# Nylon as a Dialysis Membrane

H. B. KOSTENBAUDER*, H. G. BOXENBAUM†, and P. P. DELUCA‡

**Abstract** □ Unique permeability characteristics of Nylon film make it a useful membrane for conduct of dialysis studies or separations which are not readily accomplished with semipermeable films more commonly employed for these purposes. The permeability of Nylon to drugs is not that exhibited by a porous membrane, nor that of many nonporous polymeric membranes. Nylon is relatively impermeable to small molecules and ions, such as water, urea, and sodium chloride, but many less polar, higher molecular weight, unionized species, as well as such ionic compounds as cetyl-, dodecyl-, and ethylpyridinium bromides and sodium naphthalene sulfonate, diffuse readily through Nylon films.

**Keyphrases** □ Dialysis membrane—Nylon □ Nylon membrane—permeability characteristics □ UV spectrophotometry—analysis □ IR spectrophotometry—analysis

Nylon membranes have been used extensively in these laboratories for equilibrium dialysis studies involving a number of drugs, preservatives, and dyes (1–4). Reuning and Levy (5) have further demonstrated the utility of Nylon films by introducing a technique which permits characterization of complexes between small molecules to which the film is permeable, provided that the component molecules traverse the film at sufficiently different rates.

The initial impetus toward an investigation of Nylon film as a potential dialysis membrane was a need for a highly reproducible membrane, less porous than cellophane, which would permit a dialysis study of binding of drug molecules to nonionic surfactants such as polyoxyethylene-20-sorbitan monooleate (mol. wt. approximately 2,000). Of the commercial films examined, Nylon appeared to be most promising for this purpose and, in addition, this membrane exhibited some unusual selectivity characteristics which seemed to merit further investigation. The present work was undertaken to provide an indication of the potential utility and limitations of Nylon as a dialysis membrane by providing data for the rate of transport of some representative compounds across films of this material. Data presented refer to a single lot of commercially available 0.00127-cm. (0.0005-in.) Nylon-6 (polycaprolactam) film.[1]

There has been considerable interest in rates of permeation of dialysis membranes by solutes of biological significance and there have been numerous reports of modification of commercial cellophane membranes and development of new membrane materials to enhance selectivity (6–18). Several terms have been used to describe the permeability of these membranes. Craig *et al.* (6–8, 14, 17) employed the term "half-escape time" which denotes the time required for one-half of the material to escape through the membrane under standardized experimental conditions. Gregor and Kantner (9) calculated a specific permeability constant based on diffusion of solute through pores at a rate governed by the classical diffusion coefficient, $D$, for the solute. Hoch *et al.* (11) measured first-order escape of solutes and described membranes in terms of a specific permeation coefficient, which is the fraction of the hypothetical rate which would be observed for free diffusion in the absence of the membrane (18).

## THEORETICAL

Any of the above means of describing membrane performance is satisfactory for making comparisons of membranes or solutes under a specific set of conditions; however, terms such as half-escape time or first-order rate constant are a function of the experimental design and are not always convenient if one attempts to correlate data from several sources or if one attempts to use such data to predict membrane performance under different experimental conditions. Expression of data in terms of a dialysis coefficient, $D'$, analogous to the classical diffusion coefficient, permits comparison of membrane performance in separations or makes feasible the estimation of time required for equilibration in equilibrium dialysis studies. If one is interested only in the rate at which the solute passes through the membrane it is not necessary to consider porosity (or effective area), since this term is already accounted for in the coefficient.

Indeed, the coefficient is equally applicable to porous or nonporous membranes, although it might be desirable to refer to $D'$ as "permeability" if the term dialysis is to be reserved for those processes involving diffusion through porous membranes. The calculation of $D'$ from data obtained under various experimental conditions is illustrated below. Because a number of the dialyzable compounds under investigation form micelles or aggregates in aqueous solution, it was not always possible to design studies to take advantage of some of the simplifications commonly employed experimentally in determination of dialysis or escape rates.

The expression for Fick's law as applied to transfer of solute across a membrane is:

$$J = dm/dt = D'A(C_i - C_o)/X \qquad \text{(Eq. 1)}$$

where $dm/dt$ is the amount of solute transferred per unit time, $A$ is membrane cross-section area, $C_i$ and $C_o$ are concentrations of solute in Compartment $i$ and Compartment $o$, respectively, $X$ is membrane thickness, and $D'$ is the dialysis coefficient or the permeability.

For a porous membrane, $D'$ is a function of the diffusion coefficient, or diffusivity, $D$, for the solute in a stagnant film of water, and a permeability coefficient, $p$, which is determined by the area and tortuosity of the membrane pores. $D'$ for such a membrane is always less than $D$, and can be expressed as $D' = pD$.

[1] Plaskon Nylon, Allied Chemical Corp., Morristown, N. J.

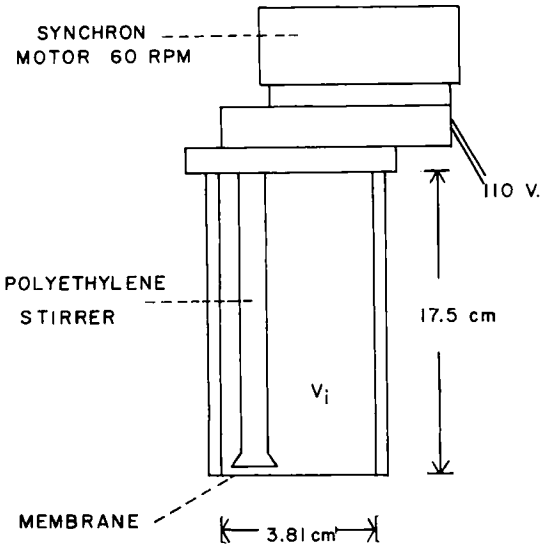


**Figure 1**—*Plexiglas dialysis cell used for escape-rate studies. Membrane is attached to base of cell with waterproof tape. In use, cell is suspended in a stirred bath of approximately 18 l., maintained at constant temperature.*

For transfer across a nonporous membrane or film, solutes must dissolve in the membrane material and $D'$ is a function of the diffusion coefficient, $D_m$, of the solute within the membrane material and the partition coefficient, $K$, which relates solute concentration in the solution phase to the solute concentration in the membrane material. For such a membrane material $D'$ may be either greater or smaller than $D_m$, depending on the choice of solvent for each side of the membrane. With the same solvent on both sides of the membrane, $D'$ can be expressed as $D' = KD_m$. Rodell *et al.* and Garrett and Chemburkar have recently determined both partition coefficients and diffusion coefficients for several drugs in relatively thick Nylon film (19) and in nonporous silastic membranes (20).

In the studies reported in this work, rate of transport of a number of compounds across Nylon film was investigated under nonsteady-state conditions for the purpose of providing data which would facilitate evaluation of the potential utility of this membrane material in dialysis and separation methods. For these systems, the concentration gradient within the barrier is assumed to be uniform, $dc/dx \approx (C_i - C_o)/X$, although it is not independent of time.

**Case I**—If the concentration $C_i$ is not independent of time, but $C_o = 0$ and the volume is constant, the integrated form of Eq. 1 corresponding to these conditions is:

$$\ln C_i = -(D'At)/(vX) + \text{constant}$$

and a plot of $\ln C_i$ *versus* $t$ is linear, with slope $= -(D'A)/(vX)$.

**Case II**—If the concentrations $C_i$ and $C_o$ are not independent of time and the volumes of the two compartments are equal ($v = v_i = v_o$), the integrated form of Eq. 1 corresponding to these conditions is:

$$\ln (C_i - C_o) = -2(D'At)/(vX) + \text{constant}$$

and a plot of $\ln (C_i - C_o)$ *versus* $t$ is linear, with slope $= -2(D'A)/(vX)$.

## EXPERIMENTAL

**Reagents**—Phenol, USP; methylparaben, USP; chloramphenicol, USP; tetracaine hydrochloride, USP; niacinamide, USP; chlorpromazine hydrochloride, USP; gentian violet (methylrosaniline chloride), USP; methyl orange, reagent grade; sodium salt of 2-naphthalene sulfonic acid, recrystallized from water; ethylpyridinium bromide, dodecylpyridinium bromide, cetylpyridinium bromide, laboratory prepared from pyridine and appropriate alkyl bromide (Eastman Organic Chemicals).

**Dialysis Rate Studies**—Film was soaked in several changes of distilled water for 24 hr. prior to use. The nylon film had a stated thickness of 0.00127 cm. (0.0005 in.). Actual thickness was determined as 0.00130 ± 0.00008 cm. for measurement of five separate sections of film. Thickness was calculated from knowledge of density and mass of an accurately measured area of film approximately 20 × 20 cm. Density was determined by means of a density gradient tube, using five pieces of Nylon, each approximately 1 mm.$^2$ in area. A density gradient was established in a 100-ml. graduated cylinder by mixing solutions of bromobenzene in kerosene according to the general procedure described by Jacobsen and Linderstrom-Lang (21). Each tube was equilibrated at least 24 hr. prior to use, and was calibrated by introducing 1-μl. drops of appropriate potassium bromide solutions from a 10-μl. Hamilton syringe. Nylon samples soaked in the bromobenzene-kerosene mixtures for 24 hr. indicated no change in IR spectra, suggesting that the Nylon did not dissolve or swell in the bromobenzene-

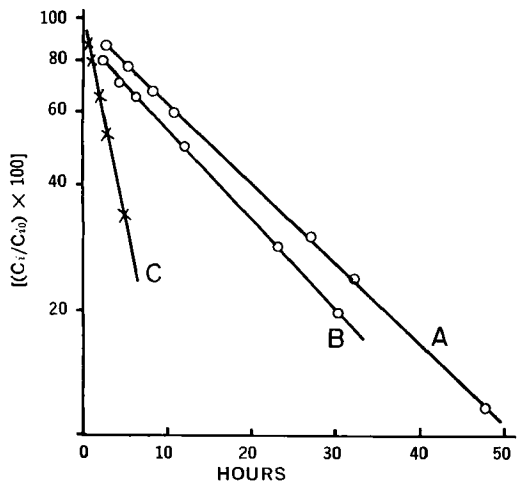


**Figure 2**—*Case I plots for rate of escape of:* A, *phenol; and* B, *methylparaben through 0.5 mil Nylon film. Plot* C *is rate of escape of methylparaben through 27|32 Visking cellulose membrane (Visking Corp., Chicago, Ill.), included for comparison.*

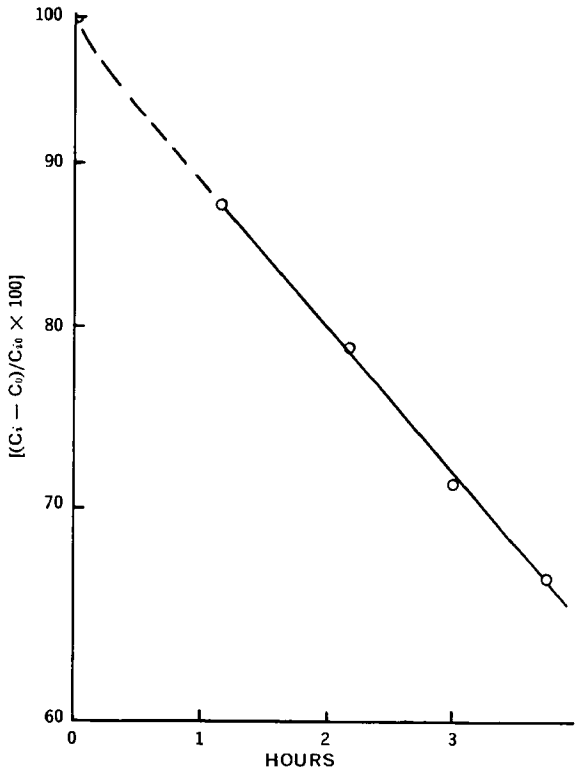


**Figure 3**—*Case II plot for rate of approach to equilibrium for methylparaben across 0.5 mil Nylon film.* $v_i = v_0 = 21.3$ *ml.*

kerosene solutions. Density determinations based upon displacement of water from a pycnometer were in good agreement with those obtained in the density gradient method, but were less precise.

The nature of several of the solutes employed in this study was such that aggregates or micelles were likely to form except in very dilute solution. It was therefore impossible, in most cases, to utilize steady-state conditions which would require a concentration sufficiently high to provide a steady concentration on the high concentration side of the membrane ($C_i$). In most studies 20 or 25 ml. of relatively dilute solution was placed on the high concentration side of a cell, such as that illustrated in Fig. 1, and essentially zero concentration was maintained on the low concentration side of the membrane by bathing the cell in approximately 18 l. of distilled water.

Since direct spectrophotometric analysis was possible, volume ($v_i$) was maintained constant by returning the sample to the dialysis cell after assay.

The determination of dialysis coefficients by study of rate of approach to equilibrium (Case II) is illustrated for one compound, methylparaben, and the results were in agreement with those for the escape-rate study (Case I). The cell used for equilibrium dialysis studies with Nylon film has been described previously (4). Cells were placed on a shaking device operating at a speed of 144 excursions per minute.

All dialysis rate studies were conducted at 30.0 ± 0.5°. Rate of dialysis of all compounds was determined by measuring absorbance of the compound at appropriate wavelength, using a spectrophotometer (Beckman DU). Studies illustrated in this report represent data points replicated in at least two separate studies with membrane samples taken from different sections of a 750-ft.² roll of Nylon film.

## RESULTS AND DISCUSSION

Typical dialysis rate data are presented in Figs. 2 to 8, and the dialysis coefficients for some typical compounds which pass through Nylon at practical rates are listed in Table I. The results indicate that selective permeability of Nylon is not typical of the selectivity which would be exhibited by a porous membrane. Although a number of high molecular weight molecules and ions pass through Nylon readily, small molecules such as urea and small ions such as sodium chloride do not pass at practical rates.

For a series of alkylpyridinium ions, the highest molecular weight compound, cetylpyridinium bromide, was shown to escape through Nylon more rapidly than the lower molecular weight dodecylpyridinium and ethylpyridinium bromides (Fig. 6). Exactly the opposite order of escape rate is observed for the dialysis of these ions through cellophane (22). A similar relationship was noted for several phenols, the higher molecular weight methyl *p*-hydroxybenzoate passing through Nylon at a greater rate than the lower molecular weight compound, phenol (Fig. 2).

The graphs illustrating decrease in $C_i$ with time indicate that those materials diffusing through Nylon are strongly adsorbed to the Nylon (as indicated by the nonlinear initial portion of the plots for those compounds for which rate of adsorption on nylon greatly exceeds rate of diffusion through the nylon). Hayashi (23) studied the diffusion of surface active ions in nylon and considered that diffusion involved transfer of the ions from adsorption site to

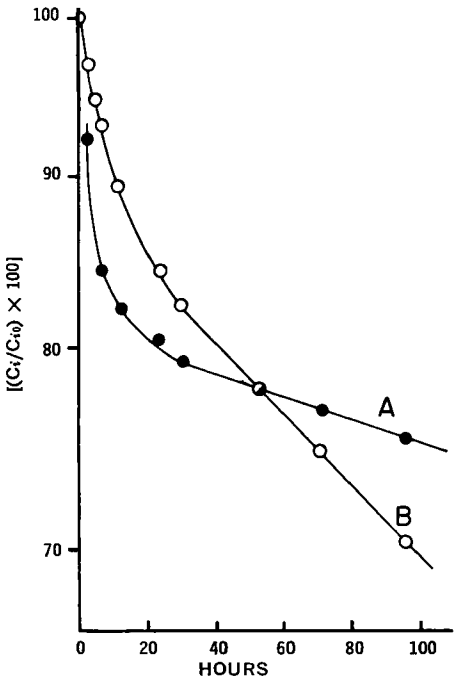


**Figure 4**—*Case I plots for rate of escape of:* A, *tetracaine hydrochloride; and* B, *chloramphenicol through 0.5 mil Nylon film.*

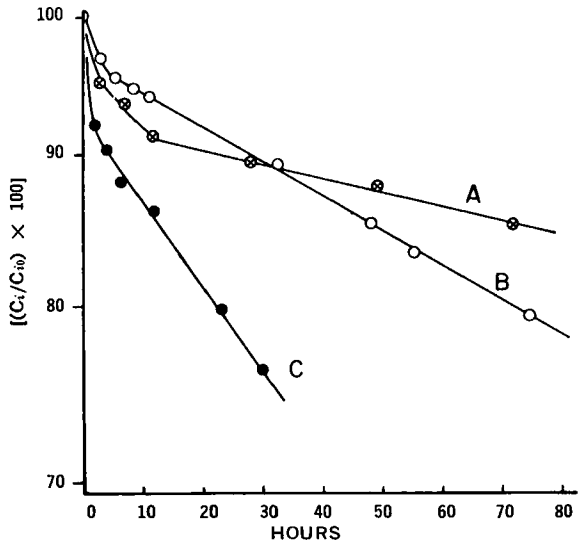


**Figure 5**—*Case I plots for rate of escape of:* A, *sodium naphthalene sulfonate;* B, *niacinamide; and* C, *chlorpromazine hydrochloride through 0.5 mil Nylon film.*

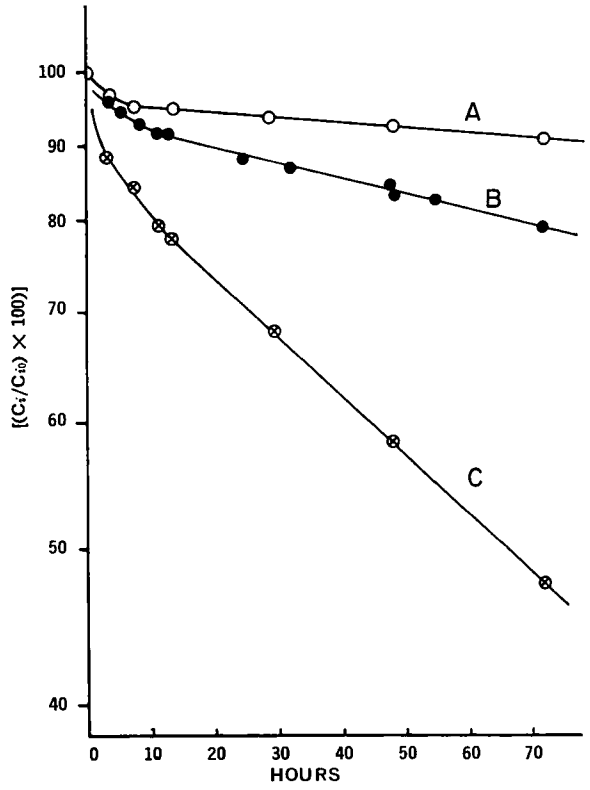


**Figure 6**—*Case I plots for rate of escape of:* A, *ethylpyridinium bromide;* B, *dodecylpyridinium bromide; and* C, *cetylpyridinium bromide through 0.5 mil Nylon film.*

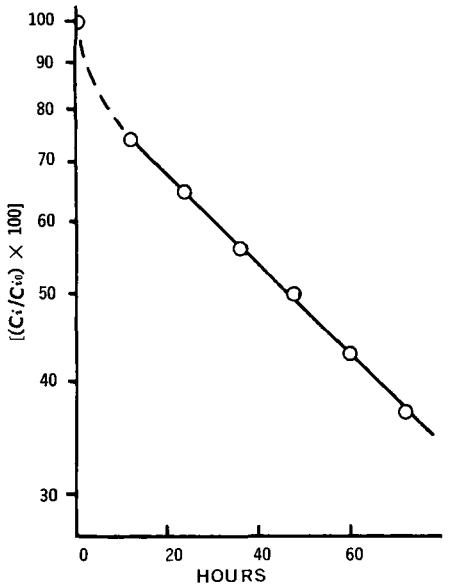


**Figure 7**—*Case I plot for rate of escape of methyl orange* ($C_{i_0}$ = *0.49* × *10*$^{-4}$ M) *through 0.5 mil Nylon film.*

adsorption site within the Nylon. All of the solutes which have been found to pass through Nylon readily are solutes which adsorb most strongly to polyamide type compounds such as polyvinylpyrrolidone in aqueous solution. All rate data were calculated from the linear portion of the plots.

Rodell *et al.* (19) investigated the permeability of Nylon to several phenols and carboxylic acids and determined permeability coefficients ($D'$), partition coefficients ($K$), and diffusion coefficients ($D_m$) for the agents in Nylon films much thicker than those used as dialysis membranes in the present study. Using the data of Rodell *et al.* obtained at 50, 60, and 70°, and assuming adherence to an Arrhenius-type plot for temperature dependency, the extrapolated value for $D'$ for methylparaben at 30° is 48.6 × $10^{-9}$ cm.$^2$/sec. Considering the assumptions stated above, and possible differences in the nylon samples in the two studies, this value is in sufficient agreement with data obtained in the present study to further support the suggestion that rate of transfer of solute through the thin Nylon dialysis membranes is determined by transfer through an intact Nylon film rather than through minute holes or "pores" in the membrane.

Both osmotic and hydraulic transport of water across Nylon membranes is insignificant when these membranes are employed in dialysis studies. Dialysis studies with a 5% aqueous solution of sodium carboxymethylcellulose on one side of a Nylon membrane were conducted for more than 1 week with no increase in volume of the polymer containing solution.

Nylon membranes have not shown changes in permeability in aqueous solutions of pH 2–9 for 1 week at 25°. At pH 7 they have been employed in dialysis studies at temperatures of 5–50°, and they have been maintained at 80° for 24 hr. with no change in permeability. Saturated aqueous solutions of phenol, as well as many nonaqueous solvents, cause denaturation of the membrane.

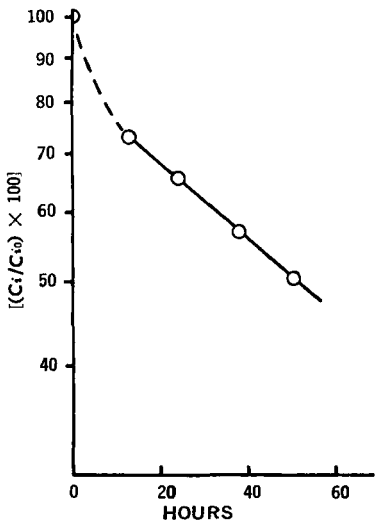


**Figure 8**—*Case I plot for rate of escape of gentian violet* ($C_{i_0}$ = *0.073* × *10*$^{-4}$ M) *through 0.5 mil Nylon film.*

**Table I**—Typical Dialysis Coefficients, Nylon Membrane, 30°

| Solute | λ mμ | Approximate Initial Solute Concn. ($C_i$)[a] moles/l. × $10^4$ | $D'$, cm.$^2$ sec.$^{-1}$ × $10^9$ | Experimental Procedure |
|---|---|---|---|---|
| Phenol | 270 | 5.3 | 34.9 | I |
| Methylparaben | 255 | 0.53 | 38.8 | I |
| Methylparaben | 255 | 0.66 | 36.8 | II |
| Chloramphenicol | 300 | 0.80 | 1.90 | I |
| Niacinamide | 261.5 | 2.8 | 2.07 | I |
| Chlorpromazine HCl | 254 | 0.26 | 5.10 | I |
| Sodium naphthalene sulfonate | 275 | 1.7 | 0.837 | I |
| Tetracaine HCl | 311 | 0.36 | 0.619 | I |
| Ethylpyridinium Br | 259 | 2.0 | 0.539 | I |
| Dodecylpyridinium Br | 259 | 2.0[b] | 1.75 | I |
| Cetylpyridinium Br | 259 | 2.0[b] | 6.68 | I |
| Methyl orange | 465 | 0.49 | 8.80[c] | I |
| Gentian violet | 595 | 0.073 | 10.4[c] | I |

[a] In distilled water. [b] Concentration is below critical micelle concentration. [c] $D'$ would be expected to exhibit concentration dependency for solutes which form aggregates in aqueous solution, and smaller dialysis rate coefficients might be expected for higher concentrations of dye.

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# NOTES

## Pharmacological Effects of Paniculatin—a Glycoside Isolated from *Ipomoea digitata* Linn.

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**Abstract** □ A glycoside (paniculatin), m.p. 134°, $C_{30}H_{54}O_{13}$ has been isolated from the tubers of *Ipomoea digitata* Linn. It elevated the blood pressure, showed a stimulant effect on myocardium and respiration, a vasoconstrictor and bronchoconstrictor effect, a spasmogenic effect on smooth muscles of gut, and also an oxytocic activity. The $LD_{50}$ (48 hr.), with 95% fiducial limits, was found to be 867.4 (755.3–985.1) mg./kg. (*SE* ± 1.03) intraperitoneally in mice.

**Keyphrases** □ Paniculatin—*Ipomoea digitata* glycoside □ Pharmacological screening—paniculatin □ $LD_{50}$ value—paniculatin

The chemical examination of the tubers of *Ipomoea digitata* Linn. (syn. *Ipomoea paniculata*, R.Br.) revealed the presence of a β-sitosterol, neutral compound m.p. 72° (1), a glycoside and fixed oil (2), besides free reducing sugars and mucilage (3). A study of this common indigenous plant of India was undertaken as it has been described as tonic, aphrodisiac, galactogogue, and stimulant in the ancient literature (4–6). Aqueous and alcoholic extracts of the tubers of the plant revealed some interesting actions (7) that required further investigations. In this communication, the pharmacological effects of a glycoside (2, 3) isolated from the tubers of *Ipomoea digitata* have been reported.

### EXPERIMENTAL

Authenticated samples of the tubers of *Ipomoea digitata* were extracted exhaustively with 95% ethanol. Recovery of the solvent and treatment of the residue with ether gave a glycoside (tentatively named paniculatin) as described earlier (2, 3). The glycoside, m.p. 134° (yield 0.02%).

*Anal.*—Calcd. for $C_{30}H_{54}O_{13}$: C, 57.87; H, 8.68; O, 33.45; mol. wt., 622. Found: C, 57.52; H, 8.98; O, 33.50; mol. wt., 618.

On acetylation in the usual way, it gave a tetraacetate, m.p. 65°.

*Anal.*—Calcd. for $C_{30}H_{54}O_{13}$ $(COCH_3)_4$: C, 57.72; H, 7.84; O, 34.44; mol. wt., 790; acetyl value % 21.77. Found: C, 57.51; H, 8.01; O, 34.48; mol. wt., 800; acetyl value % 22.21.

It was found to have three methoxyl groups, while C-methyl was found to be absent (methoxyl group found, 14.50, methoxyl group required 14.75 for three methoxyl groups). It gave a green color with ferric chloride solution and an aglycone on hydrolysis with $H_2SO_4$. It was found to be soluble in water, methanol, and ethanol and insoluble in ether, petroleum ether, chloroform, and carbon tetrachloride.

### METHODS

The pharmacological effects of paniculatin were examined on different organ systems; it has been referred as drug in the following lines. In each case 10 experiments were performed to draw the inference.

Mongrel dogs (6–10 kg.) and cats (2–4 kg.) of either sex, were anesthetized with sodium pentabarbitol (40 mg./kg., intraperitoneally) and carotid blood pressure was recorded. Respiration was recorded by means of Marey's tambour connected to the trachea. Drugs were administered through the cannulated femoral vein. Experiments were repeated after the administration of atropine sulfate (2 mg./kg.), hexamethonium bromide (5 mg./kg.), tolazoline hydrochloride (10 mg./kg.), promethazine hydrochloride (7.5 mg./kg.), and also in bilaterally vagotomized dogs and spinal cats prepared according to the method of Burn (8). In order to study the effect of drug on carotid occlusion response, the common carotid arteries of both sides were exposed in the dog. Bulldog clips were applied on both the carotid arteries for 15 sec., at a point just below the bifurcation of the common carotid into external and internal carotid arteries; this brought about occlusion and a rise in blood pressure. In carotid occlusion studies in dogs the blood pressure was recorded from the femoral artery.

To study the effect of drug on myocardium of different animals, perfusion of the frog's heart *in situ* and isolated frog's heart was