The influence of porosity upon the distribution of reserpine in calcium sulphate granules†

G. L. NICHOLSON** AND R. P. ENEVER*

Department of Pharmacy, Chelsea College, University of London, London S.W.3, U.K.

Batches of calcium sulphate granules, all of the same size fraction but of widely differing porosities, were prepared from primary granules of different sizes. Each batch was coated with reserpine by spraying a solution of the drug in methylene chloride onto the granules whilst they were tumbled in a coating pan. The relation between the macroporous nature of the granules and their reserpine content was investigated. Low-pressure mercury porosimetry was used to obtain a quantitative estimate of the porosity and macropore size distribution of the granules, and their surface areas were determined by an air permeability method. Linear relations were obtained when the reserpine content was plotted against either the volume of mercury penetrating per gram of granules at 106·5 kN m^{-2} or their surface area.

One possible cause of unit-to-unit variation in drug content of tablets is lack of uniformity in the distribution of drug throughout the granules (Johnson, 1966). Lachman & Sylwestrowicz (1964) investigated how the various stages of manufacture could influence the drug content of tablets. When incorporating a poorly water-soluble drug in tablets prepared by a moist granulation process, they found that the drug was more concentrated in the larger granules. A similar gradation of drug content with granule size was observed by Cox, Ambaum & Wijnand (1968) when preparing tablets of lynoestrenol. Unlike Lachman & Sylwestrowicz (1964), they added an ethanolic solution of the drug to prepared inert granules in an attempt to achieve uniform distribution. Cox & others (1968) suggested that the variation might be due to segregation of the granules before drying, resulting in movement of the larger ones to the surface layers. The active ingredient would subsequently tend to concentrate in this region by capillary action during drying. A second possibility was that the larger granules had more void spaces, and upon drying contained more drug than the finer granules which only had a surface coating.

   We have examined this latter suggestion in greater detail. Batches of inert granules, all of the same size fraction but of widely differing porosities, have been prepared and a solution of reserpine sprayed onto them. The macroporous nature of the granules has been quantitatively examined by low-pressure mercury porosimetry and the results correlated with the drug content of the granules.

MATERIALS AND METHODS

Materials

Calcium sulphate hemihydrate (mean surface-volume diameter 3 μm as determined by Fisher Sub-Sieve Sizer) and reserpine were supplied by British Drug Houses Ltd.

Methylene chloride and isopropyl alcohol were laboratory grade solvents from May and Baker Ltd.

Absolute ethanol was supplied by Fisons Chemicals, and polyvinylpyrrolidone (Luviskol K30) was obtained from Badische Anilin and Soda Fabrik A.G.

Methods

Preparation of calcium sulphate granules. (a) *Primary granules.* Calcium sulphate (500 g) was massed with 250 cm³ of distilled water for 2 min, and the wet mass was passed quickly through an Erweka FAG Granulator (equipped with a No. 22 cutting disc) operating at 250 rev min⁻¹. The granules were dried overnight at 80°, and subsequently screened to separate the —22 + 44, —44 + 60, —60 + 85, and —85 sieve fractions. The fractions were then used to produce the final granules.

(b) *Final granules.* From each sieve fraction of the primary granules, and the original calcium sulphate powder, 100 g samples were taken and massed with 30 cm³ of a 13·4% w/v solution of polyvinylpyrrolidone in isopropyl alcohol for 2 min, granulated through a 16 mesh sieve, and dried at 50° overnight. The five granule batches were screened to obtain the —12 + 22 mesh fractions for use in this study. The —44 + 60 mesh fraction produced from the original powder was also collected.

Characterization of granules

(a) *Low-pressure porosimetry.* A low-pressure mercury porosimeter was used to determine the volume and size distribution of the macropores (150–7 μm pore radius) in the —12 + 22 mesh granules. The apparatus measured the volume of mercury penetrating the porous material at applied pressures up to 106·5 kN m⁻². The volume of mercury penetrating at a particular pressure, p, is equivalent to the volume of all pores with radii greater than radius, r. Radius, r, is given by:

$$r = \frac{-2\gamma \cos\theta}{p} \quad \text{(Washburn, 1921)} \quad .. \quad .. \quad .. \quad (1)$$

where γ is the surface tension of mercury and θ is the angle of contact between mercury and the pore surface (an average value of $\theta = 140°$ has been found for a large variety of porous solids).

The volume and concentration of the polyvinylpyrrolidone solution for the final granulation (equivalent to 4% w/w concentration based on the dried weight of granules) was selected after preliminary experimentation had shown that with smaller quantities the granules were not sufficiently strong to withstand the pressure of mercury in the low pressure porosimetry experiments.

The porosimeter was based on the design of Cameron & Stacy (1960), and only differed in the addition of a 85 mesh stainless steel gauze to the base of the dilatometer bulb (volume 21·198 cm³) to prevent granules entering the mercury reservoir. The volume of mercury penetrating the granules was determined by measuring the height of a mercury column in a calibrated uniform-bore tube (volume coefficient 0·2025 ±

0.0008 cm³ cm⁻¹ at 20°) by means of a cathetometer (± 0.002 cm). The applied pressure was measured with a sealed mercury manometer.

A weighed quantity of granules sufficient to fill the dilatometer was used, and the porosimeter was evacuated to $0.4 - 0.65$ kN m⁻² with a rotary vacuum pump and left under vacuum for 30 min to outgas the sample. Mercury was then allowed to enter the dilatometer bulb at this pressure and permeate between the granules. Their apparent density was calculated from a knowledge of the volume of the empty dilatometer bulb, the weight of granules and the volume of mercury permeating between the granules. The pressure was then increased incrementally to 106.5 kN m⁻² and the corresponding volume of mercury penetrating the granules measured at every stage. Duplicate determinations were carried out for each of the five batches of granules, and the average values plotted in the form of volume of mercury penetrating per g of granules against applied pressure. The pore size distribution was obtained by differentiating this plot and calculating the pore size distribution function, D(r), at suitable pressure values (Ritter & Drake, 1945).

Similar porosimetry measurements were made on glass beads (av. diam. 800 μm) and two sieve fractions of sand (av. diam. 850 and 1200 μm respectively).

(b) *Surface area determination.* An air permeability method was used to measure the surface area of the granules. A known weight of each granule batch was packed into a vertically mounted precision-bore glass tube (0.78 cm internal diameter, having an 85 mesh stainless steel retaining gauze cemented to its lower end) to produce a bed with a porosity of approximately 0.45. Compressed air was fed via needle valve through a MeTeRate glass flow meter to the top of the tube to give flow rates ranging from 300 to 4000 cm³ min⁻¹. The corresponding pressure differences across the bed were measured with a water manometer.

Duplicate results were obtained for each granule batch, and from the data the specific surface was calculated at each flow rate by employing the Kozeny-Carman equation (Gregg, 1961). For all batches of granules, it was found that the specific surface was not independent of air flow rate. Thus, for the comparative purposes of this study, the linear relations were extrapolated to give the specific surfaces at zero flow rate. These values were reproducible to within 5%.

Addition of reserpine to granules

Reserpine was added to each granule batch by spraying a solution of the drug onto the granules whilst tumbling them in a coating pan. A 12 cm diameter glass pan fitted with three equally spaced baffles was rotated at 30 rev min⁻¹, and a 12.5 g sample from a batch of $-12 + 22$ mesh granules was mixed with 5 g of the $-44 + 60$ mesh granules for 5 min. Subsequently, 20 cm³ of a 0.655% w/v solution of reserpine in methylene chloride was sprayed at an even rate into the rotating pan over a period of 20 min from a hand-operated spray gun. The wide spray pattern and the shape of the bowl ensured that there was even distribution of the spray over the whole of the moving granule bed. Finally, a hot air stream was directed into the pan to dry the granules as quickly as possible. This technique was used to avoid the possibility of migration of the reserpine to the surface layers by capillary action during a static drying process. The dry granules were then separated into their two size fractions by sieving and reserved for assay of their reserpine content. Negligible quantities of fines were recovered and this indicated that little granule breakdown had occurred during addition of the spray solution. The concentration of reserpine solution was

chosen so that the granules should have contained 0·75% w/w of the drug, if all 20 cm³ of solution had been incorporated in them. In reality, an indeterminate quantity of solution was lost by rebound from the coating pan, and it was for this reason that the —44 + 60 mesh granules were included to act as a marker for the assay.

Assay for reserpine in granules

For the spectrophotometric assay 100 mg quantities of granules were weighed into 50 cm³ volumetric flasks and made up to volume with absolute ethanol. The flasks were shaken vigorously and allowed to stand for 1 h to ensure complete solution of reserpine. Subsequently, a sample was withdrawn with a syringe and filtered directly into a silica spectrophotometer cell (1 cm path length) through a Millipore Swinex filter. A Hilger and Watts H 700 spectrophotometer was used to determine the concentration of reserpine in solution by measuring at the absorbance maximum of 268 nm. Five assays were carried out on each batch of —12 + 22 granules and associated —44 + 60 granules, their mean reserpine concentrations were calculated and expressed as the ratio of granule to marker reserpine concentration.

RESULTS AND DISCUSSION

When viewed under a low power stereoscopic microscope, the five batches of —12 + 22 final granules were markedly different in appearance. Fig. 1 shows low power scanning electron photomicrographs of the granules. The granules produced directly from the calcium sulphate powder were the least irregular in shape and showed the least surface roughness. The granules made from the —85 and —60 + 85 mesh primary granules respectively showed increasing surface roughness, whilst those made
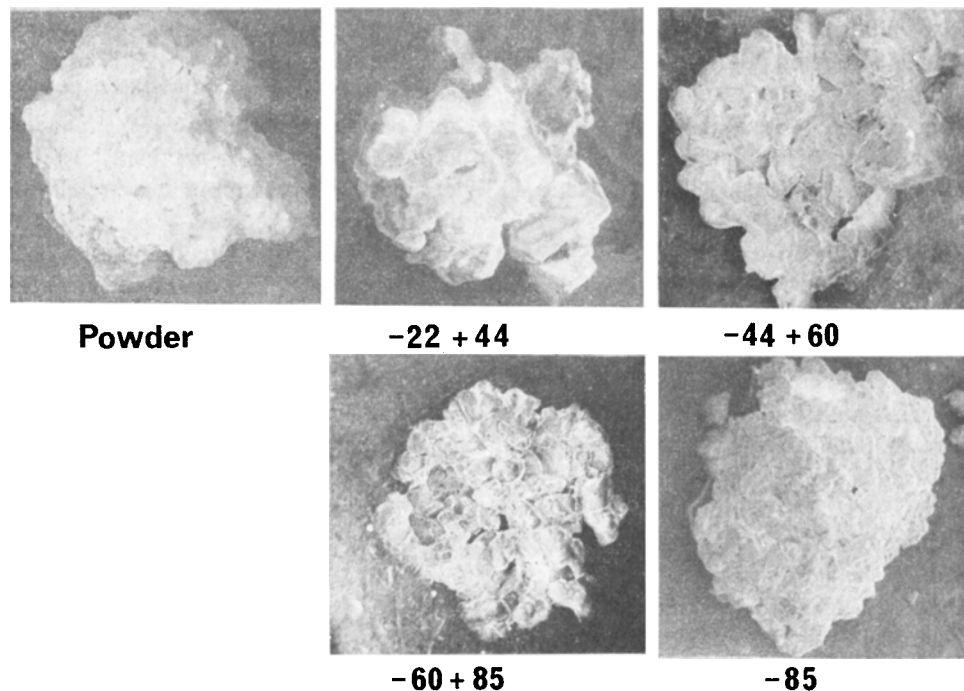

Powder   **−22 + 44**   **−44 + 60**

−60 + 85   **−85**

FIG. 1. Scanning electron photomicrographs of —12 + 22 final granules (× 57 magnification). Captions indicate the size of constituent primary granules.

from the larger primary granules ($-44 + 60$ and $-22 + 44$ mesh sizes) had the most irrregular shapes.

The pressure-volume curves obtained by porosimetry of the granule batches are shown in Fig. 2, and they indicated that there were distinct differences between the
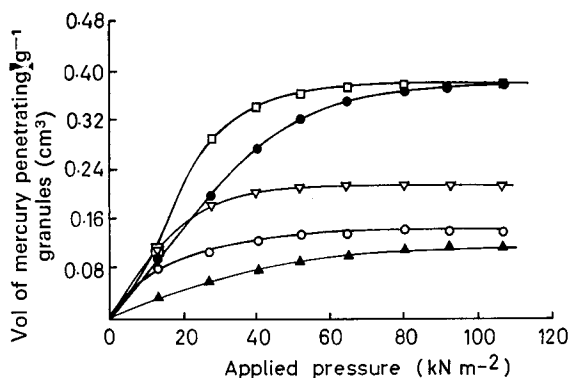

FIG. 2. Pressure—volume porosimetry curves for $-12 + 22$ final granules. Primary granule sizes: ▲ powder; ○ $-22 + 44$ mesh; ▽ $-44 + 60$ mesh; □ $-60 + 85$ mesh; ● -85 mesh.

batches. Within a sample bed of granules packed in the dilatometer bulb of the porosimeter there are two distinguishable pore systems. Firstly, there are intragranular pores within the granules themselves, which have formed during the aggregation of the primary granules, and secondly, there are intergranular pores created between the granules when closely packed within the bulb. Kruyer (1958) and Frevel & Kressley (1963) attributed the rapid penetration of mercury at low pressures to the filling of interstices of this latter type. In the present study the applied pressure used as a starting point for intragranular intrusion of the granules was 7.2 kN m^{-2}.

Since it was the intragranular porosity which was of importance in this study of drug uptake and granule porosity, it was necessary to obtain an estimate of the relative contribution of the intergranular pores to the overall porosity as obtained by low-pressure porosimetry. This was achieved by obtaining the pressure-volume relationships for the glass beads and two sieve fractions of sand whose average particle size fell within the size range of the $-12 + 22$ mesh calcium sulphate granules. The former materials had very little, if any, intragranular porosity and might be expected to exhibit a similar degree of intergranular porosity to the $-12 + 22$ mesh granules. The results, in terms of the volume of mercury penetrating per g of material at 106.5 kN m^{-2}, are shown in Table 1. It can be seen that the volumes of mercury penetrating the glass and sand samples were very low in comparison with those penetrating the granule batches. Thus, for the comparative purposes of this study, it has been assumed that the results obtained with the granules were a reflection of their different intragranular macroporosities and that intergranular porosity could be neglected. Further consideration of the porosimetry results in Table 1 shows that there was a relation between macroporosity and size of the primary granules used. For the three well-defined sieve fractions ($-22 + 44$, $-44 + 60$, $-60 + 85$), there was an increase in macroporosity as the size of the primary granules decreased. This is probably due to the fact that, the smaller the component granules, the greater the number incorporated in the increasingly regular granules and hence the greater number of intragranular void spaces. With the larger component granules, the

Table 1. *Physical characteristics of batches of final* −12 + 22 *mesh granules.*

Mesh size of component primary granules	Volume of mercury* (cm³) per g penetrating at 106·5 kN m⁻²	Specific surface at zero flow rate (cm² g⁻¹)	Apparent density (g cm⁻³)
Powder	0·114	38·5	1·352
−22 + 44	0·140	43·3	1·242
−44 + 60	0·215	48·0	1·104
−60 + 85	0·378	53·6	0·930
<85	0·377	49·5	0·910

* Values for sand (average sizes 850 and 1200 μm) and glass beads (average size 800 μm) were 0·01, 0·007 and 0·003 cm³ respectively.

resulting irregular shape of the final granules meant that a greater proportion of the void space would contribute to the extragranular porosity. Granules produced from −85 mesh primary granules had a slightly lower macroporosity than obtained with the −60 + 85 batch, whilst granules prepared directly from the calcium sulphate powder showed the least macroporosity of all batches. This phenomenon might be explained by the fact that when powder was used to produce the final granules many fine pores were formed, quite a number of which were not penetrated by mercury at 106·5 kN m⁻². Similarly with the −85 mesh primary granules, a proportion of the smallest size material could fill the void spaces and reduce porosity in the macroporous region. Fig. 3 shows some evidence for this hypothesis in the form of a plot of the
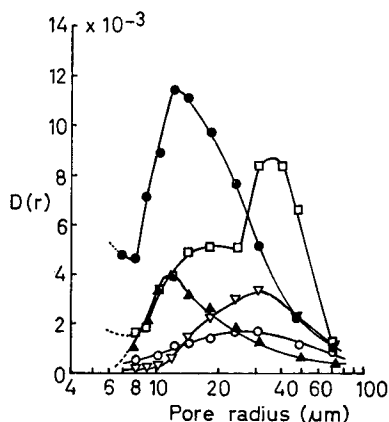

FIG. 3. Macropore size distribution plots for −12 + 22 final granules. Primary granule sizes: ▲ powder; ○ −22 + 44 mesh; ▽ −44 + 60 mesh; □ −60 + 85 mesh; ● −85 mesh.

macropore size distribution function, D (r), against logarithm of pore radius. The maxima in the distributions obtained with the final granule batches made from −22 +44, −44 + 60, and −60 + 85 mesh granules occurred in the pore radius region of 30 to 35 μm, whereas the maxima for the batches produced from the powder and −85 mesh granules were at approximately 12 μm. With these latter batches, the plots indicate that there must be a considerable number of pores with radii less than 8 μm.

Table 1 also shows the results for specific surfaces at zero flow rate and apparent granule density for the five final granule batches. The specific surface values followed the same trend as the macroporosity results, whilst the relation between apparent granule density and primary granule size for the four batches prepared from sieved fractions of primary granules agreed with the work of Awada, Ikegami & Nakamura

(1960) who found that larger apparent densities of granules were obtained when using larger primary particle sizes.

Fig. 4 shows the ratio of the reserpine content (final granule concentration to marker granule concentration) plotted against porosity at 106·5 kN m^{-2} and specific surfaces at zero flow rate respectively for the five granule batches.   The plots confirm
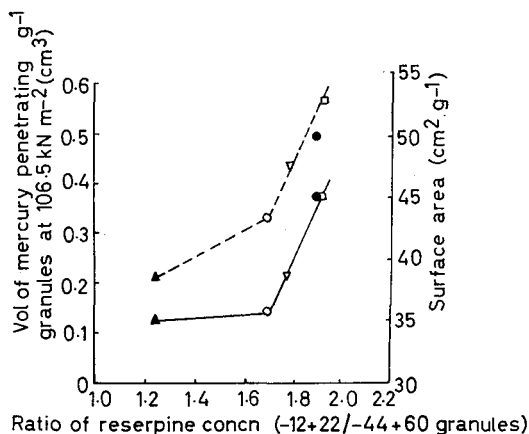

Ratio of reserpine concn (−12+22/−44+60 granules)

FIG. 4.   Ratio of reserpine concentrations (−12 + 22/−44 + 60 granules) plotted against volume of mercury penetrating per g of granules at 106·5 kN m^{-2} (———) and their surface area (- - -). Primary granule sizes: ▲ powder; ○ —22 + 44 mesh; ▽ —44 + 60 mesh; □ —60 + 85 mesh; ● —85 mesh.

the findings of Cox & others (1968) in that the drug contents of the −12 + 22 mesh granules were in every case higher than the smaller −44 + 60 marker granules. Moreover, the present work shows that reserpine concentration could be correlated with the macropore volumes of the final granules.   The trend was for a relatively low concentration of drug in the −12 + 22 mesh granules produced directly from the powder, followed by a rapid rise in content with the batch prepared with −22 + 44 mesh primary granules.   Subsequently, there was a linear increase in reserpine concentration with decrease in primary granule size (increase in macroporosity). The considerable difference in concentration of the granules produced from the powder and −22 + 44 mesh primary granules must be a reflection of the great difference in macropore size distribution (Fig. 3) of the two batches.

This correlation between macroporosity and drug content has been achieved using granule batches all of the same size fraction.   In practice, a normal batch of granules would have a wide distribution of granule sizes and porosities and the findings discussed here may very well be obscured by these other variables.

REFERENCES

AWADA, E., IKEGAMI, Y. & NAKAMURA, A. (1960).   J. pharm. Soc. Japan, **80**, 1567–1573.
CAMERON, A. & STACY, W. O. (1960).   Chem. Ind., 222–223.
COX, P. H., AMBAUM, T. J. G. & WIJNAND, H. P. (1968).   J. Pharm. Pharmac., **20**, 238–239.
FREVEL, L. K. & KRESSLEY, L. J. (1963).   Analyt. Chem., **35**, 1492–1502.
GREGG, S. J. (1961).   The surface chemistry of solids, 2nd edn, p. 236, London: Chapman & Hall.
JOHNSON, C. A. (1966).   The dosage of medicines, p. 30, London: The Pharmaceutical Society of Great Britain.
KRUYER, S. (1958).   Trans. Faraday Soc., **54**, 1758–1767.
LACHMAN, L. & SYLWESTROWICZ, H. D. (1964).   J. pharm. Sci., **53**, 1234–1242.
RITTER, H. L. & DRAKE, L. C. (1945).   Ind. Engng Chem. analyt. Edn, **17**, 782–786.
WASHBURN, E. W. (1921).   Proc. Nat. Acad. Sci., **7**, 115–116.