Poster Session 1 – Pharmaceutics

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The effects of drying process on the properties of pellets and their compacts

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Satisfactory pellet formation by extrusion and spheronisation process occurs as a consequence of several carefully optimised processing stages that include drying. Drying is used in pharmaceutical manufacturing as a unit process mainly to ensure the stability of the final products. As fibrous material microcrystalline cellulose can hold water by physically entrapping in fine capillaries and small internal pores. Moisture movement can therefore be slow during the drying process, as the liquid has to diffuse through structural obstacles caused by the molecular configuration. Different drying techniques use different mechanisms to over come this obstacle, which can have some effects on the final product. Hence, the objective of this work was to compare the effects of four different drying techniques on the properties of the microcrystalline cellulose pellets prepared by extrusion and spheronisation and their compacts.

Based on the different rate of moisture removal and means of heat and mass transfer the different drying techniques produced pellet of different properties (Table-1). The most crucial of these was the porosity, which resulted from the different extent of shrinkage of the pellets. The rapid evaporation of water as a result of the turbulent motion of the fluidized pellets (fluid-bed) and the sublimation of the expanded ice (freeze-drying) suppressed the shrinkage of the pellets during drying to produce pellets of higher porosity and of greater mean diameter. The static and slower drying nature of the oven and desiccation with silica gel produced pellets of smaller mean size and lower porosities. A fixed mass of pellets was compacted to a constant thickness at a rate of $0.5\,cm\,min^{-1}$ in a 12.0-mm diameter die with flat-faced punches.

Table 1 The effects of different drying techniques on the mechanical properties of pellets and their compacts

Drying	Freeze-dried	Fluid-bed dried	Oven	Desiccation
ε (%)	31.60 ± 1.58	16.40 ± 0.82	12.50 ± 0.63	13.80 ± 0.69
σ_p (MPa)	1.71 ± 0.17	3.33 ± 0.33	5.55 ± 0.56	6.09 ± 0.61
d (mm/kN)	20.79 ± 1.04	11.34 ± 0.57	7.33 ± 0.37	7.77 ± 0.39
P (MPa)	119.76 ± 5.99	114.5 ± 5.73	112.82 ± 5.64	97.85 ± 4.89
σ_t (MPa)	1.185 ± 0.18	0.222 ± 0.03	0.054 ± 0.01	0.063 ± 0.01
VER (%)	16.45 ± 3.29	12.62 ± 2.52	12.62 ± 2.52	8.40 ± 1.69

ε, porosity; σ_p and d, tensile strength and deformability of the pellets respectively; σ_t and VER, tensile strength and volumetric elastic recovery of the tablets; P, compaction pressure

The tensile strength, σ_p, of the pellets increased with decreasing porosity, ε, while the deformability, d, of the pellets and volumetric elastic recovery, VER, of the compacts increased with porosity. The pellets with a large porosity needed a higher compressing pressure, P, to produce tablets of the same mass and dimensions. The drying techniques, which produced porous, deformable and weak pellets, produced stronger tablets, σ_t, as measured by diametral compression Fell & Newton (1970). The value of the VER of the compacts was also observed to increase with the value of d.

Fell, J. T., Newton, J. M. (1970) *J. Pharm. Sci.* **59**: 688–691

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Sustained-release formulation of salbutamol sulfate by using pellets

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One of the most important research aspects in pharmaceutical sciences, is to optimize pharmaceutical products and formulations (Swarbrick & Boylan 1995). To achieve the best drug effect, the least side effects and better acceptance of drug by the patient in drug therapy by most of the drugs, the best way is to use sustained-release formulation. In this respect, pelletization technique based on its capabilities as well as its unique advantages is of great importance to be taken into consideration (Sellassie 1987).

The aim of this study was to produce salbutamol sulfate pellets by using flow-coater machine and coating the pellets with Eudragit RS30D and RL30D and investigation of their characteristics.

Salbutamol sulfate (Ciba Geigy, Italy), Avicel (F.M.C., USA), Eudragit RS30D and Eudragit RL30D (Pohm Pharma, Germany), and dicalcium phosphate (Dairy Crest, UK) were used. Salbutamol sulfate powder ($< 100\,\mu m$) was mixed with Avicel and dicalcium phosphate. Then the mixture was used for the preparation of pellets using a spheronization method, in a way that each one gram of prepared pellets contained 40.16 mg salbutamol sulfate. Polymer solutions containing 10% RS30D, RL30D and 1:1 RS:RL dissolved in acetone were prepared. The polymer solutions were used for coating the pellets by flowcoater apparatus. Particle size distribution, bulk density, tap density, angle of repose and friability of the pellets were determined. A dissolution test was carried out using apparatus I, in 900 mL distilled water, and a rotation rate of $100\,rev\,min^{-1}$. The amount of 240 mg pellets (equivalent to 9.64 mg salbutamol sulfate), was used for the dissolution test.

The best formulation of pure pellets contained 25% dicalcium phosphate and 36% Avicel. The best formulation of salbutamol sulfate pellets contained 2.69% drug, 25.81% dicalcium phosphate and 35.89% Avicel. The results obtained from testing salbutamol pellets are summarized as the data below:

Each g of salbutamol sulfate pellets contained 40.18 ± 0.172 mg of salbutamol sulfate. Bulk density was 0.787 ± 0.012; tap density was 0.808 ± 0.116; Carr's index was (%) 2.6 ± 0.171; Hausner ratio was 1.026 ± 0.211; bulkiness was 1.269 ± 0.151; repose angle was 21.73 ± 0.021° and friability was 0.18 ± 0.041. Rlease pattern from salbutamol pellets without coating materials flowed Highuchi low. Pellets containing 7.4% RL 30D as coating showed similar behaviour to the uncoated pellets.

Pellets containing 7.4% RS:RL in the ratio of 1:1 showed zero-order kinetics.

It was found that non-coated pellets were slow release and their drug release followed Higuchi law. Pellets coated with Eudragit RS showed first-order release kinetics, and pellets coated with Eudragit RL, followed the Higuchi law.

Sellassie, I. G. (1987) *Pharmaceutical pelletiazation technology.* Vol. 37, Marcel Dekker Inc., New York, 187–216

Swarbrick, J., Boylan, J. C. (1995) *Encyclopedia of pharmaceutical technology.* Vol. 11, Marcel Dekker Inc., New York, 369–394

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Effect of moisture on poly(vinylpyrrolidone) and the implication for accelerated stability testing

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The in-vitro dissolution profile for oral solid dosage forms is a frequently used tool for ensuring the consistent in-vivo performance of a formulation. For Class I compounds, defined as high solubility, high permeability, in the Biopharmaceutics Classification System, an in-vitro dissolution of 85% drug release after 15 min is deemed sufficient to waive the requirement for in-vivo bioequivalence testing. The use of accelerated stability studies is a common approach for predicting the long-

term physical and chemical stability of pharmaceutical formulations. In this study, a slowing of dissolution was observed for a formulation following storage at elevated temperature and humidity. In contrast to this, no change was observed on subsequent stability testing at long-term stability conditions.

For tablets prepared with PVP as a binder, there was a clear reduction in the dissolution rate following storage at elevated temperature and humidity (Table 1). In contrast, a formulation in which the PVP was replaced with HPMC as the binder did not show any slowing of dissolution following similar storage (Table 2). The moisture sorption isotherm for the binder in the PVP system shows that this material absorbs a significant quantity of water on exposure to elevated humidity. Modulated DSC has been used to demonstrate that this moisture uptake will depress the glass transition temperature (Tg) of PVP to below the conditions used in the accelerated stability study. In the case of HPMC, a classical glass transition was not observed under these conditions, which would explain why the elevated temperature and humidity had no impact on the dissolution performance.

Table 1 Dissolution following 6 months stability of PVP based formulation

Condition (°C/%RH)	% Dissolved		
	10 min	15 min	30 min
5/50	62	79	94
30/60	60	79	95
40/75	33	48	68

Table 2 Dissolution following 6 months stability of HPMC based formulation

Condition (°C/%RH)	% Dissolved (range)		
	10 min	15 min	30 min
5/50	91	97	100
30/60	95	99	100
40/75	93	98	100

During this stability study, exposure to elevated temperature and humidity would have resulted in a physical change in the PVP whereby it is converted from the glassy to the rubbery state. It is proposed that it is this conversion of the PVP that produces the observed change in the dissolution profile. Furthermore, long-term stability studies conducted at temperatures and humidities below the Tg, would not have induced this change.

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Effect of punch surface quality on the sticking tendencies of ibuprofen tablets

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Methods employed to quantify sticking problems during tableting include measurement of the tablet surface roughness (Toyoshima et al 1988) and of the adhesion force as the upper punch is detached from the tablet (Waimer et al 1999). The importance of punch face roughness on sticking has not extensively been studied.

The aim was to assess sticking to the upper punch face. Three formulations were used: 29.5%:69.5%, 49.5%:49.5% and 69.5%:29.5% ibuprofen B.P.:lactose DC (Tablettose 80, Meggle), respectively. Each contained 0.5% Aerosil 200 (Degussa Ltd) and 0.5% magnesium stearate (BDH). Compaction was performed at 10 kN or 40 kN using a Manesty F3 single-punch tablet press. Each compaction run was 1 min. Two sets of 12.5-mm flat-faced punches were used. Surface profiles (Taylor Hobson Form Talysurf 120, Liverpool John Moores University) of the upper punch indicated a large difference in quality. The punches were classified as old (Ra = 0.33 μm) or new (Ra = 0.05 μm) where Ra is the mean of all positive deviations from zero. Powder from the punch barrel was removed and the punch face immersed in 5 mL 96% ethanol; ibuprofen attached to the face was quantified by spectroscopy at 264 nm (Table 1).

Table 1 Sticking of ibuprofen (μg ± s.d.) to the upper punch face (n = 25 for each data set)

	Old punch			New punch		
Formulation	i	ii	iii	i	ii	iii
10 kN	79 ±14	169 ±26	430 ±50	44 ±15	71 ±17	149 ±43
40 kN	86 ±16	208 ±34	311 ±63	69 ±26	275 ±65	1031 ±286

Statistical analyses showed that surface roughness, compaction force and the blend composition were all individually significant ($P < 0.05$) factors in sticking. However, 3-way analysis of variance showed that the interaction of all factors was significant ($P < 0.05$) and, therefore, sticking was inter-dependent on all parameters studied.

The greatest degree of sticking occurred with the new punch at 40 kN with formulation iii. More sticking might be anticipated with the punch with poorer surface quality. An explanation for this apparent anomaly may be that the imperfections in the old punch surface provided areas that disrupted the force of attraction between the tablet and punch surfaces. The lack of imperfections in the new punch face did not allow the force between tablet and punch to be disrupted, increasing the amount of the tablet sticking to the punch face. This possibility was suggested by Schumann & Searle (1992).

Schumann, S., Searle, G. D. (1992) *Drug. Dev. Ind. Pharm.* **18:** 1037–1061

Toyoshima, K., Yasumura, M., Ohnishi, N., et al (1988) *Int. J. Pharm*aceutics **46** 211–215

Waimer, F., Krumme, M., Danz, P., et al (1999) *Pharm. Dev. Tech.* **4:** 359–367

060
Relationship between powder cohesion and tablet tensile strength

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Compactibility is the ability of a powder to cohere together during compression to attain a specific tensile strength (Fell 1996). Nystrom et al (1993) suggested that tablet tensile strength is related to the strength of interparticle forces (cohesion and adhesion) and the surface area over which they operate and that bond surface area is influenced by the yield pressure of the material. This paper studies the relationship between particle cohesion and tablet tensile strength using powder rheometry.

Riboflavin phosphate 3% w/w was wet granulated with 66.8% w/w lactose monohydrate, 2% w/w croscarmellose sodium, 5% w/w PVP K30 and 23.2% w/w of Avicel (PH101, 103, 112, 113, 200 or 105). Cohesivity of the dried granule and each individual grade of Avicel were separately measured using a Manumit powder rheometer (200 mL glass vessel, 48 mm diameter blade, 50 mm s^{-1} blade speed, 5° helix angle downward compaction cycle, 178° helix angle upward aeration cycle). The cohesion coefficient was calculated by measuring the area under a force-displacement plot of the aeration cycle. The tensile strength of tablets compacted on an ESH compaction simulator at five pressures (1.5–370 MPa) and a speed of 3 mm s^{-1} were measured. Yield pressures were determined using Heckel analysis.

At any given compression pressure, the tensile strength of tablets compressed from the individual Avicel powders and riboflavin phosphate granules containing Avicel, were higher with increasing cohesion coefficient.

This study suggests that for a series of test systems, tablet tensile strength may be predicted from the cohesion coefficient of the powder. The yield pressure of the powders were similar (119 MPa to 129 MPa), suggesting that bond surface area effects will be negligible and that tablet tensile strength for Avicel and riboflavin

phosphate granules may be determined predominantly by the cohesive forces between particles.

In this system, the changes in powder cohesion can be correlated with the change in tablet tensile strength. If these findings can be replicated across a wider range of systems, cohesion measurements by powder rheology may complement compaction simulation in powder characterisation. This could offer the possibility of at-line study of batch variation prior to compression.

Fell, J. (1996) Compaction properties of binary mixtures. In: Alderborn, G., Nystrom, C. (eds) *Pharmaceutical powder compaction technology.* Marcell Dekker Inc., New York

Nystrom, C., et al (1993) *Drug. Dev. Ind. Pharm.* **19**: 2143–2196

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The effect of tooling geometry on the tensile strength of tablets

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Pharmaceutical tablets come in a wide range of shapes and sizes. Investigating the effects of different shapes of tablet tooling on the properties of the tablets produced will help to understand why some formulations are difficult to produce in non-standard shapes.

Five flat-faced punch and die sets were used, (I. Holland, Nottingham, UK) having dimensions of 10 mm square with corner radii ranging from 1 to 5 mm in 1-mm steps. Tablets were manufactured using a compaction simulator (ESH Testing Ltd, Dudley, UK) following a single-ended saw-tooth profile. The fill depth (6 mm), compression height (3 mm) and speed (3 mm s^{-1}) were kept constant. The average density was therefore similar. The material compressed was Ludipress (BASF, Germany). Magnesium stearate 0.5% w/w was added for lubrication. A number of tablets were made and five tablets of each shape were tested. Their weight and dimensions were recorded. The tablets were compressed along their diagonal using a hardness tester (Schleuniger Model 4D, Pharmatron, Germany) and the fracture force recorded. The tensile strength (σ_t) was calculated from the force needed to fracture the tablet (F_t) and the cross sectional area of the fracture plane (A_t) according to the following equation:

$\sigma_t = F_t/A_t$ (Olsson & Nyström 2001)

Table 1 Summary of tablet data (n = 5)

Corner radius	Fracture force F_t (N)	Tensile strength σ_t (N mm^{-2})
1 mm	77.8 ± 5.2	0.509 ± 0.033
2 mm	72.0 ± 4.5	0.491 ± 0.030
3 mm	93.0 ± 7.2	0.641 ± 0.048
4 mm	94.8 ± 3.4	0.691 ± 0.023
5 mm	103.6 ± 8.5	0.862 ± 0.069

The round tablets showed a higher tensile strength (Table 1) than the square-shaped tablets. An attempt was made to attribute these effects to the properties recorded by the compaction simulator during compression. However, analysis of the peak forces, punch displacements and calculations of power, elastic recovery and work done, failed to show any correlation. Calculation of the average density from the thickness, punch area and weight showed the tablets were of similar densities. The effects seen may indicate that there are differences within the internal structure of the tablets, across the shape range, leading to differences in distribution of the stresses and crack propagation during crushing.

In conclusion, changing the geometry of the tablet shape from round to square has marked effects upon the tensile strength of the tablets produced, which cannot be accounted for by differences in the mechanics of the compaction event.

Olsson H, Nyström C. (2001) Assessing tablet bond types from structural features that affect tablet tensile strength. *Pharm. Res.* **18**: 203–210

062
Characterisation of wet masses using a powder rheometer

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Quantitative understanding of granulation and of variables that impact granulation behaviour, has increased enormously over the last decade (Faure et al 2001). Introduction of the mixer torque rheometer has fuelled research, such that end-point is no longer difficult to determine, and consistency of wet mass to achieve satisfactory final product, easier to predict. The aim of this study was to evaluate a powder rheometer (FT3 Powder Rheometer, Freeman Technology, UK) to assess rheological properties of wet masses processed using large-scale equipment; to investigate whether data could be used to predict the quantity of granulating liquid required to produce optimum wet granule and final tablets with appropriate properties.

Haloperidol masses to manufacture 5 mg tablets were produced and end-point recorded. Multiple 200-g samples were removed from each batch and moisture contents determined. Tests performed with the FT3 included a standard compaction test and piston compaction of the mass followed by the blade used with a shearing action. Granules were dried at 65°C, milled, lubricated and tabletted at a range of compression forces. The data for tablets prepared from 4 different wet masses at two compression forces is shown (Table 1). Experiments using masses produced with various process parameters on small-scale equipment, have also been conducted.

Table 1 Energy values of wet masses, properties of tablets

	Water content (% w/w)	Energy (mJ)	CF (KN)	H (kP)	DT (s) (n = 12)
A	12.3	629.6 (22.3%)	22.4	4.3	128.4 (12.7)
B	11.7	311.9 (9.4%)	22.6	3.8	103.2 (10.6)
C	9.6	165.7 (3.4%)	19.2	3.0	61.8 (11.9)
D	7.4	101.1 (6.2%)	22.9	2.9	16.9 (2.7)
B	11.7	311.9 (9.4%)	12.3	2.6	50.9 (10.2)
C	9.6	165.7 (3.4%)	14.4	2.1	36.1 (6.2)
D	7.4	101.1 (6.2%)	13.6	2.0	28.0 (8.9)

CF = compression force, H = hardness, DT = disintegration time

Total energy results show a high sensitivity to water content indicating the rheometer can discriminate between samples that differ by less than 1%w/w water. Plots of energy versus water content for various blending times were constructed and used to accurately determine water contents of masses based on rheological behaviour. Energy profiles indicate that at low water content more energy is required when mixed for shorter times, and as water increased to 12%w/w the energy requirement increased with longer mixing times.

Table 1 shows there is a rank order with regards water content, energy requirement for blade to traverse mass and tablet hardness and disintegration time for both a high (22 KN) and low (12 KN) compression force. A comparison of data between the two compression forces demonstrates that tablet hardness and disintegration times do not always correlate with respect to water content. This would suggest that different bonding mechanisms in tablets result from increased water content of granules than those that result from compression.

The study demonstrated the ability of the rheometer to determine the relationship between flowability characteristics and proportions of granulation liquid, and there is potential to detect optimum levels of liquid to produce suitable final product.

Faure, A., et al (2001) *Eur. J. Pharm. Biopharm.* **52**: 269–277

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Density distribution and surface topography of flat-face tablets

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As a powder is compacted in a die, various zones are subject to differing intensities of pressure and shear. This is due to the frictional properties of the powder and tooling and will result in differing density and flaw distributions within the tablet. These resulting inhomogeneities give rise to the potential for subsequent poor tabletting performance such as capping and lamination. Previous workers have used a number of techniques to characterise the density distribution seen in compacts, such as turning, measuring and weighing (Train 1957), autoradiography (Macleod & Marshall 1977) or NMR microscopy of tablets (Nebgen et al 1995). In this investigation, the density distributions in microcrystalline cellulose tablets were determined experimentally using a coloured layer technique. Non-destructive topography measurements were taken to assess the variation in surface roughness of the tablets and relate this to the forming pressure and density distribution.

Weighed amounts of alternate-coloured layers of microcrystalline cellulose alone and of microcrystalline cellulose coloured with 1% Black iron oxide were filled into a 25-mm unlubricated die fitted with flat face punches and then uniaxially compressed to various compaction pressures between 6 and 200 MPa using an automated instrumented press. Dwell time and punch speed were kept constant. The compacts were then ejected and cross-sectioned. The thickness of the exposed layers as a function of the radius and of the tablet height were determined by a digital imaging system to obtain the compact density distribution within a section. These values were then combined to give 2-dimensional plots of density distribution across the tablet. To confirm that the coloured and white powders have the same compressional properties, single-layer tablets were compressed from both. Results showed that there were no significant differences between the densities of the resulting compacts.

High areas of density were found at the top corners and middle bottom halves of the layered tablets, with low density being found in the bottom outer circumferences of the compact. The density decreased down the sides of the tablets. The data also showed that the density distributions increased as the applied compaction pressure increased, with the overall difference of maximum to minimum density remaining fairly constant at 30% over the pressure range examined. Surface topographies of the top, bottom and sides of the compacts at the different compaction pressures were then obtained by using a laser proximity sensor to obtain 3-dimensional images of surface roughness images. Surface roughness values were then related to the applied pressure.

The variation in pressure could then be determined across the tablets with the conclusion that the final density distribution could be correlated with the pressure distribution occurring during compression.

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064
Investigation of protein denaturation upon spray-drying by differential scanning calorimetry

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Increasing interest in developing pulmonary delivery systems for proteins and peptides has led to the proposal that spray-drying may provide one of the principal means of producing inhalable powders of the labile biopharmaceuticals (Broadhead et al 1994). However, the processes involved in spray-drying impose potential stresses on the product, including exposure to high temperatures, distribution of air-water interfaces and dehydration. Any of these could denature proteins and thus compromise the biological activity (Tzannis & Prestrelski 1999). To preserve the native structure of proteins, disaccharides such as sucrose or trehalose are generally included as stabilisers. In addition, it is preferable to determine the integrity of processed proteins in the dried form since the denatured states have limited storage stability and may be unable to be detected after immediately rehydration due to their partial or complete reversibility. FTIR spectroscopy is the most commonly utilised method for such a purpose, although FTIR is limited for the evaluation of secondary structure (Dong et al 1992). In this study, the aim was to investigate the feasibility of using the denaturation/melting enthalpy (ΔH_m) of the model protein lysozyme, obtained by differential scanning calorimetry (DSC) to evaluate the integrity of the native structure.

Lysozyme samples were spray-dried with or without excipient using a Model 190 Buchi mini spray-dryer. Second derivative FTIR spectra and DSC were employed to determine the secondary structure and the integrity of the native structure of spray-dried lysozyme. Spray-dried lysozyme in the absence of stabiliser underwent a sizeable perturbation of the structure as indicated by 2nd derivative FTIR spectra and DSC analysis. The intensity of the α-helix band in the amide I region of spray-dried lysozyme obtained from FTIR was only 60% of that exhibited by the native protein in the liquid form, whilst the ΔH_m as measured by DSC in the dried form was $12.98 \pm 0.98\,J\,g^{-1}$, only about one-third of $33.61 \pm 0.72\,J\,g^{-1}$ of the native protein in the liquid state.

Inclusion of sucrose or trehalose to the lysozyme solution was found to stabilise spray-dried lysozyme as indicated by the intensity of α-helix band obtained from FTIR and the ΔH_m measured by DSC, with the protective capacity of each excipient found to be a function of the sugar concentration. In addition, the stabilising profiles by both sucrose and trehalose determined by either DSC or FTIR were similar. Indeed, ΔH_m was found to be a linear correlation between the intensity of α-helix band and ΔH_m for both sucrose ($R^2 = 0.990$) and trehalose ($R^2 = 0.997$). Such results indicated that ΔH_m is applicable to evaluate the stabilisation of proteins in the dried form. In addition, the stabilisation of lysozyme led to increases in ΔH_m of ~280% (from $12.98\,J\,g^{-1}$ to $36.89\,J\,g^{-1}$), while the corresponding increases in the intensity of FTIR was only ~134%. In other words, DSC may contain a high precision relative to FTIR to determine the content of the native structure preserved after processing.

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Dong, A., et al (1992) *Biochemistry* **31**: 182–189
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065
Dissolution and mechanical properties of nifedipine pellet prepared by pan-layering

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A prolonged-release form of nifedipine pellets (30 mg), made up of a particulate core, a nifedipine coating layer which covers the particulate core and a surface coating layer which covers the nifedipine coating layer, was invented. A conventional pan coating method was utilized to prepare the nifedipine sustained-release coated beads. The layer was applied on the particulate core and was comprised of excipients which can be water-soluble or water-insoluble, and were obtained by pan coating technique. The prepared pellets were subjected to physicochemical tests and physicomechanical investigation (e.g. tensile strength, Young's modulus, particle size measurement by sieve analysis and shape analysis by scanning electron microscopy (SEM)). Drug release profiles were achieved by using USP apparatus No. 1 (rotating basket) in different dissolution media including GF with and without pepsin and buffer phosphate consisting 1% SLS. The formulated nifedipine pellets demonstrated slower and prolonged dissolution profile than a similar tablet of Slodipine (Table 1). The mechanical strength of prepared seeds and nifedipine pellets showed good resistance by increasing core diameter from 0.8 mm to 2.0 mm. The core diameter was optimized at 1.0 mm. Meanwhile, the comprised component of the cores showed significant effect on the mechanical characteristics. Size and shape analysis determined the suitability of

pelletization technology in producing narrow size distribution and spherical shape for dosage forms. These obtained results were proved by statistical parameters.

Table 1 Nifedipine release from tablet and pellet dosage forms in the medium of phosphate buffer containing 1% SLS

Time (min.)	Drug release (%)	
	Tablet	Pellet
30	0.34	7.74
60	0.50	14.17
120	1.64	19.90
240	3.41	29.65
360	5.73	42.92
480	7.96	54.16
1260	35.82	86.20
1320	38.00	86.70
1380	40.37	87.00

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Preparation of non-pareil seeds (NPS) by pan layering method

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Interest in using spherical substrates for oral dosage forms is now growing, with an increasinge desire to offer products that have better controlled-release characteristics. Small coated spheres of active ingredients are the most popular way to deliver drugs in a controlled-release format. Making these spheres round, smooth and uniform from powders can sometimes be the most difficult part of development or production (Ghebre-Sellassie 1989; Zhang et al 1990).

A popular alternative to sphere formation from powders is to layer the drug onto a starting core by spray coating. Uniformity of size and the sphericity of prepared seeds, not only allow for more accurate and consisting coating of granules, but also provide for more consistent dissolution of active ingredients (Sherington & Olivet 1981; Raman et al 1993).

Conventional pan coating method was utilized to prepare non-pareil seeds. A known amount of sugar (20–25 mesh) was held in the pan. PVP was dissolved in isopropanolol and then sprayed on the sugar in the rotating pan (at 30 rev min^{-1}). The processes of wetting with binder, dusting with talc and drying the seeds were repeated until the seeds were built to the desired size. The seeds were then screened for particle size distribution using sieve analysis.

A universal material testing machine mounted with a suitable load cell was used to test physicomechanical properties of seeds. The physicomechanical parameters assessed for 1.0-mm microsphers are shown in Table 1.

Table 1 Physicomechanical parameters for 1.0-mm non-pareil seeds

No.	E-modulus ($N mm^{-2}$)	Rm (N)	W_{Rm} (J)	εFmax (%)	σ_t (N)
1	11880	8.18	0.000285	0.15	20.84
2	6623	7.04	0.000234	0.15	17.93
3	13233	7.76	0.000229	0.13	19.77
11	6126	7.22	0.000209	0.14	18.41
12	9308	8.76	0.000223	0.12	22.32
χ	8301	7.98	0.000242	0.14	20.33
A	2183	0.77	0.000382	0.01	1.97
v	26.30	9.70	15.97	9.57	9.700

E-modulus, Young's modulus of elasticity; σ_t, tensile strength Rm, W_{Rm}, εFmax, maximum stress, work upto maximum load and deformation up to maximum force, respectively

Scanning electron microscopy was used to evaluate surface shape of the microspheres prepared under the various conditions. The prepared seeds showed good resistance of mechanical strength, and size and shape analysis determined the suitability of pan layering method in forming microspheres.

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067

The effect of crystal habit on milling induced crystallinity changes

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Milling is a key process in the preparation of an oral dosage form. One possible milling-induced change is the production of small levels of amorphous material found predominantly at the surface of a powder (Krycer & Hersey 1980), which could lead to significant chemical and physical instability. The influence of crystal habit on this change is investigated using succinic acid as a model compound.

During crystal growth, heterogeneous solvent adsorption can produce change in crystal habit. Plate-like and needle-like crystals of succinic acid were produced by crystallisation from water and isopropanol, respectively. Both materials were subjected to at least 12 h drying in a vacuum oven at 40°C and stored over silica gel before any milling was commenced. Milling was performed in two mills: a ball mill (BM) (for 2 h) and a jet mill (JM). Characterisation was performed using powder X-ray diffraction (PXRD) (Siemans D5000) and solution calorimetry (Tronac 459 isoperibol calorimeter). All experiments were performed in triplicate. PXRD diffraction patterns of the milled materials were similar to the unmilled, indicating no gross change. While changes in the peak intensities of the milled samples compared with the unmilled samples were observed, these were deemed minor and difficult to quantify. Solution calorimetry was sufficiently sensitive to detect that all milled samples had reduced heats of solution (ΔH_s) (Table 1), a characteristic of a material that has lost crystallinity (Hogan & Buckton 2000).

Table 1 ΔH_s of milled and unmilled materials together with their respective changes in crystallinity

	ΔH_s (kJ mol^{-1}) (n=3)	Percentage change
Plates	+27.21 ± 0.77	
Needles	+27.14 ± 0.27	
BM plates	+25.65 ± 0.03	−5.23
BM needles	+26.78 ± 0.22	−1.32
JM plates	+26.41 ± 0.31	−2.95
JM needles	+26.64 ± 0.19	−1.84

BM = ball milled, JM = jet milled

From Table 1, the greatest change was seen in BM plates, which decreased by 5.23%, compared with the BM needles which had decreased by 1.32%. Similarly the JM plates were found to have a greater drop in their heats of solution when compared with the JM needles and an implied greater loss of crystallinity. The data indicate that, in the case of succinic acid, crystal habit is one factor influencing the susceptibility of a material to crystallinity change on milling, as demonstrated by plates exhibiting greater changes than needles. The use of solution calorimetry to detect crystallinity change on milling is also highlighted.

In conclusion, succinic acid crystals with plate-like morphology appear to be more prone to crystallinity loss on milling compared to the needle-like morphology. Such findings suggest that habit modification is a valid approach to overcome process-related problems of pharmaceutical materials.

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068

The rheological evaluation of polymers in optimization of nasal gel sprayability

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Absorption from the nasal route has been studied widely to improve drug bioavailability. It has been shown that the spray form is one of the most suitable systems for this purpose. Deposition pattern and residence time of nasally administered drugs are two of the most important factors which affect the therapeutic efficiency. On the other hand droplet size distribution and viscosity of the prepared formulations alter these factors predominantly (Hughes et al 1993).

Development of a sprayable gel formula which causes longer residence time of drug in the nasal cavity would be a potential way to improve effectiveness especially for those drugs that exhibit poor absorption like peptides and proteins.

In this study, prepared gels from four different polymers were evaluated for possible correlation of rheological properties with sprayability. Different concentrations of polymers were used to achieve the same viscosity (about 2000 cps). The rheological study was carried out using a Brookfield Viscometer Model DV-I+ (Brookfield, US) at 25 $\pm$ 1°C and 34 $\pm$ 1°C (temperature of nasal mocusa). The results of rheological study are shown inTable 1.

Table 1 Viscosities (cps) of four different polymers at 25°C and 34°C

RPM	0.3	0.6	1.5	3	6	12	30	60
Carbopol[a]								
(25°C)	104000	58000	28080	16800	10240	6220	3240	1992
Na CMC[b]	9200	7200	6400	5600	4660	3740	2608	1904
MC[c]	4400	3000	2560	2400	2280	2190	2056	1942
HPMC[d]	4400	3400	2960	2800	2660	2490	2200	1922
Carbopol								
(34°C)	71600	41400	22640	14400	9260	5860	3104	1902
Na CMC	6000	5200	4480	4080	3480	2860	2072	1542
MC	3200	2000	1680	1560	1500	1440	1388	1322
HPMC	3200	2200	1920	1800	1700	1610	1468	1314

[a]Carbopol 934P (0.4%); [b]sodium carboxymethylcellulose 5000 (0.9%); [c]methylcellulose 400 (3.1%); [d]hydroxypropyl methylcellulose 4000 (1.6%)

The resulting rheograms show that Carbopol gels provide plastic flow curves whereas Na CMC, MC and HPMC gels provide pseudoplastic flow at both temperatures. Carbopol gel has extremely higher apparent viscosity at low shear rates in comparison with other polymers also its viscosity is less affected by temperature. Based on this reasoning, among the mentioned polymers Carbopol gel would causes a longer residence time in contact with nasal mocusa.

To study the sprayability of different gels, samples were filled into bottles with metered spray pumps (Erich Pfeiffer, GmbH). Carbopol gel was the only one which was sprayable as uniform droplets. It had been shown that Carbopol gels with a viscosity up to 5000 cps could be sprayed (Moslemi et al 2002), whereas other polymers show poor sprayability properties even in viscosities less than 50 cps. It seems that this is related to rheological properties of the polymers which is explained above.

Further investigation should be carry out to study the rule of rheological properties in characterization of a nasal gel spray.

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069

Mechanical properties of spray-coated layers on solid-core microcapsules

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Microcapsules used for oral administration of medication essentially consist of one or more drugs in a liquid or solid core enclosed with an outer coating, with a diameter in the region of 1–2000 μm. They are often employed in drug delivery to provide controlled or modified release of the drug.

Microcapsules were manufactured consisting of a cellulose core (385 μm diameter) surrounded by a drug-loaded layer which in turn was enclosed with a diffusion membrane to give finished microcapsules with a typical diameter of 500 μm. The layered core contained up to 10% (by weight) of active material and was spray-coated onto the cellulose spheres using water as a suspending medium. The composition of the outermost layer could be altered to give modified-release profiles and was applied using either aqueous or organic solvents.

The study attempted to measure differences in mechanical properties of the outer layers arising from the two different manufacturing processes. Such information could prove useful for the purposes of quality control and processing. The micromanipulation technique (Zhang et al 1991) was used to examine these microcapsules.

The technique is based on the compression of a particle between two notionally infinite parallel plates and has previously been used to study both organic and inorganic materials, including microcapsules with liquid cores (Sun & Zhang 2001). In such instances, the technique relied on compression of the particle until breakage. Given that the coatings were of interest and their small thickness, the microcapsules were not subjected to compression until rupture, but rather strains of up to 20%.

As stresses and strains were not uniform across each individual microcapsule, data were obtained in the form of pseudo-stress–strain curves taking into account the diameter of the particles. From the initial slopes of the pseudo-stress–strain curves, it was possible to determine pseudo-elasticity moduli (Table 1).

Table 1 Pseudo-elasticity moduli (MPa) for cellulose cores and coated microcapsules

Cellulose cores	Active aqueous-organic	Active all-aqueous	Placebo all-aqueous
4.19	3.53	3.97	1.99

Placebo microcapsules required much smaller forces to achieve given deformations, with a pseudo-elasticity modulus 40–50% less than that of the microcapsules with drug-loaded layers. Given that the aqueous and organic coating processes used different materials, it was expected that the microcapsules produced would have different material properties. However, as Table 1 shows, this was not the case and microcapsules produced using the organic and aqueous processes have similar material properties.

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070

The influence of protein concentration on the activity recovery of freeze-dried LDH under different storage conditions

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Protein therapeutics are often prepared in a solid form to overcome their limited physical and chemical stability. Lyophilisation (freeze-drying) is one of the most common methods for manufacturing proteins in powder form. However, this

process generates a variety of freezing and drying stresses (e.g. solution concentration, pH changes, etc.) that can denature the protein to various degrees (Franks 1993). Even protein preparations successfully freeze-dried can undergo different degradation pathways during storage (Wang 2000). In this study the stability of lyophilised lactate dehydrogenase (LDH) was investigated under different storage conditions. LDH, a tetrameric enzyme, was chosen as the model protein because of its high sensitivity to freeze-drying stresses, particularly at low concentrations (Jiang & Nail 1998). Two solutions at 5 and 50 $\mu g\,mL^{-1}$ of LDH in phosphate buffer (pH 7.4, 0.05 M) were analysed. Phosphate buffer was selected to exacerbate the freeze-drying stress, due to the well-documented pH drop from 7.4 to 4.5 during freezing; no lyoprotectants were added (Wang 2000). Mannitol (5% w/v) was used as caking agent to avoid any loss of the solutes during freeze-drying. The solutions were evaluated after freeze-thawing and freeze-drying. Freeze-thawed samples were frozen in a −20°C freezer. Freeze-dried samples were prepared using a shelf temperature −20°C during primary drying and +25°C during secondary drying with a pressure of 100 μbar throughout the process. The freeze-dried samples were stored for 10 days at storage temperatures of –20, 4, 25 and 60°C. Samples were analysed on day 0, 6, and 10. Results are summarised in Table 1.

The data indicate that most of the enzymatic activity loss is due to freezing induced stress and only partially due to drying. Additionally, the protective effect of the higher concentration is notable under all storage conditions. There is no major difference in storing samples at –20, 4, and 25°C but the incubation at 60°C completely inactivates both formulations of LDH, with the formulation at $5\,\mu g\,mL^{-1}$ being denaturated more rapidly then the one at $50\,\mu g\,mL^{-1}$. This study highlights the importance of protein concentration on activity recovery and sets the basis for further investigations on the protective capability of excipients.

Table 1 Stability of lyophilised LDH

Condition (days)	LDH residual activity (%)	
	$5\,\mu g\,mL^{-1}$	$50\,\mu g\,mL^{-1}$
Solution refrigerated (6)	50	64
Solution refrigerated (10)	43	50
Freeze-thawed solution	23	59
Freeze-dried (0)	10	40
FD stored at 25°C (6)	9	35
FD stored at 25°C (10)	7	33
FD stored at −20°C (6)	14	40
FD stored at 4°C (6)	8	34
FD stored at 4°C (10)	4	27
FD stored at 60°C (6)	0	3
FD stored at 60°C (10)	0	0

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071

Physical characterisation of bioadhesive ternary component semi-solid systems designed as platforms for drug delivery to the periodontal pocket

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It is accepted that the rheological and mechanical properties of periodontal drug delivery systems are direct determinants of the clinical performance (e.g. influencing the ease of administration into and retention within the periodontal pocket and the rate and mechanism of drug release (Jones et al 1999, 2000)). In recent reports, the successful clinical uses of bioadhesive polymer networks composed of three polymeric components, hydroxyethylcellulose (HEC), polyvinylpyrrolidone (PVP) and polycarbophil (PC) and containing either tetracycline or flurbiprofen were described (Jones et al 1999, 2000). However, a fundamental understanding of the rheological properties of these systems is required to optimise their clinical performance. Therefore, in this study the rheological and mucoadhesive properties of bioadhesive semi-solids containing HEC, PVP and PC are described.

Semisolids containing HEC (3% w/w), PVP (1–5% w/w) and PC (1–5% w/w) were prepared as previously described (Jones et al 2000). Dynamic (oscillatory) and continuous shear (flow) rheology was performed using a Carri-Med CSL^2-100 rheometer with a 2-, 4- or 6-cm diameter parallel plate geometry and a 1-mm plate gap, as previously reported (Jones et al 2001). The mucoadhesive strength was determined by measuring the force required to detach the formulations from a mucin disc using a texture analyser as previously described (Jones et al 1999). Results are shown in Table 1. The effect of each polymeric component on the storage modulus (G'), loss modulus (G''), loss tangent (tan δ), hardness, compressibility, adhesiveness and mucoadhesive strength were statistically examined using a three-way analysis of variance in conjunction with a Tukey's HSD post hoc test. In all cases $P < 0.05$ denoted significance.

Table 1 Mucoadhesive strength (M) and storage modulus (G') of gel systems

HEC/PVP/PC	M (N)	G' (1 Hz) (Pa s)
3/1/1	0.18 ± 0.01	712.31 ± 62.14
3/1/5	0.29 ± 0.02	4494.02 ± 128.12
3/3/1	0.19 ± 0.02	954.72 ± 23.21
3/3/5	0.33 ± 0.03	4441.67 ± 112.78
3/5/1	0.22 ± 0.02	1043.83 ± 64.21
3/5/5	0.39 ± 0.01	4672.33 ± 321.92

Increasing the concentration of each polymeric component significantly increased the rheological and textural parameters, with the exception of tan δ, which decreased. This may be accredited to increased polymer entanglement and the effects of dispersed PC on the elastic properties. Furthermore, as the concentration of HEC, PVP and PC increased there was an increase in mucoadhesion, consistency and the pseudoplasticity, evident from the increased gel strength (308.87–1720.51 Pa s^n) and decreased flow behaviour index (0.13–0.34). As the concentration of PVP or HEC increased the amount of free water decreased and hence the swelling of PC decreased. This increase in the relative amount of unswollen PC increased the mucoadhesive strength and the rheological/mechanical properties and may be attributed to the increase in the number of free (uncharged) carboxylic acid groups associated with increased concentrations of HEC and PVP. The mucoadhesive strength and rheological parameters increased as the physical state of PC changed. The wide ranges of mucoadhesive strengths and rheological properties observed in this study suggest these formulations may be ideal candidates as bioadhesive platforms for the delivery of therapeutic agents to the periodontal pocket. In particular, formulations in which the swelling of PC is minimized (and hence mucoadhesion and rheological properties are enhanced) offer particular promise in this respect.

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072

An investigation into the interactions present within bioadhesive polymer gel systems using physical and spectroscopic characterisation techniques

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Bioadhesive/mucoadhesive formulations have gained increasing interest since they have the ability to adhere to mucosal surfaces thus increasing their therapeutic efficacy. It is common for bioadhesive formulations to contain more than one

polymeric component. These multi-component systems offer maximized potential as drug delivery platforms in comparison with mono-polymeric units. The interactions between polymeric components and or therapeutic entities are pertinent to the success of the drug delivery platform. Raman spectroscopy has previously been used to study the interaction between chlorhexidine (Jones et al 2000) and bioadhesive polymers incorporated into semi-solid formulations. This investigation has highlighted that when combinations of specific bioadhesive polymers are utilised, there is a change in the rheological, textural and release properties of the formulations. To investigate the cause of these changes, Raman spectroscopy was employed as a means of providing further information on intermolecular interactions present within the polymer matrix.

Polymer gels were manufactured by dissolving the required amount of HEC or Gantrez (or both) into a continuous aqueous phase with the aid of a mechanical stirrer. Dynamic, continuous shear rheology and texture profile analysis (TPA) was performed using a Carri-Med CSL^2-100 rheometer, as previously reported (Jones et al 2000). The effect of each polymeric component on the storage modulus (G'), loss modulus (G"), loss tangent (tan δ), hardness, compressibility, and adhesiveness were statistically examined using a three-way analysis of variance in conjunction with a Tukey's HSD post-hoc test. In all cases $P < 0.05$ denoted significance. Raman spectra were recorded at 785 nm using a backscattering geometry and a 45° mirror to focus the beam onto the horizontally mounted sample.

The combination of HEC and Gantrez resulted in increased formulation hardness, compressibility, storage modulus, loss modulus, and dynamic viscosity in comparison to the values obtained by the single component systems. These increases were attributed to hydrogen bonding between the two polymeric components. The Raman spectra obtained supported this theory with a shift in the Gantrez band at 1709 cm^{-1} to 1715 cm^{-1}. A shift in the Gantrez band at 1709 cm^{-1} in the gel to 1715 cm^{-1} in the solid may be attributed to hydrogen bonding between the Gantrez carbonyls and the hydroxyl groups on the HEC. This suggests that the H-bonding is mediated by water molecules, present within the gel formulations. It was proposed that this shift may be due to water of crystallization, so in an attempt to clarify the nature of this shift, dry virgin samples of Gantrez and HEC were also recorded. The spectra obtained were identical to the film forms of the monopolymers, proving that the shift in the carbonyl band may be attributed to intermolecular hydrogen bonding.

In conclusion, this study has identified that the change in the physical response of polymeric gels is attributed not only to polymer–polymer entanglements but also hydrogen bonding between the two components. This is of primary importance in the development of gel platforms for use as drug delivery systems, given that the physical properties determine the end use performance.

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073
Raft resilience in alginate anti-reflux products

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One of the primary treatments for gastro-oesophageal reflux disease (GORD) is the administration of alginate based anti-reflux preparations, which provide a physical barrier on contact with the stomach contents in the form of a neutral floating gel or raft. The strength of such a barrier is an important factor in its ability to resist reflux, because a strong cohesive barrier is more resilient and resistant to break-up with continued movement in the gut.

The aim of this study was to develop a reliable quantitative in-vitro measure of the ability of rafts to resist break-up in simulated gastric conditions, and to test this on leading UK alginate products.

Alginate rafts were prepared by dispersing the maximum product dose in 0.1 M HCl at 37°C, inside cylindrical 250-mL plastic specimen bottles. These were tightly capped after full raft development and agitated in a water bath at 37°C with a side-to-side oscillating action. Each bottle was fitted with a filter plate at the base, which could be lifted out to retain only raft pieces larger than 15 mm diameter (approximately the diameter of the oesophageal opening). The amount of raft remaining after a fixed agitation time was measured by weight.

The raft resilience of three liquid alginate anti-reflux products was tested. (Tables 1 and 2) Product A contained 10% sodium alginate and products B and C both contained 5% sodium alginate. In Table 1, a product was classified as having a raft remaining if one or more large pieces remained on the filter plate after agitation.

Table 1 Number of rafts remaining (n = 6)

Time	Product		
(min)	A	B	C
0	6	6	6
5	6	6	5
10	6	6	3
20	6	6	2
30	6	4	1
45	6	3	0
60	5	0	0

Table 2 Mean weight % raft remaining (s.d.) (n = 6)

Time	Product		
(min)	A	B	C
0	100	100	100
5	71.3 (4.4)	20.1 (1.5)	12.7 (7.3)
10	50.3 (2.8)	15.8 (1.3)	5.2 (7.2)
20	32.6 (2.9)	12.5 (2.6)	4.1 (7.2)
30	19.3 (1.5)	6.5 (5.5)	2.1 (5.0)
45	14.2 (1.6)	4.8 (5.2)	0 (0.0)
60	10.4 (5.6)	0 (0.0)	0 (0.0)

Products were statistically compared by computing the area under the percentage remaining by time curve (AUC) for each raft, and comparing the AUCs between products using one-way analysis of variance. The means (s.d.) of the AUCs were: Product A, 1842 (100); Product B, 746 (179) and Product C, 419 (212). There was an overall statistically significant difference between products ($P < 0.0001$) and each product was statistically significantly different from the other two ($P < 0.05$). This method therefore shows clear differences between products, with the product containing 10% alginate being the most resistant to break-up by agitation.

074
The preparation and properties of solvent cast films

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Solvent cast films are used as drug delivery formulations to moist surfaces, including wounds. This study aims to characterise the mechanical properties of carboxymethylcellulose (CMC) films in the presence of plasticiser (glycerol GLY) at ratios CMC:GLY from 2:1 to 1:1.5. The mechanical properties are known to affect the clinical acceptability of film dressings, regarding ease of application (Thomas 1990). Films were prepared by drying aqueous solutions of 2% w/v CMC (Blanose 7H4XF, Hercules, USA) with or without GLY, to constant weight over saturated salt solutions at relative humidities (RH) in the range 0–50%, and temperatures of 25–45°C. The water content was determined by thermogravimetric analysis, TGA (Mettler, UK), using heating rates of 5°C min^{-1} to 150°C. The tensile properties of the films were evaluated by stretching dumb-bell shaped sections to break using a Texture Analyser (TA.XTi2, Stable Microsystems, UK). The time to break (s), % strain at break, linear elastic region (mm) elastic modulus, (mPa) and work (J) in breaking the films (Aulton 1982) were compared at different stretching speeds.

It was shown that drying rates increased and water content of the films decreased at high temperature and low RH. In contrast, increasing glycerol content decreased the drying rates and increased the residual water in the films. The % of water (TGA)

correlated well with the theoretical values of water content calculated by weighing. The fastest drying times to produce reproducible films were achieved at 6% RH and 45°C, and these conditions were used to produce films for subsequent mechanical experiments.

For all films, the time to break and the % strain at break decreased with increasing stretching speed whereas elastic moduli and the work done to break the films changed only slightly. The linear elastic range depended on glycerol content and varied from a stretching length of 6 mm for the most brittle film (CMC:GLY 2:1) to 20 mm for the least brittle formulation (CMC:GLY 1:1.5). Typical data for the effect of glycerol on the tensile properties of films are shown in Table 1.

Table 1 Tensile properties of films at stretching speed of 0.5 mm s^{-1}

Film CMC:GLY	Time to Break (s)	% Strain at break	Elastic modulus (m Pa)	Work to break (J)
2:1	4.8	2.2	27.1	0.1
1:1	97.1	44.0	1.0	1.2
1:1.5	175.0	80.0	0.03	0.6

The results indicate that GLY modifies the mechanical properties of CMC films. Elastic moduli decreased with increasing glycerol content as the films became less rigid and more flexible. Work to break data indicated that a 1:1 (CMC:GLY) ratio formed the toughest films. It was concluded that the formulation prepared with a 1:1 ratio (CMC:GLY) by drying at 45°C and 6% RH shows an acceptable balance between elasticity and toughness and films of this composition will be used in future drug delivery studies.

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075

The influence of process spray rate and airflow on the adhesion of HPMC films to placebo tablet cores

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Inherent poor adhesion between a film coating and a tablet will alter the mechanical and functional properties of the final dosage form. This communication reports an investigation into two process parameters, air flow and spray rate, on the adhesion of HPMC coats to placebo tablet cores

Tablets consisting of 75.2% microcrystalline cellulose (MCC) (Avicel PH 102, FMC, USA), 24.2% lactose (Tablettose 80,Meggle, Germany), 0.4% magnesium stearate (BDH, UK) and 0.2% colloidal silicon dioxide (Aerosil 200, Degussa, Germany) (all w/w), were manufactured using a rotary tablet press (Unipress Diamond, Manesty, UK) equipped with flat bevelled-edge punches, 10 mm in diameter, to an average crushing force of 190 N and an average weight of 360 mg. Two-kilogram batches were coated using a Manesty Spray Gun in a fully perforated drum coater (XL coater, Manesty, UK) fitted with four ploughshare baffles. The drum speed was kept at a constant 20 rev min^{-1}, inlet temperature was 60°C, atomising air pressure and fan air pressure were a constant 0.5 bar, and the gun-bed distance, 150 mm. The film coating formulation consisted of 9% hydroxypropylmethylcellulose (Pharmacoat 606, Shin-Etsu Chemical Co. Japan), and 1% PEG 400 (BDH, UK). For each coating run samples were coated to an actual weight gain of 3%.

For the coating studies, a two-level full factorial design (Design-Expert Version 6, Stat-Ease, Minneapolis, USA) was used to conduct the experiment. Levels used were 200 and 300 $m^3 h^{-1}$ airflow and 12 and 22 g min^{-1} for the spray rate.

The adhesion of the films to the tablet cores were tested using a specially designed tablet tester (Force Measurement Systems, Glasgow, UK). The method of testing has been described in an earlier communication (Khan et al 2001).

The measured values of adhesion, area under the force/displacement curve (AUC) and elongation are shown in Table 1.

Table 1 Force of adhesion, AUC and elongation values

Airflow ($m^3 h^{-1}$)	Spray rate (g min^{-1})	AUC (N mm)	Adhesion (N)	Elongation (mm)
300	12	1.1 ± 0.1	13.4 ± 0.6	0.16 ± 7.6 E-017
300	22	0.7 ± 0.1	10.4 ± 0.6	0.13 ± 7.6 E-017
400	12	1.1 ± 0.1	13.8 ± 0.6	0.16 ± 7.6 E-017
400	22	0.7 ± 0.1	10.0 ± 0.6	0.13 ± 7.6 E-017

± Least significant difference (LSD) at the 95% confidence level.

Results show that airflow has no significant effect on the AUC, adhesion or elongation, whereas spray rate significantly influences all three responses.

The authors would like to thank Manesty, Kitling Road, Knowsley, Merseyside, U.K for part funding of this work.

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076

Preparation and in-vitro release study of theophylline slow-release microparticles

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Theophylline has been used for the relief of bronchospasm in asthma for more than 50 years. Due to a narrow therapeutic dosage range, rapid absorption and adverse effects which commonly affect the gastrointestinal tract and CNS, attempts have been made to design slow-release formulations to minimize the fluctuations in serum concentrations and adverse effects that follow the administration of conventional dosage forms.

In this study, different techniques, including coating beads or pellets by polymers, solid dispersion and matrix-based pelletization, were employed and the release pattern for various formulations was then evaluated.

Beads composed of theophylline and additives including soluble and insoluble diluents and binding agents, were prepared by the methods of dry and wet granulation and pelletization, using a laboratory pelletizer. The prepared beads were evaluated for appearance, particle size distribution, flowability and friability. Particles with the size range of 500–1200 μm were coated by various polymeric solutions, using a laboratory coating pan.

Matrix-based pellets were prepared with the aid of Eudragit RS. In solid dispersion, Eudragit RS, ethyl cellulose and 96° ethyl alcohol were used as retarding polymers and solvent, respectively.

Following the assay test, the release pattern of each formulation was determined based on test number 5 of theophylline monograph in US Pharmacopeia. The results of the dissolution test on matrix-based pellets showed a rapid release of theophylline in the first hour, even with high Eudragit content or in the presence of hydroxypropylmethyl cellulose as a binding agent. The same trend was observed for slow-release granules, prepared by solid dispersion technique, when the amount of Eudragit RS 100 and ethyl cellulose increased up to 10 and 12 times that of the active ingredients.

It was observed that the beads composed of drug, dicalcium phosphate and polyvinyl pyrrolidone, which were prepared by wet granulation, did not agglomerate and adhere to the internal wall of the coating pan. This formulation was then selected as the core for the coating stage to prepare beads with various coating thickness. Coating was performed in laboratory pans with the aid of Eudragit RS 100 alcoholic solution. The release pattern of the drug from the coated beads prepared with one, two and three times coating (f_1, f_2, and f_3, respectively) did not comply with the pharmacopeial criteria. However, desired dissolution behaviour was obtained when specific proportions of these formulations were combined. The correlation coefficients of the release–time curves obtained for the combined formulations showed that release rate followed first-order kinetics over 10 h.

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Characterisation of the bioadhesive properties of a polymer-coated powder

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There are no standard in-vitro methods for measuring bioadhesion. Some methods use mucin as a substrate to which a bioadhesive formulation is added and mucus adsorption measured by turbidimetric and colorimetric methods. Zeta potential measurements have also been used to estimate adhesion between a positively charged delivery system and negatively charged mucin (Ping et al 1998). Other methods for predicting bioadhesion involve measuring the force of detachment from a tissue substrate. This work investigates whether such methods, generally used for solid dosage forms or microspheres, can be adapted to predict bioadhesion of a polymer-coated drug powder and also the suitability of non-tissue substrates in detachment studies.

Fluticasone propionate (FP) was coated with different amounts of hyaluronic acid (HA). Mucin adsorption of the polymer-coated drug was quantified using turbidimetric and colorimetric methods (Ping et al 1998). The force of detachment of a known quantity of powder, HA/FP or FP, scattered onto a disc of agar ($12\,g\,L^{-1}$) or agar/mucin was measured (perspex disc probe, ~1.5" diameter, applied force 50 N held for 120 s) using a Texture Expert TA-XT2 (Stable Microsystems, UK). Zeta potential measurements (Zetasizer 3000, Malvern UK) of suspensions of FP and HA/FP, ($2\,mg\,mL^{-1}$) in pH 7 acetate buffer were also made initially and 30 min after preparation.

Table 1 Summary of data after 30 min adhesion

% HA	Force of detachment after 30 min (g)		Free mucin (% w/v)[a]	Turbidity[b]
	Agar substrate	Agar/mucin substrate		
0.04	39.2 ± 38.1	17.4 ± 8.1	0.084	0.162
0.07	95.1 ± 50.8	156.6 ± 67.6	0.076	0.126
0.11	43.8 ± 34.7	26.9 ± 10.6	0.030	0.131
0.2	125.3 ± 20.9	153.8 ± 57.6	0.0093	0.104
0.0	1.1 ± 3.3	14.8 ± 4.6	0.092	0.186

[a]0.1% mucin: 0.6% sample. Colourimetric assay, ± s.d. ˜10^{-4}. [b]90% mucin: 10% test suspension ($2\,mg\,mL^{-1}$)

The results in Table 1 indicate that the same general trends are shown by both mucus adsorption and detachment methods after 30 min, even though the latter showed large standard deviations. As the quantity of HA increases, adhesion also increases indicated by the decreasing free mucin concentrations (turbidimetric and colourimetric data) and increasing force of detachment values. Both techniques indicate that there is a massive increase in adhesion with the 0.07% HA sample. The zeta potential values become more negative as the amount of HA increased from 5.2 ± 5.4 mV at 0.04% HA to −45.3 ± 3.6 mV at 0.2% HA implying that electrostatic attraction is not the initial driving force for bioadhesion,

The data, when used in conjunction with visual observations, suggest that hydration forces and polymer entanglement account for the muco-adhesive properties of the formulation. They also imply that a conformational change occurs in the HA solution at ~0.07% w/v allowing greater adsorption of HA to the FP particles.

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078

The influence of macrostructural properties on the dielectric response of hydrated proteins

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Our previous studies have discussed in some length the dielectric properties of various hydrated globular proteins (Suherman & Smith 2002) and the application of dielectric measurement to the in-situ measurement of residual moisture during lyophilisation (Suherman et al 2002). Our work in this paper aims to establish the significance of the macrostructural properties of proteins to the dielectric response. Various hydrated globular proteins (ovalbumin, lysozyme and pepsin) with a range of water contents (0.05–0.2 g water (g dry protein)$^{-1}$) and various microstructural properties have been investigated. For dielectric measurement, each sample was placed between two circular brass electrodes and subjected to a stepwise increase in temperatures from 253 to 363 K. Low frequency dielectric measurements (0.1 Hz to 1 MHz) were carried out using a Solartron 1296 dielectric interface connected to a Solartron 1255 frequency response analyser and Oxford temperature controller ITC^{503}. The properties of the samples were also investigated by other complementary methods (i.e. DSC, FTIR, SEM and XRD).

As shown in previous results, the dielectric response from hydrated proteins was characterised by a low-frequency dispersion (LFD) and an ε_3-dispersion (observed at high frequency). The ε_3-dispersion in the spray-dried ovalbumin was observed more clearly than that in the freeze-dried ovalbumin. The difference between these ovalbumins relates mainly to the macrostructural properties of the two materials. SEM results show that spray-dried ovabumin has a regular globular shape, whereas the freeze-dried ovalbumin has a thin flake shape. The elusive ε_3-dispersion was also found for hydrated pepsin, which has more crystalline state compared with ovalbumin, and has an irregular shape for its macrostructure.

Our previous work assumed that the ε_3-dispersion was due to intra-cluster proton transport, that was limited to the molecular geometries of each protein. In this study it can be confirmed that the macrostructural properties of the powder (i.e., particle size and shape) have little effect on the ε_3-dispersion and one may infer that the dispersion originates from, and is confined to, the microscopic rather than macroscopic properties of each material. Instead, it was possible to show that the LFD response was sensitive to the polarization of larger scale domains that are defined by the macroscopic geometry of materials. The observations clarify our previous work on in-situ dielectric measurements, in which the ε_3-dispersion from freeze-dried sample in the glass vial was not observed as clearly as that for the spray-dried sample. The reason can be associated with the influence of the LFD of the powder and the glass vial in obscuring the ε_3-dispersion.

The impact of the macrostructural properties of freeze-dried materials on their dielectric response may be exploited to provide some in-situ or end-testing of the product matrix derived from the freeze-drying process.

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079

Forces that develop during tapping of powders

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Packing and densification of powders due to compression or vibration is important to many pharmaceutical processes and have been traditionally described by following the volumetric changes during tapping in a glass cylinder. The disadvantages of this method are mainly associated with the lack of reading precision due to the uneven powder surface, and to the fact that measurement is restricted to static conditions due to the downward movement of the powder by its inertia between two cylinder drops, not simulating the processing conditions. Little attention has been given to the measurement of forces in the powder column during tapping and, in particular, to the compressive stress applied (Fukuoka & Kimura 1992).

A load cell was fitted to a tester that complies with the BS 1460 for the dimensions of the cylinder (250 mL), the drop height (25 mm), and the falling rate (30 taps per minute) and a system was developed for recording of the force exerted on the base of the cylinder during tapping. Rapid data acquisition at speeds of 5000 samples per second (one measurement every 0.2 ms) allowed dynamic recording of the maximum compressive force exerted at every cylinder drop. Four size fractions of

maize starch (Cerestar, Italy) of mean diameters 11, 45, 75 and 150 μm, obtained by using sieves and the zig-zag classifier, were tested.

Table 1 Compressive pressure and number of taps at kink point and at plateau conditions, determined from plots of compressive pressure against number of taps, for the four size fractions of starch powder

Mean starch size (μm)	Pressure ($kN\,m^{-2}$)		Number of taps	
	At kink	At plateau	At kink	At plateau
11	7.4 (0.4)[a]	10.2 (0.5	44 (5)	81 (11)
45	7.1 (0.2)	8.9 (0.3)	12 (4)	65 (3)
75	7.0 (0.2)	9.3 (0.3)	4 (1)	48 (2)
150	—	9.3 (0.1)	—	37 (3)

[a]s.d. is given in parentheses (n = 3)

The plot of the pressure transferred to the base of the tapping cylinder against the number of taps applied, showed differences for the four experimental size fractions. For the two cohesive powders (11 and 45 μm) and, to a lesser extent, for the intermediate size fraction (75 μm), there is initially a slow pressure increase with tap number, reaching a kink point after a certain number of taps, above which the pressure increases rapidly, reaching plateau at higher tap numbers. From Table 1 it can be seen that the number of taps and, to a lesser extent, the pressure corresponding to the kink point increase as the mean particle size decreases. This is because of structural irregularities due to high frictional forces existing in the mass of cohesive powders, which require more energy to break than in the case of free-flowing powders. Once this number of taps has been exceeded, the densification of cohesive powders progresses faster, leading to a coherent plug-like structure. This transmits the stress more efficiently to the base of the tapping cylinder, as indicated by the higher pressure value of 10.2 ($kN\,m^{-2}$) at plateau conditions (near the end of tapping test), seen in Table 1 for the lower size fraction of starch powder.

The above findings indicate that measurement of forces exerted by the powder on the cylinder base during the tapping test, provide a new approach for the characterization of powder packing and densification, which could be useful for the selection of processing conditions during pharmaceutical formulation.

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080
Small-scale high-throughput screening for polymorphs using D8 GADDS to determine x-ray powder diffraction patterns

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Polymorph screening to find both stable and metastable forms by varying solvent and crystallisation conditions is an important step in the development of an API since inappropriate nomination of a solid form/polymorph can have important financial and regulatory consequences. Traditional methods of screening involve variation of crystallisation conditions (e.g., solvent, temperature and super-saturation) and usually involve significant quantities of material (1–5 g). There is an increasing need for this activity to be carried out earlier in discovery when limited compound is available and this has its own associated problems of scale. To screen for polymorphs earlier, small-scale crystallisation, solid-state characterisation and data analysis need to be automated and integrated to handle a large number of compounds.

The first stage of this approach is to crystallise samples onto multiwell plates for subsequent high-throughput analysis using D8 GADDS powder diffraction. This will be demonstrated using examples of drugs exhibiting polymorphism. These include primidone, indometacin and chloroquinaldol which were used as supplied (Sigma-Aldrich). The drugs were slurried separately for 24 h in a variety of solvents at room temperature and filtered to give clear solutions free from solid material. Each drug solution was titrated (200 μL) into a split multiwell plate and 3 replicates carried out. Solutions were allowed to evaporate at variable temperature to give crystals on a split-bottom plate and this was presented to the X, Y, Z stage of the D8 GADDS X-Ray Diffractometer. The PXRD patterns of all the crystal sample spots were measured using the automated stage and focused and aligned using the integrated laser system. A 300-μm collimator was used to illuminate the sample with x-rays and the data collected for 300 s using the 2D detector. Each of the frames of 2D diffraction data consist of large 2-theta and chi ranges which contain diffraction spots or Debye rings. These were integrated to give conventional 1D diffraction data. All of the diffraction data from the crystallisation plates were analysed using SNAP1D (Glasgow University Chemistry Dept). This process involved background subtraction, extraction of 2 theta peak positions and then pattern matching.

For primidone and indometacin, the two known anhydrous polymorphs were identified by cross correlating the results from SNAP1D and comparison of the XRD patterns by manual visualisation confirmed these findings. For chloroquinaldol only one pure form was identified, the remaining samples being a mixture of two forms. This was confirmed by examination of the PXRD patterns.

The method of small-scale high-throughput polymorph screening using D8 GADDS X-Ray diffractometry to characterise material prepared by crystallisation on multiwell plates and analysis of data, was shown to be a valuable approach. At present, a method of automating the process of preparation of multiwell plates for an integrated system is progressing.

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Diffusion of acid through calcium alginate gels

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Alginates are a main ingredient in many products used to treat the symptoms of gastric reflux, working by interacting with calcium to form a physical floating barrier that may be refluxed in place of the gastric contents. If these alginates also adhered to mucous membranes they may exert a further barrier action against potentially harmful agents such as pepsin and acid. This Franz cell study investigates the ability of calcium alginate gels formed from four different sodium alginates (Table 1) to inhibit the diffusion of acid.

Table 1 Physical properties of the alginates used in the study

Alginate	MW ($g\,mol^{-1}$)	F_G (fraction guluronate)	Concn used (% w/v)
A	34 700	0.64	5
B	75 000	0.45	5
C	387 000	0.69	1
D	397 000	0.46	1

Sodium alginate, calcium carbonate and D-gluconic acid lactone solution (0.5 mL) was pipetted onto a dialysis tubing membrane in a Franz diffusion cell, and left for 12 h at 4°C to gel. This gave a layer 113 mm^2 in area and 3 mm depth. The receptor phase, 12 mL freshly boiled and cooled water at pH 7, was constantly stirred and the system kept at 37°C throughout the study. HCl (0.1 M, 200 μL) was layered on the top of the gel as the donor phase and the pH of the receptor phase was recorded using a microelectrode at set time intervals.

Table 2 pH of the receptor phase with time

Alginate	pH		
	30 min	60 min	120 min
A	5.41 ± 0.18	5.14 ± 0.15	4.71 ± 0.10
B	5.38 ± 0.26	5.14 ± 0.23	4.79 ± 0.12
C	4.66 ± 0.24	4.48 ± 0.20	4.18 ± 0.07
D	5.32 ± 0.24	4.31 ± 0.28	4.03 ± 0.07

Data are expressed as means ± s.d., n = 5

The pH decline was initially rapid, then slowed to give values of 4–5.5 between 30 and 120 min (Table 2). These pH values are significantly higher ($P < 0.0001$, analysis of variance) than when 200 μL 0.1 M HCl was placed directly onto the dialysis tubing (pH 2.95 within 5 min). The diffusion through alginates C and D fell to significantly lower pH values than alginates A and B at 60 min and 120 min ($P < 0.05$ analysis of variance). The high guluronate-containing alginate (C) was seen to shrink in the Franz cell, allowing the acid to pass around rather than through the gel. This could be an important factor limiting any possible barrier action. It is probable that the high number of carboxyl groups present in A and B (relative to C and D) may also act as a buffer.

It was concluded that an adhering alginate layer could act as a barrier to acid. Alginates with high mannuronate contents may be preferred, as they are less likely to shrink in the presence of acid.

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Adhesion of formulation coatings to the oesophagus: a novel in-vitro test system

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The adhesion of solid formulations to the oesophagus during swallowing has been widely implicated in medication-induced injury to this organ (Jaspersen 2000) and may also significantly delay the onset of drug action. Formulation size, shape and surface characteristics are factors that affect dosage-form adhesion in the oesophagus (Marvola et al 1982; Channer & Virjee 1985). In this study, a novel in-vitro apparatus to investigate the oesophageal adhesion was developed, based upon a previously described test system (Young & Smart 1998).

Porcine oesophageal mucosa, dissected free of encapsulating musculature, was rinsed with saline, flash frozen and stored at –20°C until required. This was mounted to yield a flat test surface, ca. 120 mm × 15 mm and inclined at 10° from horizontal. The tissue was maintained at 37°C and ca. 100% relative humidity and an artificial saliva (a mucin saline solution) flowed across the epithelial surface at a rate of 1 mL min^{-1}. Polymer-coated glass discs (8 mm diameter, dried film weight 2 ± 0.3 mg) with a 2.0-g consolidation weight were then drawn upwards across the mucosal surface at a rate of 0.22 mm s^{-1} and the forces required to achieve this recorded. The maximum detachment force (MDF, the largest shear force recorded in mN) and the total work of adhesion (TWA, area under the force–distance curve in μJ) was then calculated and expressed relative to an uncoated glass disc tested on the same tissue.

It was found that all polymer coatings were significantly ($P < 0.05$, Student's *t*-test) more adhesive than the non-adhesive control paraffin wax (Table 1). Alginic acid and hydroxypropylmethylcellulose (HPMC) demonstrated the highest TWA. For HPMC, a high initial force was obtained which was considered to reflect initial adhesion due to polymer hydration yielding a hydrogel which was subsequently sloughed from the substrate surface during transit across the epithelium. Conversely, alginic acid was seen to re-adhere to the mucosal surface. The pluronic surfactant F127 also adhered, while polyethylene glycol (MW 6000) and gelatin exhibited comparatively low adhesion.

Table 1 Adherence of test materials to isolated porcine oesophageal tissue (n = 6)

Test Material	Relative TWA	s.d.	Relative MDF	s.d.
Alginica acid	6.35	2.43	19.57	9.09
HPMC	2.32	0.73	8.53	3.42
F127	2.12	0.57	7.08	2.86
Polyethylene glycol	1.14	0.43	0.99	0.13
Gelatin	1.01	0.15	1.62	0.69
Paraffin wax	0.76	0.09	0.73	0.11

It was concluded that this novel in-vitro test system can be used to investigate polymeric coatings for their adhesive properties.

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083
Temperature modulated differential scanning calorimetry of a spray-dried model protein: catalase

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The spray-drying of thermal sensitive proteins can lead to different amounts of denaturation and degradation, depending on the spray-drying conditions. Spray-drying temperature, feed rate and atomisation pressure are some of the spray-drying parameters that can be optimised to minimise denaturation and degradation of labile molecules (Masters 1991). In addition, stabilisers such as sugars and polyols are often used to preserve the native conformational structure of proteins (Reinhard et al 1997). An added advantage of using sugars is that generally, the spray-drying of such molecules leads to the formation of amorphous material. The latter can be characterised by its glass transition (Tg). In this study, temperature modulated differential scanning calorimetry (TMDSC) was used to determine the stability of spray-dried catalase formulations and to provide some insight into the mechanisms involved in the thermal denaturation of a model protein. The effect of a variety of stabilisers on the preservation of the native structure of spray-dried catalase was investigated thermally using two spray-drying inlet temperatures (95°C and 180°C).

The thermal analyses of catalase formulations showed two endothermic transitions, the glass transition of the sugar excipient and the denaturation of catalase. The glass transition of each catalase/sugar formulation did not show any trend with regard to the catalase:sugar ratio. However, the trehalose/catalase formulations were found to have higher glass transition temperatures than the sucrose/catalase formulations when prepared at either of the inlet temperatures. The difference in glass transition temperatures was more apparent employing a spray-drying inlet temperature of 95°C than at the higher temperature of 180°C. The glass transitions for both the sucrose and trehalose-based formulations increased as the inlet temperature of the spray-drying process was increased.

The denaturation endotherm of catalase without excipient had the lowest enthalpy whilst sucrose and trehalose formulations of catalase showed the highest denaturation enthalpy. However, the denaturation temperature for these disaccharide formulations was lower than that for catalase spray-dried without any excipients. Generally, the denaturation enthalpy decreased as the denaturation temperature increased. The thermal profile of catalase spray-dried with the disaccharides displayed a denaturation peak with a leading or tailing shoulder depending on the inlet temperature, type of disaccharide and ratio of catalase to stabilising excipient. Such results suggested the existence of two denaturation processes that are closely related thermodynamically. In this study it has been illustrated that TMDSC has the capacity to measure thermal changes related to the conformational structure of catalase. Furthermore, the preservation of the native structure of catalase is dependent on the spray-drying temperature and the type and amount of stabiliser used during the spray-drying process. It has been demonstrated that TMDSC is able to differentiate between the possible stabilisation mechanisms of sucrose and trehalose for the labile catalase molecule.

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084

The development of a microwave-moisture sensor for fluid-bed drying

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Fluid-bed drying is used widely in the preparation of pharmaceutical granules for tablet manufacture (Hlinak & Saleki-Gerhardt 2000). The variable density of a fluidized-bed necessitates the application of density-independent methods for measuring moisture. In a number of cases, a solution has been found through the use of microwave sensors combined with radioactivity densitometry. However, the problem of safety restricts the widespread use of the radioactive source in a number of industries.

In this paper we describe the design and operation of a new microwave-only moisture sensor that overcomes the inherent problems associated with measurements on fluid-bed drying installations, viz. the low density and low moisture content of the materials under test; the elevated, and rather high, temperature of the fluidized bed; and the loading/unloading of the measurement cuvette. The device is based on a two parameter resonator measurement with an extended central conductor and a sample cuvette which is unloaded by pneumatic discharge. Tests involving fluid-bed drying of range of materials (including milk powder, casein, wheat flour, and talc) were carried out within a simulated industrial framework.

A laboratory fluid-bed dryer was constructed from a 60 L stainless vessel with a bottom mesh, through which the hot air was fed. The pressure was adjusted to obtain the fluidised bed exceeding the height of sensor installation. The air temperature in the fluid bed was maintained at 85 $\pm$ 5°C. Material was removed through a sample hatch, at timed intervals during the process of fluid bed drying. The moisture of these samples was determined by drying to constant weight (at 103°C). Simultaneous measurements of the resonance frequency and signal amplitude of the sensor were taken at each time point, and then converted to a meter reading of the moisture content (via a pre-determined calibration algorithm). The accuracy of the measurement was then determined as 2 standard deviations of the difference between the meter moisture reading and that from the loss-on-drying method (Table 1).

Table 1 Sensor performance characteristics

Material	Moisture (5)*	2 s.d. (%)	R
Spray-dried milk	2–6.5	0.12	0.996
Casein	3.5–8	0.16	0.994
Flour	11–14.5	0.13	0.993
Talc	0.05–0.3	0.01	0.985

*Moisture determined by loss-on-drying

The new sensor was shown to provide adequate metrology for the control of fluid-bed drying, with a measurement accuracy of ~0.14% (typically). The results obtained for talc suggest that it is possible to measure moisture contents as low as 0.05%, with an accuracy quite sufficient for process control of pharmaceutical processes. Moreover, precise correction for fluctuations in material density was demonstrated which enabled the authors to suppose that the density variations in the real industrial processes will not affect the accuracy of moisture determination.

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085

A study of the partitioning behaviour of phenoxyethanol in aqueous cream BP and hydrous ointment BP

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Aqueous cream BP and hydrous ointment (oily cream) BP are both semi-solid emulsions used for the topical delivery of drugs. Such systems primarily contain oil and aqueous phases and, it has been proposed, a number of other structured phases (Junginger 1984). They contain a preservative, phenoxyethanol (PE, 1% w/v), to protect them from microbial challenge. The preservative, added to the aqueous phase (in which it is active) before emulsification, may partition into other, lipophilic, phases during cream manufacture. As a result, the aqueous PE concentration may be lower than expected with the consequence that the formulations may not adequately withstand unintentional microbial contamination. In this work the partitioning behaviour of PE in both formulations was investigated. Since partitioning of the preservative only occurs during manufacture (the only time the oil phase is liquid), experiments were conducted to determine the effects on partitioning of emulsification time. Samples were prepared both with and without emulsifier.

Batches of creams (600 g) were manufactured in accordance with the relevant BP monograph either with or without emulsifier. Thornton & Ross Ltd. supplied BP-grade materials and samples of industrially manufactured creams for comparison. Batches were homogenised for various periods of time (1, 2 and 3 h) at 60°C. Samples without emulsifier readily separated into aqueous and oil fractions. Samples with emulsifier were centrifuged (6700 *g*, 1 h) to effect a separation. In all cases analysing the aqueous layer at 269 nm and comparing the data with a Beer–Lambert plot constructed for PE in water determined the PE concentration in the aqueous phase, Tables 1 and 2.

Table 1 Aqueous cream BP. — PE concentration (M)

Time (h)	0	1	2	3
With emulsifier	0.105	0.104	0.099	0.095
s.d.	n/a	0.001	0.004	0.003
Without emulsifier	0.105	0.107	0.104	0.104
s.d.	n/a	0.003	0.003	0.003

Table 2 Hydrous ointment BP. — PE concentration (M)

Time (h)	0	1	2	3
With emulsifier	0.149	0.110	0.082	0.086
s.d.	n/a	0.026	0.027	0.032
Without emulsfier	0.149	0.117	0.118	0.119
s.d.	n/a	0.003	0.004	0.003

In the case of aqueous cream BP it can be seen that the PE concentration in the aqueous phase remains relatively constant irrespective of either mixing time or the presence of surfactant. The aqueous PE concentration in the sample of industrial cream was found to be 0.096 M (s.d. 0.002). However, in the case of hydrous ointment some partitioning seems to occur. With no surfactant the aqueous PE concentration drops to 0.119 M within 1 h, subsequently remaining at this value. With surfactant the aqueous PE concentration drops to 0.110 M in 1 h and to 0.082 M after 2 h. This implies that PE partitions into the lipophilic phases of this formulation and that this effect is greater in the presence of emulsifier. The aqueous PE concentration in the commercial sample was 0.099 M (s.d. 0.025). It is notable that the two formulations had the same aqueous PE concentration despite having different compositions. It is concluded that care must be taken when formulating preservatives into complex systems such as creams, as the final concentration of active preservative may be significantly lower than expected.

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086

An assessment of novel lubricants using an instrumented Minipress

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Lubrication is a key issue for successful tablet development, with Magnesiun Stearate (MS) being the most prevalent of all solid dose excipients. Despite its

popularity, overlubrication results in protracted dissolution times from the resultant weak tablets. Compritol 888 ATO (COM) and Precirol ATO 5 (PRE) are novel materials with lubricant, glidant and putative binding action which, by virtue of their sphericity, do not over-mix upon prolonged agitation (Gattefossé 2000). Lanthanum carbonate (LAC) is a new safe orally active treatment for renal failure associated hyperphosphataemia. High unit doses necessitate blend lubrication, glidant addition and facilitated die filling to ensure efficient tablet production at scale. Using an instrumented table-top rotary press to conserve drug, an evaluation has been performed of blending time on tablet performance using MS and newer lubricants to evaluate their additional functionality.

Screened LAC–Emdex–Aerosil 200 (46:51:2) (2 kg) were blended at 20 rev min^{-1} for 20 min, whereupon lubricant (1%) was added and blended for 5 (−) or 20 (+) min as listed in Table 1. Compression and ejection forces were continuously measured during tableting (10 mm) on a Picola Table-top mini rotary tablet press (Picola, Argentina). Powder blend was continually fed by twin paddle. Mean ejection force (n = 10) and hardness were plotted versus compression force from which data at specific forces were interpolated. Disintegration was assessed on BP apparatus.

Table 1 Tablet characterisation (n ⩾ 6)

		Attribute Ejection/kg			Hardness/kp			
Compression force/kg		250	500	750	250	500	750	Disin/min
Lub	Mixing							
MS	−	4.0	8.0	12	1.4	2.3	3.2	7.2
	+	2.5	4.9	7.0	1.0	1.8	2.8	12.2
COM	−	32	42	41	1.5	2.5	3.7	2.75
	+	22	28	36	1.3	2.3	3.8	3.0
PRE	−	27	35	43	1.4	2.7	4.1	3.75
	+	13	17	24	1.3	1.8	2.6	2.5

Lub – Lubricant; Disin – Disintegration (tablets of 1.8–2.2 kp)

From ejection forces, die-wall stress increased with compression force as did tablet strength, irrespective of additive or blending time. Appreciable ejection forces were recorded for PRE and COM at the lower end of their recommended range (1–5%), where MS was shown to be effective. Tablet strength was significantly reduced for overlubricated MS and PRE ($P < 0.05$) but unaffected with COM ($P > 0.05$). COM and PRE have been shown to resist over-mixing and assist binding of plastic materials, which are notoriously sensitive to overlubrication (Gattefosse 2000). From compaction simulation, LAC binds by predominantly brittle fracture. De Boer et al (1978) found dramatic loss of tensile strength for MS overlubricated plastic materials, an effect that is reduced with brittle materials (e.g. LAC) but clearly not eliminated. In contrast to MS, COM — being a spherical lubricant — does not undergo delamination with an increased surface coverage and hence tablet bonding was unaffected. Disintegration was most affected by overlubrication of MS, whereas COM and PRE samples did not prolong tablet wetting and break-up.

Although less efficient than MS, COM is a robust alternative for tablet lubrication that is insensitive to over-mixing and does not interfere with tablet strength and disintegration time, whereas PRE offers little advantage based on these data.

Compritol ATO 888 and Precirol ATO 5, Gattefossé Brochure, 2000

De Boer, A. H., Bolhuis, G. K., Smedema, C. F. (1978) *Powder Technol.* **52**: 33–43

087
An assessment of novel lubricants using the FT3 Powder Rheometer

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Lanthanum carbonate (LAC) is a new safe and effective treatment for hyperphosphataemia. Presented as a chewable tablet, the cohesivity of LAC inhibits flow under gravity, paddle-feed being indicated to ensure reproducible die fill and consistent tablet mass. Overblending magnesium stearate (MS) is not uncommon during force-feed die filling (Bossert & Stamm 1980). Compritol 888 ATO (COM) and Precirol ATO 5 (PRE) are novel lubricants with glidant functionality and putative binding properties that purportedly resist overlubrication (Gattefossé 2000). The FT3 Powder Rheometer (Freeman Technology, UK) measures torque as a pitched blade is traversed helically through a column of test powder from which total energy is derived as a Basic Flowability Energy (BFE). Variation of test program permits discrimination between blends, optimisation of excipients, their levels, and their interplay with process, while committing minimal quantities of limited active during early development. As an alternative to traditional empirical means, the work described examines the performance of two novel lubricants using this new equipment.

Screened LAC–Emdex–Aerosil 200 (46:51:2) (2 kg) were blended at 20 rev min^{-1} for 20 min, whereupon lubricant (1%) was added and blended for 5 (−) or 20 (+) min as listed in Table 1.

Table 1 Blend attributes (n ⩾ 3)

	Blend					
	1	2	3	4	5	6
Lubricant	MS		COM		PRE	
Mixing	−	+	−	+	−	+
AoR [(o)]	57.3	55.2	60.3	55.4	57.3	55.6
CCI (%)	18.3	14.0	14.3	12.5	12.5	12
BFE (mJ)	145	125	134	133	141	135
FRI	2.34	2.06	2.11	2.12	2.27	2.27
SI	1.10	1.07	0.99	1.00	1.09	1.00

Traditional flowability indices, angle of repose (AoR) and Carr's index (CCI), did not differentiate between lubricants, although extensive blending resulted in values indicating improved flow, particularly the CCI for MS. After 5 min mixing, BFE for MS had the greatest energy requirement consistent with the low glidant efficiency of this material, whereas overlubrication of this blend after 20 min resulted in the greatest drop in BFE. Shearing of the plates and high surface coverage account for overlubrication with MS. In contrast, the sphericity of COM resisted overlubrication and the BFE remained constant, whereas PRE had intermediate behaviour. Prospectively, optimum blending for these additives could be identified by the time for the BFE to plateau.

Flow rate index (FRI) is a measure of response to flow rate and is calculated by division of the BFE at a tip speed of 10 by that at 100 mm s^{-1}. Greatest and lowest FRI was noted for MS at 2 and 20 min mixing, respectively, suggesting blend instability. In contrast, COM blends had a lower BFE and hence should result in improved die filling, and a lower but constant FRI indicating improved flowability under low external force (e.g., flow in a hopper). Division of the BFE for the initial conditioned sample by the corresponding value following aggressive mixing by FT3 gives the Stability Index (SI). Values approximating unity for COM (and PRE at 20 min) blends indicated stable flow properties unaffected by additional processing such as that experienced during force-feeding.

The FT3 Rheometer provides profile information on powder flowability over and above standard powder testing, in this instance on lubricant performance. COM is a useful alternative to MS that effectively resists overlubrication and possesses additional glidant activity, whereas the role of PRE requires further investigation.

Bossert, J., Stamm, A. (1980) *Drug Dev. Ind. Pharm.* **6:** 573–589

Compritol ATO 888 and Precirol ATO 5 (2000) Gattefossé Brochure

088
An in-vitro investigation of a topical prophylaxis for schistosomiasis

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Schistosomiasis is endemic in seventy-four countries and infects 200 million people, with a further 500–600 million at risk. Infection occurs when larvae (cercariae) penetrate the skin in infected freshwater. Previous work by the authors

(Bartlett et al 2000) has demonstrated that an in-vitro model based on the Franz cell using excised human skin can be utilised to study the temporal dynamics of cercarial penetration. In the past, the lack of such a model has meant that the topical application of barrier formulations to prevent infection has never been systematically studied. However, early field trial studies (Pellegrino 1967) showed that motor oil and Vaseline might give some protection. Thus, the aim of this study was to test the potential of novel barrier preparations to inhibit cercarial penetration using a novel in-vitro model of infection.

Human skin from patients undergoing elective abdominoplasty was assembled in Franz cells, as described by Bartlett et al (2000). Cells were treated with either a formulation developed within King's College London (KCL), silicone oil, liquid paraffin or Vaseline. Control untreated skin and inert-membrane cells were also included. The test formulations were left in contact with the skin for 1 h before cercarial exposure. A known number of cercariae were applied to the skin for 20 min and then removed. The recovered cercariae were counted and it was assumed that any cercariae not recovered had become attached to or penetrated the skin; this was confirmed by microscopy. The procedure was repeated 24 and 48 h after application of the formulations.

It was possible to recover 100% of the applied cercariae from the control inert membranes. Roughly 20% of the cercariae were recovered from the cells containing untreated skin confirming that the cercariae were readily attracted and attached.

Vaseline, liquid paraffin and silicone oil all offered some degree of protection, but the most striking result was achieved with the KCL formulation where all the cercariae were recovered. These results were not statistically different from the inert membrane ($P = 0.9942$).

The anti-penetration activity of the formulation was shown to persist for at least 48 h and if the formulation was left in contact with the skin for 3 h then the skin could be washed without any loss of protection.

The formulation was shown to be effective against both a laboratory maintained strain of *S. mansoni* and one recently isolated from an endemic area, as well as *S. haematobium*, *S. intercalatum* and *S. bovis*.

This study suggests that a simple barrier formulation could dramatically reduce schistosome infection levels. The formulation is inexpensive, all components are included in the GRAS list of the US Food & Drug Administration and have been widely used as pharmaceutical excipients for many years. The lack of need to include a novel bioactive compound means that the treatment should not be associated with unwanted side effects and thus could quickly enter field trials.

Bartlett, A., Brown, M., Marriott, C., et al (2000) *Parasitology* **121**: 49–54
Pellegrino, J. (1967) *Exp. Parasitol.* **21**: 112–131

089
The effect of hydro-alcoholic co-solvents on the physical behaviour of hydroxyethylcellulose

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Hydroxyethylcellulose (HEC) has been employed as a component of semisolid bioadhesive topical drug delivery systems (Jones et al 1997). The performance (e.g. transdermal drug permeation) of these systems may be influenced by the incorporation of various water-miscible solvents (e.g. ethanol and propylene glycol), but their influence on the physical properties of the dosage form have not been previously described. In the presence of organic co-solvents, it has been reported that structural changes in water occur (Cowie et al 1961), and accordingly, this may influence the rheological performance of gel formulations. This study highlights the effect of model co-solvents, ethanol, propylene glycol and glycerol on the mechanical and rheological properties of HEC gel systems.

Gels were manufactured by dissolving HEC (1–5%w/w) in the appropriate co-solvent with the aid of mechanical stirring. Glycerol, ethanol and propylene glycol (0–100% w/w)/water co-solvents were formulated. The oscillatory and continuous shear properties of the formulations were assessed using a Carri-Med CSL^2-100 rheometer whereas texture profile analysis (TPA) of all formulations was performed as previously described (Jones et al 1997). Results are shown in Table 1. All measurements were performed, at least, in quadruplicate. The effects of solvent type and concentration on the mechanical properties were statistically examined using a two-way analysis of variance ($P < 0.05$ denoted significance).

Table 1 Mechanical parameters of representative gels composed of 5% HEC in various glycerol/water solvents

Gly/H_2O ratio	Hardness (N)	G' (1Hz) (Pa s)
10/90	1.61 ± 0.07	1388.33 ± 86.23
30/70	2.23 ± 0.05	2038.86 ± 145.89
60/40	2.76 ± 0.06	3097.41 ± 89.02
80/20	0.62 ± 0.01	744.47 ± 21.63

Each of the polymer systems used in this study displayed physical properties that were dependent on the concentration of organic co-solvent present. Increases in the concentration of ethanol, propylene glycol and glycerol significantly increased the rheological and textural properties of all gels, reaching a peak or plateau at a concentration that was dependent upon the co-solvent. The formation of an enhanced structure may be due to the ability of the alcoholic solvents to reduce the dielectric constant of the overall co-solvent mixture, reducing the energy barrier to the formation of secondary interactions. This may increase the mechanical parameters within specific limits for each different alcohol. It is believed that as alcohol is added HEC begins to form hydrogen bonds with the alcohol which, in turn, may be bonded to itself and to water, reducing the mobility of the solvent and allowing expansion of the HEC polymer chain. As the percentage of alcohol increases the number of bonds between polymer and solvent reduces and the number of solvent-solvent bonds increases (Kim et al 1994), thereby, reducing the physical properties of the gel. The concentration at which this exchange from a highly structured gel to a weak gel structure is influenced by the nature of the alcohol and the number of hydroxyl groups.

While the mechanisms of gel formation and rheological properties of cellulose ethers in aqueous solvents is well understood, little is known about their rheological properties in mixed solvent systems. The results of this study have shown that the presence of water-miscible organic solvents (ethanol, propylene glycol and glycerol) can have significant effects on the rheological and textural properties of cellulose ether gel systems.

Cowie, J. M. G., Toporowski, P. M. (1961). *Can. J. Chem.* **39**: 2240
Kim, S.S., et al (1994) *Polymer* **35**: 3212–3216
Jones, D. S., et al (1997) *Pharm. Res.* **14**: 450–457

090
The use of texture analysis to assess the physical properties of sputum collected from adult cystic fibrosis patients

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Cystic fibrosis (CF) is the largest autosomally recessive genetic disorder in children worldwide. It is caused by a mutated transmembrane conductance regulator, which acts as a chloride channel. This defect leads to abnormal epithelial Na^+ and Cl^- transport, resulting in the production of thick, viscous mucus in the lungs (Boat et al 1989). The physical properties of CF sputum are paramount in determining its adherence to lung walls and its ease of clearance by cough or ciliary action. We have investigated using the TA.XT2 Texture Analyser (SMS, Godalming) (TA) for the measurement of sputum adhesivity, work of adhesion and cohesiveness. In TA, a probe is inserted into and withdrawn from the sample under specified conditions. The adhesiveness is measured by the force required to detach the probe from the test material and the cohesiveness corresponds to the distance travelled before separation of the probe and test substance occurs. The TA.XT2 allows sputum to be studied as received, without pre-treatment. Sputum samples were collected from

patients at Belfast City Hospital and immediately refrigerated at 4°C until used. The sputum samples were classed according to their physical appearance and also on a qualitative viscosity index (VI) from 1 to 4. Samples (10 mL) of sputum were studied using a 10-mm diameter Perspex cylinder probe to ensure good sample contact and consistent probe insertion volume. Five replicate adhesion tests were performed on each sample, with the results shown in Table 1.

Table 1 Comparison of visual assessment with quantitative analysis using Texture Analysis

Sample	Adhesiveness (g)	Work of adhesion (g mm)	Cohesiveness (mm)
1. Pale green, VI 2	0.69 ± 0.03	2.54 ± 0.15	7.82 ± 0.30
2. Clear green, VI 2	0.72 ± 0.08	2.60 ± 0.24	8.48 ± 0.95
3. Yellow green VI 3	1.18 ± 0.08	13.02 ± 1.56	26.34 ± 1.73
4. Khaki VI 3	1.10 ± 0.23	6.78 ± 0.69	13.05 ± 0.98
5. Khaki green VI 4	1.23 ± 0.05	11.30 ± 1.49	23.23 ± 1.77
6. Khaki green VI 4	1.24 ± 0.10	12.74 ± 2.03	24.94 ± 5.15

From Table 1 it is clear that viscous samples (as measured visually) had a tendency to be greener; perhaps due to more severe bacterial infection. Values for all three parameters increased with increasing sputum viscosity. The standard deviations for all parameters were comparatively low and of the same order of magnitude as would be expected for conventional rheological investigations. To conclude, these results have shown that it is possible that the TA.TX2 may be developed to provide a numerical description of the consistency of CF sputum.

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091
Mechanical and surface characteristics of copolymers and interpenetrating networks of methacrylic acid and hydroxyethylmethacrylate

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Polyhydroxyethylmethacrylate (pHEMA) is one of the most widely studied hydrogels for use as a medical device biomaterial due, in part, to the known biocompatibility. The physicochemical properties of HEMA may be modified by co-polymerisation with other acrylate monomers, although the mechanical properties of these systems may be inappropriate for clinical use as, for example, drug delivery systems and medical devices. One method by which the mechanical properties of polymer systems may be increased is through the formation of interpenetrating polymer networks (IPN) (Jones et al 1997a). Therefore, this study investigated the mechanical and surface properties of copolymers of HEMA and methacrylic acid (MA) and IPN prepared from these components as biomaterials for medical device applications.

Sheets of copolymer were formed by mixing HEMA with MA (10, 30, 50%) following polymerisation (AIBN 0.5%, 60°C for 18 h) within lined moulds. The IPN was formed by immersion of sheets of pHEMA in a 90% MA solution (0.5% AIBN) following which polymerisation was performed, as described above. The mechanical and surface properties of the copolymers and IPN were analysed, as previously described (Jones et al 1997a). In addition, the uptake of buffer (pH 7.4) was examined using gravimetric analysis whereas the resistance to bacterial adherence was also examined (Jones et al 1997b). The effect of polymer composition on buffer uptake, mechanical and surface properties and resistance to bacterial adherence (a prequel to medical device related infection) were evaluated using a two-way analysis of variance ($P < 0.05$, denoting significance).

Table 1 Physicochemical properties of HEMA/MA copolymers and IPNs

Polymer	Tensile strength (MPa)	% of initial inoculum	Advancing contact angle (°)	% Buffer uptake pH 7.4
Hema	0.47 ± 0.01	0.16 ± 0.01	62.90 ± 2.25	96.93 ± 9.90
Hema/ MA 9:1 copolymer	0.60 ± 0.04	0.14 ± 0.01	52.87 ± 2.04	122.04 ± 11.42
Hema/MA 7:3 copolymer	1.11 ± 0.10	0.13 ± 0.01	49.05 ± 1.25	154.85 ± 4.86
Hema/MA 5:5	3.29 ± 0.60	0.11 ± 0.01	46.15 ± 2.80	184.40 ± 8.50
Hema/Ma IPN (1:1)	4.13 ± 0.09	0.14 ± 0.01	46.78 ± 1.19	16.55 ± 1.86

Increasing the ratio of MA to HEMA in the copolymers significantly increased tensile strength, % elongation, Young's modulus, buffer uptake, yet decreased the advancing contact angle. These properties offered a significant improvement over pHEMA as medical device biomaterials. Significantly, IPNs exhibited greater tensile strength and Young's modulus however the uptake of buffer was markedly lower. The resistance of copolymers of HEMA and MA or IPNs to bacterial adherence was significantly greater than for pHEMA, representing an advantage as a medical device biomaterial. In conclusion, this study highlighted the potential of copolymers of HEMA and MA and, in particular, IPN composed of these components as medical device biomaterials.

Jones, D. S., et al (1997a) *J. Mat. Sci: Mat. Med.* **8**: 713–717
Jones, D. S., et al (1997b) *Biomaterials* **18**: 503–510

092
Rheological synergy within binary aqueous gels of cellulose derivatives

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Polymer–polymer interactions, facilitated by hydrogen bonding, directly influence the mechanical properties of gel networks. Such interactions often result in excluded volumes and ordered structures thus forming interpolymer complexes (Sivadasan et al 1991). Accordingly, mutli-component interactive gels may be formulated to possess unique mechanical properties that cannot be obtained using individual polymer components. Several studies concerning the rheological/mechanical properties of gels composed of hydroxyethylcellulose (HEC) or sodium carboxymethylcellulose (NaCMC) (e.g. Jones et al 1997), have been reported although there have been no reports concerning the mechanical/rheological properties of gels composed of binary mixtures of these two polymers. Therefore this study describes the physical behaviour of single-component gel systems composed of either HEC or NaCMC and, in addition, binary gels. From this study, information concerning the interaction between HEC and NaCMC and the rheological and mechanical implications of this interaction may be derived.

Mono- and binary component systems were prepared by dissolving the required mass of HEC or NaCMC (10% w/w total solids) in distilled water with the aid of mechanical stirring. The samples were analysed by dynamic and continuous shear rheology using a Carri-Med CSL^2-100 rheometer, as previously reported whereas texture profile analysis (TPA), of all formulations was performed as previously described (Jones et al 1997). All measurements were performed at least in quadruplicate. The rheological properties of the various gel formulations were statistically compared using a one-way analysis of variance with an associated post-hoc test (Tukey's HSD). ($P < 0.05$ denoted significance.)

Table 1 Rheological parameters of representative gel systems

HEC/NaCMC (% w/w)	Relaxation exponent[a]	
	G' (pa.s)	(1Hz)
0/10	0.45 ± 0.00	407.02 ± 15.62
2/8	0.21 ± 0.00	1590.57 ± 43.32
10/0	0.30 ± 0.00	8110.25 ± 81.13

[a]Derived from the power law model

Increasing the concentration of HEC or NaCMC significantly increased all mechanical parameters (Table 1), with the exception of tan δ, observations that are consistent with increased polymer entanglement. For example, G' of HEC ranged from 63.71 ± 4.76 (% w/w) to 8100.25 ± 81.13 (10% w/w) whereas, for NaCMC, G' ranged from 0.36 ± 0.05 (% w/w) to 407.02 ± 15.62 (10% w/w). The relaxation exponent of gels composed of 10% w/w HEC was significantly less than for systems composed of 10% w/w NaCMC, suggesting, increased interaction between polymer chains. Relaxation exponents for all binary mixtures were less than those of the individual polymer gels. This suggests that there is increased cross-linking within the polymer blends. A minimum relaxation exponent was observed for the binary gel containing a HEC weight fraction of 0.2. This gel exhibited a storage modulus that was frequency independent; indicative of a cross-linked rather than an entangled system. According to Catsiff (1962), the observed binary gels exhibited a positive deviation at all blend ratios for all mechanical parameters indicative of an associative interaction. In conclusion, this study has illustrated the interaction between HEC and NaCMC in an aqueous vehicle and the effects of blend composition on the resultant rheological and mechanical properties. By appropriate alteration of blend composition, gels may be prepared that offer unique mechanical properties. For this reason such systems show initial promise as platforms for drug delivery systems.

Catsiff, E. H. (1962) *J. Appl. Polym. Sci.* **6**: 530
Jones, D. S., et al (1997) *Int. J. Pharmaceutics* **151**: 223–233
Sivadasan, K., et al (1991) *Colloid Polym. Sci.* **269**: 131

093
An examination of the mechanical and surface properties of novel p(HEMA)-chitosan polymer blends

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Chitosan is a natural non-toxic, biocompatible, biodegradable polymer, which is prepared by the N-deacetylation of chitin. It has been proposed for use as a medical device biomaterial due to its biodegradable, non-allergic and inherent antimicrobial properties (Muzzareli et al 1990). Polyhydroxyethylmethacrylate (pHEMA) is a hydrogel that is used as a component of medical devices such as urethral catheters, soft contact lenses and wound dressings. p(HEMA) is capable of swelling to a predetermined volume in biological fluids and is resilient, pliable, chemically stable, lubricious and biocompatible (Peppas et al 2000). In this study, blends of chitosan and pHEMA have been prepared to produce biomaterials of acceptable biocompatibility which possess optimal mechanical and antimicrobial properties.
Initially, sheets of p(HEMA) were formed by free radical polymerisation (AIBN, 60°C for 18 h) using lined moulds. p(HEMA) was incubated in solutions of chitosan (1 or 3% w/w) for 24 h at 37°C. The sheets were then cross-linked by immersion in solutions of glutaraldehyde (0.125–0.25% w/w) for 4 or 18 h. The tensile properties and advancing/receding contact angle of the hydrated blends and pHEMA were examined. Bacterial adherence was evaluated as previously reported (Jones et al 1997), using a strain of *Escherichia coli* isolated from a retrieved urethral stent. The effect of polymer composition on the mechanical and surface properties and bacterial adherence were evaluated using a one-way analysis of variance ($P < 0.05$, denoting significance) (Table 1).

Table 1 Mechanical and surface properties and resistance to bacterial adherence of various of p(HEMA)–chitosan blends

Polymer	Tensile strength (MPa)	Elongation (%)	% of initial inoculum	Advancing contact angle (°)
Hema	0.47 ± 0.01	99.77 ± 1.27	0.16 ± .01	62.9 ± 2.25
Hema/Chit 1% Glut 0.25%	0.29 ± 0.06	95.45 ± 5.22	0.15 ± .01	57.4 ± 2.38
Hema/Chit 3% Glut 0.25%	0.16 ± 0.02	52.96 ± 10.35	0.10 ± .01	50.5 ± 1.96
Chitosan 100%	0.06 ± 0.001	17.66 ± 2.55	0.10 ± .01	34.7 ± 2.17

Data are expressed as means ± s.d.

These results show that the mechanical properties of chitosan were significantly enhanced by the formation of blends with pHEMA. The formed blends offered a range of contact angles intermediary between pHEMA and chitosan and exhibited similar resistance to bacterial adherence to chitosan films. Using confocal laser scanning microscopy it was shown that the two polymers existed independently and confirmed the presence of a blend. In conclusion, due to the mechanical and surface properties of the blends of pHEMA and chitosan and their resistance to microbial adherence (the initial step in medical device related infection) these materials offer initial promise as medical device biomaterials.

Jones, D. S., et al (1997) *Biomaterials* **18**: 503–510.
Muzzareli, R., et al (1990) *Antimicrob. Agents Chemother.* **34**: 2019–2023
Peppas, N. A., et al (2000) *Eur. J. Pharm. Biopharm.* **50**: 27–46

094
The use of DVS-NIR to characterise the different crystal lattices of theophylline

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It has been reported (Lehto & Laine 2000) that anhydrous theophylline converts to the monohydrate form at room temperature and high relative humidities (> 80% RH). At low relative humidities (< 20% RH), the monohydrate is dehydrated to a form which differs from the initial anhydrate (Phadnis & Suryanarayanan 1997). The aim of this study was to examine whether the combination of the techniques of Dynamic Vapour Sorption (DVS) (Surface Management Systems) and Near Infrared (NIR) (Foss NIRSystems) allows the study of these changes in forms to be undertaken.
The effect of subjecting anhydrous theophylline (Sigma) to changes in relative humidity on both the mass and the NIR spectra was determined at 25°C. At 95% RH the sample gained approximately 9% mass (equivalent to 1 mole of water), indicating that the monohydrate may have formed. When this presumed hydrate was subjected to 5% RH the mass dropped, again by 9%, suggesting a further change of morphological state (the dehydrate).

Table 1 Presence of major peaks in the three theophylline lattices

Main peaks/nm	Anhydrous	Hydrate	Dehydrate
1440	Yes	No	Yes
1478	No	Yes	No
1660	Double peak	Single peak	Single peak
1710	Yes	No	No
1725	No	Yes	Yes
1972	No	Yes	No
2260	Double peak	Single peak	Single peak
2322	Yes	No	No
2343	No	Yes	Yes

NIR spectra of the different theophylline forms were compared (Table 1). The hydrate spectrum was consistent with the presumed formation of a monohydrate. The sharp (negative) peaks at 1478 nm and 1972 nm were only present on the spectra for the hydrate. They probably correspond to the –OH deformation conformation signifying the presence of the monhydrate. Furthermore the peaks at 1660 nm and 2260 nm are seen as single peaks for the hydrate but as a double peaks in the anhydrous lattice.
The spectra for the original anhydrous material and that formed by dehydration of the monohydrate both lack the characteristic –OH deformation at approximately

1972nm, thus proving that neither is a hydrate. However, the spectra for the dehydrate contains some peaks that are found on the theophylline monohydrate spectra. These are at 1660, 1725, 2260 and 2343 nm. The data is therefore consistent with the earlier finding that a dehydrated hydrate differs from the original anhydrous state, and show that DVS-NIR is a powerful tool in studies of changes in physical form.

The combination of DVS and NIR data therefore is valuable in aiding the characterisation of different crystal lattices of a powder, which should aid the understanding of how materials of pharmaceutical relevance may behave under environments they may meet during processing.

Lehto, V., Laine, E. (2000) *Pharm. Res.* **17**: 701–706
Phadnis, N. V., Suryanarayanan, R. (1997) *J. Pharm. Sci.* **86**: 1256–1263

095
The use of DVS/NIR in the analysis of protein stabilisation by carbohydrates

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Protein formulations are commonly produced in solid form due to the tendency of proteins in solution to degrade. The hydration layer surrounding proteins is lost in the final stages of the drying process (Prestrelski et al 1993). This can result in denaturation of the protein structure and hence loss of activity (Arakawa & Timasheff 1982).

Stabilising additives such as carbohydrates are used to protect proteins from denaturation during drying. It is widely accepted that the stabilising additive must be amorphous to confer such stability, however the mechanism of stabilisation is a matter of debate. Franks et al (1991) proposed that protein molecules are immobilised within a glassy matrix. Others have stated that the amorphous state of the stabilising additive alone is not enough to confer protein stability upon drying (Chang et al 1996). The water replacement hypothesis proposes that as water is removed from the protein hydration shell in the final stage of drying; sugar molecules hydrogen bond with polar groups of the protein to conserve its native conformation (Carpenter & Crowe 1989).

This project aims to examine the water distribution between amorphous carbohydrates and proteins to try to elucidate the mechanism of carbohydrate-protein stabilisation upon desiccation.

The unique hyphenated technique of dynamic vapour sorption (DVS, Surface Measurement Systems, UK) and Near Infrared Spectroscopy (NIR, Foss NIRSystems, USA) was used to study the behaviour of two model proteins, hen egg white lysozyme and catalase from bovine liver, after spray drying with the disaccharides, lactose and trehalose. The spray-dried products were exposed to varying humidity within the DVS instrument and the mass change of the sample recorded with simultaneous measurement of near infrared spectra. The influence of protein:disaccharide ratio was also examined.

Preliminary water sorption/desorption data indicate distinct differences between formulations with differing protein:disaccharide ratios suggesting inhibition of disaccharide crystallisation. Formulations containing an excess of the disaccharide show mass loss at higher relative humidity. However, those containing an excess of the protein fail to show mass loss, suggesting that crystallisation has not occurred. NIR spectra agree with this hypothesis, with marked differences in the peak for –OH deformation at approx. 1934 nm. Differences in the NIR spectra between 2000–2400 nm are indicative of protein conformational change and may be used to assess the degree of protein stability in each formulation.

The combination technique of DVS/NIR has been shown to be a useful tool in the study of protein-carbohydrate stabilisation. Further work will aim to utilise other thermal analytical techniques to further characterise these systems and determine ideal stabilising agents for use in protein drug delivery.

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096
Liposome–alginic acid formulations for oral delivery: stability in gastrointestinal fluids

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The suitability of alginates as systems for oral delivery has been investigated in recent years. Vlachou et al (2000) examined the factors determining swelling behaviour in a number of polymers, and concluded that for alginates pH is less of a dominant factor than water solubility. This therefore, opens the possibility of using alginic acid in formulations intended for oral delivery of macromolecules.

Machluf et al (1996) developed a combined alginate–liposome system, with the alginate component present as an encapsulating calcium-cross-linked alginate microsphere. The aim of this study was to evaluate the stability of liposomes loaded with alginic acid (AA; low viscosity) gel in three different gastrointestinal (GI) fluids. Liposomes composed of phosphatidylcholine (PC; Lipoid) and cholesterol (CH; Sigma; 2:1 molar ratio) entrapping 1 % (w/v) FITC-BSA were prepared by the Reverse-Phase Evaporation Vesicle (REV) method (Szoka & Papahadjopoulos 1978). The required concentration (1.0 and 0.1% w/v) of AA was added as the aqueous phase during REV formation, thus forming loaded liposomes, using a method similar to that used by Perugini et al (2000).

The GI fluids in question were simulated gastric fluids (SGF, Sigma) with 0.32% (w/v) pepsin (5536 U mL^{-1} activity; Sigma), bile salts (BS, sodium cholate and deoxycholate, 10 mM final concentration when added to each liposome sample; Sigma) and Pancreatin (P, Sigma) with calcium chloride (5 mM) to facilitate enzyme activation (0.1 U mL^{-1} lipase, 1.25 U mL^{-1} protease and 1.25 U mL^{-1} amylase activity). For each of the GI fluids, a liposome sample was added in a 1:1 volumetric ratio to give a total volume of 1 mL. Results were assessed in terms of entrapped fluorescence (Table 1).

Table 1 EE % of FITC-BSA following 1 h incubation in GI fluid

	Loaded 1.0%	0.1%	Control
SGF	0.37	1.14	0.44
	(0.05)	(0.04)	(0.03)
BS	8.01	10.15	11.40
	(0.27)	(0.14)	(0.15)
P	50.20	44.33	32.13
	(0.90)	(0.32)	(0.94)

Values in parentheses denote s.d.

AA-loaded liposomes displayed the greatest stability in pancreatin with respect to EE %, despite the presence of lipase. Liposomes loaded with 1.0% (w/v) AA retained more FITC-BSA than those formulated with 0.1% (w/v) AA after 1 h incubation. A parallel study conducted with coated liposomes, revealed that consistently less FITC-BSA was retained, supporting the hypothesis that when less AA is used in formulations there is a reduction in encapsulation efficiency, indicative of reduced stability.

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097

Investigation of the effects of excipients and plasma treatment on the dissolution rate of griseofulvin and frusemide (furosemide)

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Excipients are usually inert substances used in drug formulations to enhance dissolution of drugs, assist drug delivery, control drug release rates and enhance flow characteristics and the compressibility of the drug substance (Florence & Attwood 1998). This communication describes the use of plasma treatment of excipient-containing formulations to enhance drug release from compressed tablets.

The drugs (300 mg) were mixed with 2 mL of the solution of the excipient (2% for starch and 5% for PVA) in 24-plate wells, which were frozen overnight. The water was then removed by lyophilisation and the mixtures analysed by FTIR, DSC and SEM to investigate any possible interactions between the drug and polymer and to assess the degree of coating of the drug by the polymer. The freeze-dried mixtures were compacted at a pressure of 5 tonnes using a KBr press. The compacts were treated using oxygen plasma at 10 W for 15 s. Dissolution was carried out using the paddle apparatus method using HCl with 2% SDS and phosphate buffer as the dissolution media for griseofulvin and frusemide (furosemide), respectively. Contact angle measurements were carried out using the sessile drop technique with the contact angle being measured every second for 10 s.

Both PVA and starch were found to decrease the dissolution rate of griseofulvin and furosemide but in the case of furosemide the dissolution rate of the plasma treated furosemide compacts containing 5% PVA was slightly higher then the untreated samples but still lower than that of the drug alone. In the case of griseofulvin with 2% starch and 5% PVA the dissolution rate of the drug was found to decrease; with furosemide, only PVA was found to decrease the dissolution rate of the drug. The contact angle of griseofulvin with 2% starch was found to be the same for the untreated and treated samples whilst the contact angle of plasma treated furosemide with 5% PVA samples was found to be half that of the untreated samples. No interaction was indicated between the drug and polymer by both the FTIR and DSC analysis.

In conclusion, neither starch nor PVA were found to interact with either drug and in both cases the presence of the additional matrix decreased the dissolution rate of both griseofulvin and furosmide. Plasma treatment was however found to modify the wettability of the PVA-containing compacts causing an increase in the dissolution rate of furosemide with 5% PVA compared with the untreated.

This work was kindly supported by a BBSRC Industrial CASE Studentship Award with GlaxoSmithKline in memory of Chris Banton.

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098

An investigation of the use of oxygen plasma as a possible technique for increasing the solubility of frusemide (furosemide)

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Improving the dissolution of poorly soluble drugs represents a major challenge to the pharmaceutical industry (Wotton 2001). Various methods have been previously used for improving drug dissolution including the use of surfactants, complexing agents, co-solvents and the use of different drug forms. Plasma irradiation is widely used in the polymer, automotive and biomaterial industries to increase the wettability of surfaces. As wetting is the preliminary step to dissolution it has been postulated that increasing the wettability of a drug may lead to increased rate of dissolution. This communication describes an investigation into the potential use of this technology to enhance dissolution of frusemide (furosemide), a diuretic with a limited aqueous solubility of 0.029 g L^{-1}.

Compacts of furosemide (300 mg) were produced using a stainless-steel die and punch assembly which was placed into a KBr press, under a pressure of 5 tonnes under vacuum. Half the compacts produced were plasma-treated with an oxygen plasma at 80 W for 15, 30 and 60 s. Dissolution of the plasma-treated and untreated compacts were carried out using the paddle apparatus method (BP 1999). Dissolution was carried out at 37°C using 0.1 M HCl (1 L). Samples were taken at 10-min intervals. The absorbance of the samples was measured at 270 nm. The wettability was assessed by contact angle measurements using the sessile drop technique. A syringe was used to place a drop of pure water onto the compact. The contact angle was measured every second for a period of 10 s. Untreated and plasma treated samples were analysed by SEM at × 5000 magnification.

Plasma treatment was found to lower the equilibrium contact angle from approximately 50 to 35° although the dissolution rate was not significantly reduced with respect to the untreated samples. SEM analysis indicated that plasma treatment for 30 s caused fusion of the surface of the compact thus decreasing the dissolution rate. Further plasma treatment was shown to increase the dissolution rate possibly reflecting an increase in oxidation and hence wettability of the fused surface.

Therefore even though plasma irradiation increases the wettability of furosemide compacts it does not seem to have any potential as a technique for increasing the rate of dissolution of poorly soluble drugs.

This work was kindly supported by a BBSRC Industrial CASE Studentship Award with GlaxoSmithKline in memory of Chris Banton.

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Osteointegrative anti-inflammatory phosphatidylserine coatings

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Phosphatidylserine (PS) is a calcium-binding phospholipid liberated by osteoblasts in bone extracellular matrix where it plays a major role in mineralization. Recently, Santin et al (2001) demonstrated that PS-coated medical-grade titanium induces the rapid formation of a mineral phase in simulated body fluid (SBF). Concerns over the use of this coating arise for two reasons: the potential inhibition of the mineralization process by the adsorption of plasma proteins in-vivo; and the receptor role of PS for macrophages during cell phagocytosis. This work analyses the mineralization of a PS-coated commercial dental fixture in a clinically-reflective model including plasma proteins. The inflammatory response towards uncoated titanium and PS-coated materials was followed by adsorption of the C3 fragment of complement and macrophage adhesion.

Titanium alloy fixtures (Plan1 Health, Italy) were dip-coated under rotation into a 111 mM synthetic dilauroylphosphatidyl serine (DLPS) (Avanti Lipids Polar, USA) chloroform solution for 20 s and the samples kept at room temperature overnight to ensure complete chloroform evaporation. Uncoated and PS-coated implants were incubated for 1 h in human plasma, washed 3 times with deionised water and incubated in SBF (Radin & Ducheyne 1993) for 1 h at 37°C. Specimens were prepared for scanning electron microscopy (SEM) and elemental analysis (EDX) by a standard procedure. Uncoated titanium coupons (SAMO, Italy) as well as samples coated with pure PS and with a phosphatidylcholine (PC):PS:cholesterol (C) (7:2:1) formulation were incubated for 30 min in human plasma, washed 3 times with water and the adsorbed proteins eluted with 2% (w/v) sodium dodecyl sulphate. Supernatants were dialysed overnight against water, freeze-dried, resuspended in a non-reducing sample buffer for electrophoresis and

analysed by Western blot for the C3 fragment of complement. Human peripheral blood monocyte/macrophage adhesion was also evaluated by incubating the specimens with 10^5 cells for 3 h at 37°C. The fixed specimens were analysed by SEM.

SEM analysis clearly showed the formation of a PS matrix in the trough of the fixture thread after incubation in SBF. EDX demonstrated the complete calcium-phosphate mineralization of the matrix indicating no significant inhibitory effect of the adsorbed proteins. C3 adsorbed on all the tested surfaces as a single band suggesting no fragment activation. Macrophages adhered exhaustively onto control titanium and PC:PS:C coatings as activated giant cells, whereas on PS coatings only a few round-shaped cells were found. These data corroborate previous findings that showed no oxidative burst by the monocytes incubated with PS coatings.

The data collected in this work indicate that because of its calcium-binding and anti-inflammatory potential PS can represent the material of choice for a new generation of osteointegrative coatings.

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A comparison of crosslinkable sulfobetaine and phosphobetaine co-polymers as potential anti-bioadherent coatings

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Various polymeric coatings are used in the medical device industry to enhance the surface properties of medical devices including vascular, orthopeadic and opthalmologic implants; catheters; haemodialysers and bloodbags (Meritt et al 1998). Phosphobetaine-based polymers have been extensively evaluated as anti-thrombogenic and anti-bioadherent coatings (Lloyd et al 1999). More recently, studies have suggested that sulfobetaine polymers may have similar applications in reducing mammalian cell attachment and bacterial adhesion to biomedical materials (Lowe et al 2000). To evaluate the relative performance of the phosphobetaine and sulfobetaine coatings. the mammalian cell attachment and bacterial adhesion to similar phosphobetaine and sulfobetaine copolymers has been investigated.

Poly[2-(methacryloyloxyethyl)-2'-(trimethylammoniomethyl)phosphate, inner salt]-co-(n-dodecyl methacrylate)-co-(hydroxypropyl methacrylate)-co-(3-(trimethoxysilyl)propyl methacrylate) (PC1036) and poly[2-(methacryloyloxyethyl)-dimethyl(3-sulfopropyl) ammonium hydroxide, inner salt hydrate]-co-(n-dodecyl methacrylate)-co-(hydroxypropyl methacrylate)-co-(3-(trimethoxysilyl)propyl methacrylate) (SB1036) were prepared and fully characterised by Biocompatibles Ltd and the Sussex polymer group respectively. The polymers were dip-coated onto 13-mm glass and poly(methyl methacrylate) disks from a methanolic solution and cured in an oven overnight at 70°C. Mammalian cell attachment was determined using 3T3 mouse fibroblasts by incubating the materials with 10^4 cells mL^{-1} for 72 h at 37°C. The disks were then rinsed with PBS and the adherent cells stained using calcein AM solution. The adherent cells in 20 fields of view on each disk were counted under a fluorescent microscope. Macrophage adhesion was assessed using human mononuclear cells. Mononuclear cells (2×10^5) were placed onto each polymer coated disc and incubated overnight at 37°C. The adherent cells were fixed with 4% paraformaldehyde and stained using Sigma Quik-1 staining kit. The number of adherent macrophages were determined for 30 fields on each disk using light microscopy.

Significantly few 3T3 fibroblast cells were found to attach to the PC1036 coated materials than the SB1036 coated materials or the glass controls($P > 0.05$). A significant reduction in macrophage adhesion was observed between PC1036 and both glass and PMMA and between PC1036 and SB1036 on both substrates. No significant reduction in macrophage adhesion between SB1036 coated substrates and either PMMA or glass was observed. This study suggests that the phosphobetaine polymers offer clear advantages over the sulfobetaine polymers with respect to reducing mammalian cell adhesion.

The authors gratefully acknowledge the financial support of BBSRC for this collaborative research programme.

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Betaine siloxanes as potential anti-bioadherent coatings

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Surface engineering offers the potential to reduce bioadherence to materials while retaining the mechanical and physical properties of the underlying substrate. For example, zwitterionic betaine polymers have previously been shown to reduce bacterial adhesion and mammalian cell attachment to various biomedical materials (Lloyd et al 1999; Lowe et al 2000). Highly reactive siloxane-based betaines may offer an alternative approach to the use of polymeric substrates for the functionalisation of surface hydroxylated biomedical materials. This communication describes the synthesis and bioevaluation of phospho- and sulfo-betaine siloxanes for such applications.

The sulfobetaine siloxane (SBDAMS) was prepared by reacting 3-(dimethylamino)propyltrimethylsilane (DAMS) with 1,3-propanesultone. The phosphobetaine siloxane (PBDAMS) was prepared by firstly reacting *n*-butanol with 2-chloro-1,3,2-dioxaphospholane-2-oxide in the presence of triethylamine. The resultant 2-butoxy-1,3,2-dioxaphospholane-2-oxide was reacted with DAMS give the desired product. Glass coverslips (13 mm diameter) were coated by immersion in an methanolic solution of the betaine siloxane (10 mg mL^{-1}). The coated disks were then allowed to dry in a dessicator overnight.

Mammalian cell attachment was determined using 3T3 mouse fibroblasts by incubating the materials with 10^4 cells mL^{-1} for 72 h at 37°C. The disks were then rinsed with PBS and the adherent cells stained using calcein AM solution. The adherent cells in 20 fields of view on each disk were counted under a fluorescent microscope. Bacterial adhesion was determined using a bioluminescence-based biomass assay following incubating the disks with *S. epidermidis*, *Ps. aeruginosa* or *S.aureus* (10^8 cells mL^{-1}) for 4 h.

Mammalian cell adhesion was found to be significantly greater ($P < 0.05$; Student's *t*-test) to SBDAMS than PBDAMS. Bacterial adhesion of *S. epidermidis* and *Ps. aeruginosa* was greater to SBDAMS treated surfaces than PBDAMS treated surfaces. There was however no significant difference between the bacterial adhesion between the PBDAMS and SBDAMS treated surfaces following ncubation with *S. aureus*. These studies indicate that the phosphobetaine functionalised glass surfaces offer greater potential as anti-bioadherent coatings than SBDAMS functionalised surfaces. PBDAMS may therefore have application in the treatment of hydroxylated surfaces to reduce bioadherence in certain medical device applications.

The authors gratefully acknowledge the financial support of BBSRC for this collaborative research programme.

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Investigation of processing parameters affecting the drug homogeneity of a blend formulation processed by high shear wet granulation

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The wet granulation process is commonly used to improve powder flow and compressibility characteristics of a formulation via densification. While low shear is often the method of choice for wet-granulation, pharmaceutical industries increasingly employ high shear as the preferred commercial process due to the shorter processing time, enhanced product containment, reduced steps (one-pot process) and the availability of large scale production facilities (Holm 1997). Compared with conventional low shear granulators, high shear mixers also offer the advantage of employing high agitation speeds, thus ensuring uniformity of drug distribution both of the blend and the final granule product. Despite the inherent advantages of high shear wet granulation, process optimisation still remains complex due to the large number, and interdependencies, of the process parameters. Such parameters also need to be evaluated at a small scale to enable scale-up with minimal bulk consumption.

In this paper, we report an efficient and rapid method for optimising the process parameters of a solid dosage formulation consisting of an active principle, a polymer (polyethylene oxide), and a diluent (xylitol) using a SP1 high shear granulator. A $2^3 \times 3$ experimental design was used in which key process parameters such as impeller speed, excipient pre-screening chopper utility and mixing time were investigated at 3 time points (1, 5 and 10 min). To obtain an estimate of the variability of the process, two of the experimental runs were run in duplicate, resulting in a total of 8 batches. The end points used to determine the effectiveness of the process were based on the Pharmacopeial specification requirements for drug uniformity and potency of blends. Blend samples were analysed using a HPLC-UV method. Statistical analysis of the homogeneity data generated was carried out using analysis of variance techniques using the Genstat package.

Results indicated that impeller speed and blending time had the most significant impact on drug uniformity, whereas the other factors investigated (pre-screening and chopper) had a negligible effect. Unexpectedly, homogenous blends were obtained with low agitation speeds in combination with short blending times. Conversely, high agitation speeds and long blending times produced inhomogeneous blends, indicating that demixing may be occurring. Similar observations specifically related to the granulation step were reported by Vromans et al (1999). In conclusion, optimum processing conditions to obtain blend homogeneity in a high shear mixer were identified by using an experimental design approach. Compared with an empirical or a full factorial experimental design, the fractional method used has shown the clear advantage of reducing the investigation time and the bulk consumption whilst providing useful guidance for an effective process scale-up.

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Effect of cannabis extracts on nocturnal sleep and early morning behaviour in healthy volunteers

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The effects of tetrahydrocannabinol (THC) and cannabidiol (CBD) on the sleep process in young, healthy adults were studies. Mood, performance and sleep latencies during the morning of the day after administration were also studied.

The experiment was performed at QinetiQ Ltd sleep laboratory. Effects on nocturnal sleep, early morning performance, memory and sleepiness were investigated in a randomised, double-blind, placebo-controlled, four-way crossover study. The acute effects of two cannabis extracts; a THC-rich extract (15 mg) and two different 50/50 combinations of THC and CBD, (5 mg:5 mg and 15 mg:15 mg), were investigated in 8 subjects, (4M, 4F; 21–34 years). Sleep was assessed by measuring electroencephalograph (EEG) activity along with bilateral electro-oculograms (EOG) and the submental electromyograms (EMG).

Finished dosage preparations were presented as a Pump Action Sublingual Spray (PASS) in a metered dose container for sublingual application.

THC extract, (15 mg THC), did not significantly alter the sleep process, although in some individuals slow wave sleep, (stages 3 and 4), may be increased. THC at this dose produced impaired memory, increased sleepiness, changes in mood, and reduced latencies to early morning sleep the next morning. When an equal dose of CBD was co-administered with 15 mg THC these effects on memory and sleep latency were no longer observed, though increased sleepiness and mood changes were recorded.

CBD extract (5 mg or 15 mg CBD) when administered with the same doses of THC, modulated slow wave sleep, with no changes in total sleep duration. At the lower dose combination (5 mg:5 mg, THC:CBD) there were no changes in mood, sleepiness, fatigue or performance the next morning. For both combination doses (5 mg:5 mg and 15 mg:15 mg, THC:CBD), there were no changes in performance on the memory tests, except for a reduced reaction time with the lower doses in the digit recall test.

An increase in wakefulness occurred from administration of the higher dose combination (15 mg:15 mg, THC:CBD), but this was not seen with the lower combination dose or with THC extract alone.

Conclusions from the study are as follows: the central activity of THC and CBD appears to differ; THC in the dose range 5–15mg has little if any effect on the sleep process; CBD in the dose range 5–15mg appears to modulate slow wave sleep, when administered with equal doses of THC; CBD may shift slow wave sleep activity from stage 3 to 4; CBD may have alerting properties, which may be minimised when given with equal doses of THC.

CBD may modulate some of the residual effects of THC the morning after the evening dosing, particularly memory impairments and sleep latency.

The co-administration of THC and CBD may have advantages beyond the therapeutic benefits of both drugs given alone.

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Protection of liposomes from lyophilization induced aggregation

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The use of liposomes in marketed formulations is proof of their potential as delivery systems (Naeff 1996). Developments within gene and vaccine delivery have shown that carefully defined population sizes aid in targeting or avoiding body organs (Zelphati et al 1998). With these precise monodisperse or mixed blends of liposome populations comes the requirement for suitable storage to maintain stability for a 2-year shelf life.

Pharmaceutical products with longer shelf lives have improved economic viability, increased with the possibility of storage and handling at room temperature. Freeze-drying allows the transformation of unstable solutions to stable solids. However, stresses imposed on the material during this process often result in damage to the active moieties.

The focus of this work was to elucidate the mechanism(s) for lyophilization-induced damage of macromolecules of gene and vaccine delivery systems. Conventionally, sugars have been used to protect materials and liposomes during lyophilization, but their mode of action is not precisely known. Using the theories of particle isolation (Alison et al 2000), and that of glass transition avoidance, we

set out to develop a rationalised method for universally protecting drug delivery formulations from degradation during freeze-drying. The final formulation should protect a wide range of macromolecules, so that minimum optimisation is required before a lyophilization step.

We have used an atypical protection method, capable of retaining the particle size with equal efficiency to that observed using sugars. The protection of phosphatidyl choline (PC) and PC-cholesterol (PC-chol) liposomes against aggregation has been assessed using laser diffraction (Mastersizer S) (Table 1). Using our developed method, liposomes stored in open vials at room temperature showed no significant change in size after more than forty days.

Table 1 Size distribution data for liposomes before and after freeze-drying and with differing lyoprotectants

Sample	Average modal peak (μm)	Average vol. of particles (μm)	80% of particles are below (μm)
Liposomes before freeze drying	1.08 ± 0.02	1.41 ± 0.61	1.38 ± 0.07
Liposomes after freeze drying	14.90 ± 0.03	14.36 ± 0.47	21.20 ± 0.33
Trehalose:			
7.6% w/v	0.96 ± 0.00	2.59 ± 0.50	3.95 ± 0.63
4.7% w/v	0.96 ± 0.01	2.94 ± 0.40	4.04 ± .19
3.8% w/v	4.66 ± 2.1	3.81 ± 0.09	6.15 ± 0.12
2.80% w/v	5.45 ± 0.40	4.70 ± .46	6.94 ± 0.20
In house:			
0.75% w/v	1.70 ± 0.11	2.08 ± 0.33	2.76 ± 0.21

Data are presented as means ± s.d., n ⩾ 5

With the in house method, 80% of the particles were below 2.76 μm compared with 6.94 μm using 2.8%w/v trehalose, indicating decreased levels of aggregation. Further results in house show a different method of protection to the conventional sugars. Sugars typically prevent transformation to a new specific size. Our method shows a gradual progression to a new size and requires less additives (%w/v) for its protective effect.

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