peak was dominant and the peak associated with the lower molecular weight fraction was smaller being approximately 3000 g/mol. In the absence of polyoxyethylene–polyoxypropylene, a monomodal distribution was observed, as in most of the samples of the present investigation, except that the peak molecular weight was again much lower at around 600 g/mol. The molecular weight of poly (*iso*-butyl cyanoacrylate) nanocapsules prepared by interfacial polymerisation of an oil-in-water system was also reported to be bimodal having distributions centred around 9000 and

150 000 g/mol (Damgé et al., 1990). Further, the molecular weight of alkyl cyanoacrylate polymers formed by interfacial polymerisation also appears to increase if ethanol is used to solubilise the monomer (Gallardo et al., 1993). Thus, it would appear that the molecular weight of the polymer in PACA nanoparticles is dependent on the process used in their preparation, whether interfacial polymerisation occurs in a water-in-oil microemulsion, micellar polymerisation in an aqueous vehicle or interfacial polymerisation of an oil-in-water system and also the type of monomer used. This may have important implications in terms of carrier degradation and toxicity (Leonard et al., 1966; Vezin and Florence, 1980; Vansnick et al., 1985; Lherm et al., 1992).

The Mn associated with the lower molecular weight polymer fraction progressively increased with increasing pH of the aqueous component of the microemulsion (Fig. 6). PACAs are formed by an anionic polymerisation mechanism initiated by nucleophilic attack on the β carbon of the monomer resulting in a reactive carbanion. The reaction is initiated by hydroxyl ions and terminated by protons (Donnelly, 1977; Pepper, 1978). Thus, as pH of the aqueous is reduced, polymer termination is encouraged leading to a lowering of the Mn. In all cases, the polydispersity of the molecular weight was in the range 1.5–2.1.

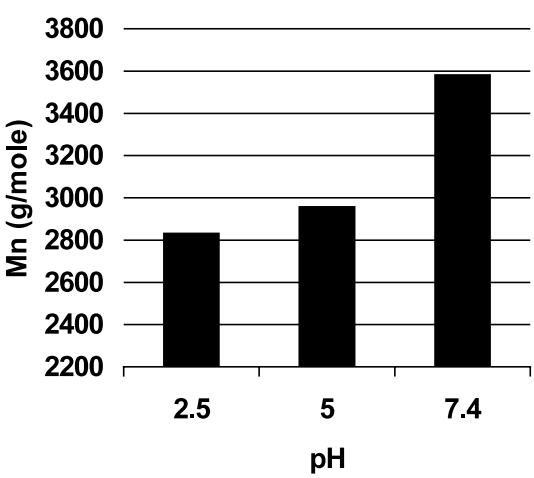

Fig. 6. Effect of pH on the number average molecular weight (Mn) of PECA. Values represent mean of six samples prepared at three monomer concentrations and two water volume fractions.

The gel permeation chromatography elution profile of PECA obtained from nanocapsules prepared from a microemulsion in which the aqueous component is a proprietary insulin solution (100 U/ml, pH 7.4) is also included in Fig. 5. Again a bimodal profile is observed which is characteristic of that obtained from a microemulsion having an aqueous component of pH 7.4 and weight fraction of 10%. However, it can be noted that in the presence of insulin, a larger percentage of the polymer mass is associated with the higher molecular weight peak, resulting in the Mw of the polymer increasing from 21 000 to 32 000 g/mol. A similar increase in the percentage of polymer associated with the higher molecular weight fraction was also reported by Damgé et al. (1990) for the preparation of poly (butyl cyanoacrylate) nanocapsules containing insulin from oil-in-water systems in which insulin was incorporated into the disperse phase. This may be a result of the intervention of some insulin in the polymerisation reaction as has been reported for other active compounds having basic residues (Vansnick et al., 1985; Grangier et al., 1991).

3.5. Encapsulation efficiency of insulin

The efficiency of insulin entrapment within PECA nanocapsules was influenced by the mass of monomer used in the polymerisation. Insulin (62.5%) was found to be associated with nanocapsules prepared by the addition of 50 mg of monomer to a microemulsion having an aqueous weight fraction of 10% while 79.4 and 78.6% of insulin was associated with nanocapsules prepared by addition of 100 and 150 mg of monomer, respectively. An increase in the entrapment of fluorescein (a hydrophilic marker) upon increasing the amount of alkyl cyanoacrylate used to polymerise a kinetically stabilised sub-micron water-in-oil emulsion was reported by El-Samaligy et al. (1986) and was considered to be a result of the increased resistance of thicker coats to the washing process.

To obtain a high drug entrapment efficiency within PACA nanoparticles produced by interfacial polymerisation of dispersed systems, the drug

should have a high affinity for the dispersed phase to be encapsulated so as to reduce loss to the continuous phase (Krause et al., 1986; Fresta et al., 1996). Thus, polymerisation of water-in-oil dispersions would appear to be a preferred method for the entrapment of hydrophilic proteins and peptides (Lambert et al., 2000). Surprisingly, high efficiencies of peptide entrapment (in excess of 90%) have been reported for nanoparticles prepared by the interfacial polymerisation of oil-in-water systems, (in which the peptide is dispersed in the oil phase) and following micellar polymerisation where the peptide is adsorbed onto nanoparticles either during or following polymerisation in an aqueous medium (Gallardo et al., 1993; Damgé et al., 1997). Successful preparation of nanocapsules containing peptide by the polymerisation of oil-in-water systems, however, would depend on the stability of the peptide in the dispersed oil phase which usually contains a large quantity of an organic solvent, for example ethanol or acetone, while for nanoparticles prepared by micellar polymerisation, a protein or peptide having a high affinity for the polymer interface would seem necessary. Preparation of nanocapsules by interfacial polymerisation of a water-in-oil microemulsion overcomes these requirements and can result in a relatively high entrapment efficiency, in this case of insulin, as the water-soluble peptide is present only in the aqueous phase of the water-in-oil microemulsion.

3.6. Effects on the release of insulin from nanocapsules

The estimated zero-order release rates of insulin from PECA nanocapsules prepared by addition of different masses of monomer to microemulsions having an aqueous weight fraction of either 5 or 10% are shown in Table 2. The duration of the period of zero-order release rate varied from between 60 to in excess of 240 min, and was influenced by monomer mass with a longer period of zero-order release noted at higher mass of monomer added to microemulsion (Table 2). The estimated zero-order release rates of insulin from PECA nanocapsules were influenced, like wall thickness, by both the mass of monomer used for polymerisation and the aqueous weight fraction of the microemulsion. The rate of insulin release was noted to decrease with increasing the ratio of monomer mass to aqueous weight fraction.

Table 2
Effect of aqueous weight fraction of the microemulsion and mass of ethyl 2-cyanoacrylate used for the preparation of nanocapsules on the zero-order rate constants (% per min) of insulin release

Aqueous (%w/w)	Monomer mass (mg)		
	50	100	150
5	0.064 (60–120)	0.035 (120–300)	0.016 (120–300)
10	0.107 (60–180)	0.062 (60–240)	0.038 (120–300)

Values represent means, $n = 2$. Values in parenthesis represent time (min) over which zero-order release was observed.

Fig. 7 is a plot of the measured zero-order rate of insulin release against the reciprocal of the estimated wall thickness of PECA nanocapsules containing insulin prepared using different masses of monomer and microemulsions having an aqueous weight fraction of either 5 or 10%. The wall thickness was estimated from a plot of

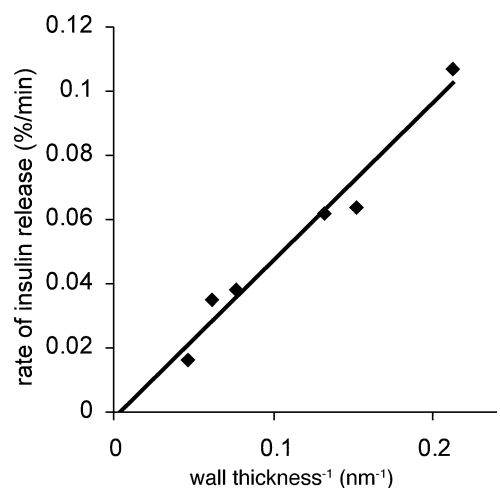

Fig. 7. Correlation of measured zero-order rate of insulin release with reciprocal of estimated wall thickness of nanocapsules.

nanocapsule size against monomer mass as described for insulin-free nanocapsules (Section 3.2). A good correlation is observed between zero-order release rate (as would be expected from a nanocapsule reservoir system) and the reciprocal of the wall thickness ($r^2 = 0.966$). This would imply that the release rate of a bioactive from PACA nanocapsules prepared by the interfacial polymerisation of a water-in-oil microemulsion could readily be controlled by manipulation of the ratio of monomer mass to the water weight fraction. Indeed from such a plot of release rate against reciprocal of the estimated wall thickness, conditions can be chosen to tailor-make nanocapsules having specified release rates provided that the relationship between particle size and monomer mass is first established.

The maximum amount of insulin released from PECA nanocapsules over the time period of the release study, however, was only about 80% of the total insulin added to the microemulsion prior to polymerisation. This again proposes some intervention of the insulin in the polymerisation reaction as suggested by the change in the molecular weight distribution of the polymer formed in the presence and absence of insulin.

4. Conclusions

This study demonstrates that PECA nanocapsules can be readily prepared by the interfacial polymerisation of water-in-oil microemulsions and that certain physico-chemical characteristics of the nanocapsules (size, wall thickness, polymer molecular weight, and release rate) can be controlled, at least to a certain extent, by manipulation of some of the formulation variables including; the amount of monomer used, the water weight fraction of the aqueous component of the microemulsion and the pH. The advantages offered in preparing PECA nanocapsules by interfacial polymerisation of water-in-oil microemulsions over other reported methods may have implications with regards to maintaining the stability (during the preparation process) and achieving efficient entrapment of certain bioactives, particularly proteins and peptides. The use of microemulsions as a polymerisation template also renders this process suitable for industrial scale-up.

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