HT content, while animals treated with 6-OHDA showed an insignificant change in brain 5-HT content. Thus the data suggest that pCPA may act through the serotoninergic systems to exert its hypothermic action.

In fact, there is growing evidence suggesting a hypothermic role for brain 5-HT in rats. For example, it has been found that intraventricular administration of 5-HT led to hypothermia in rats[14,15]. Moreover, in this laboratory, we found that elevating 5-HT content in brain with 5-hydroxytryptophan reduced rectal temperature in rats (Lin et al., unpublished data) after peripheral decarboxylase inhibition with R04-4602 at ambient temperatures of both 8 and 22 °C. Also, elevating 5-HT concentration in 5-HT receptor sites or increasing functional 5-HT in brain with inhibitors of 5-HT re-uptake such as Lilly 110140 and chlorimipramine reduced rectal temperature in rats at room temperature and below (Lin et al., unpublished data). This raises the possibility that pCPA may act through the enhanced release of brain 5-HT to induced hypothermia. However, it must be acknowledged that pCPA is used as a 5-HT synthesis inhibitor rather than a 5-HT precursor[5]. It is not known whether the immediate inhibition of 5-HT synthesis after pCPA treatment would induce an enhanced release of 5-HT into synaptic clefts and result in a transient acute hypohtermia. The possible feedback mechanism needs further verification.

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Permeability coefficients of the egg-case membrane of *Scyliorhinus canicula* L.

David J. Hornsey

School of Biological Sciences, University of Bath, Bath BA2 7AY (England), 25 May 1978

Summary. The cleidoic egg-case of the dogfish appears to have a highly porous and permeable outer membrane, the pore radius being computed to be 13.6 Å. It does not present any physiological barrier to small molecules and therefore constitutes an open ionic and osmotic system for the embryo. Being a porous protein membrane it may be of value as a model for molecular transport mechanisms.

The oviparous dogfish, *Scyliorhinus canicula* L. lays egg-cases throughout the year and for the first 3 months of their development, until the embryo is 3 cm long, the eggs are cleidoic[1]. Because of the osmotic importance of urea to Elasmobranchs, both Wood[2] and Baldwin[3] have suggested that the egg-case is impermeant to this solute, it being considered necessary to retain urea within the case for the embryo's survival. The urea molecule has a radius of approximately 2.3 Å which is much the same size as the water molecule. Does this imply that the egg-case is impermeant to water as well? The shark gill has been shown[4] to be relatively impermeant to urea but freely permeable to water. Can the mechanism of urea retention in the living gill be similar to that in the dead collagenous egg-case? In this report, the permeability of the egg-case to water and some selected solutes is established.

3 coefficients of permeability categorise a membrane[5], L_p, the hydraulic coefficient, ω, the diffusional coefficient, and σ_s, the reflection coefficient. ω is related to the conventional permeability coefficient P, by $P=\omega RT$ (R and T being the gas constant and absolute temperature respectively). The volume flux, J_v, brought about by an osmotic gradient, $\Delta\pi_s$ is given by $J_v=-L_p\sigma_s\Delta\pi_s$[6], the negative sign being convention. The reflection coefficient is a measure of the 'leakiness' of the membrane to the solute. When $\sigma_s=1$, s is impermeant and when $\sigma_s=0$, s is permeant.

Methods. Areas of washed membranes were clamped between 2 glass reservoirs containing the experimental solutions. The apparatus was shaken horizontally in a water bath at 293 °K. To measure L_p, a solution of an impermeant solute, polyvinylpyrrolidone PVP ($\Delta\pi_s=21.6$ at) was placed in 1 reservoir with a weighed amount of distilled water in the opposing one. Gravimetrically, J_v was measured and L_p calculated. ω was determined for water with ^3H_2O (0.2 $\mu Ci\,ml^{-1}$) in 1 compartment and assaying its appearance in the water of the opposing one. This experiment returned a value for ω_T (T for tritiated water) calculated from the modified Fick equation:

$$P_T=\omega_T RT=\frac{Q}{\Delta C\cdot A\cdot t}$$

where Q is the total amount of label moved in time t across an area A cm^2 under an isotope gradient ΔC. P_s or ω_s for various labelled solutes may similarly be determined. With the measured value of L_p calculated using PVP, the J_v produced by a permeant solute was used to measure the relative σ_s for that particular solute. Corrections for the unstirred layers[7] of water were unnecessary for a membrane this thick (0.4 mm).

The values for the permeability coefficients for the egg-case are shown in table 1. Whether or not there is an osmotic gradient across the membrane, the permeability coefficient, P_T, for water movement is high compared to the J_v values and is fairly constant indicating that the major contribution to water movement is purely diffusional and not bulk flow. By converting the units of ω_T to those of L_p, by multiplying by the molal volume of water

Table 1. The permeability coefficients, P_T, P_s and σ_S measured under various osmotic gradients

Osmotic gradient $\Delta\pi_s$ (at)	Solute	$P_T(\omega_T RT)$ (cm sec^{-1})	ω_T (of mean) (mole cm^{-2} sec^{-1} at^{-1})	$\omega_T \bar{V}_w$ (of mean) (cm sec^{-1} at^{-1})	Osmotic water flow, J_v (ml cm^{-2} sec^{-1})	σ_s	P_s (cm sec^{-1})	$P_s M^{1/2}$
zero	Sea-water	$1.49 \pm 0.03 \times 10^{-4}$ (n=5)	6.2×10^{-9}	1.12×10^{-7}	-	-	$7.4 \pm 0.05 \times 10^{-5}$ (n=5) (for Na ions)	3.47×10^{-4}
zero	Distilled water	$1.3 \pm 0.04 \times 10^{-4}$ (n=5)	5.41×10^{-9}	9.74×10^{-8}	-	-	-	-
zero	Urea	-	-	-	-	-	$4.66 \pm 0.02 \times 10^{-5}$ (n=5)	3.6×10^{-4}
22.4	Urea	$1.3 \pm 0.05 \times 10^{-4}$ (n=7)	5.41×10^{-9}	9.74×10^{-8}	1.25×10^{-6} (mean from table 2)	0.04	-	-
30.24	Sucrose	$1.33 \pm 0.05 \times 10^{-4}$ (n=5)	5.53×10^{-9}	9.96×10^{-8}	$4.12 \pm 0.3 \times 10^{-5}$ (n=5)	1.06	-	-
23.7	Sea-water	$1.54 \pm 0.05 \times 10^{-4}$ (n=5)	6.41×10^{-9}	1.17×10^{-7}	$2.47 \pm 0.2 \times 10^{-5}$ (n=7)	0.87	-	-

$L_p = 1.27 \pm 0.12 \times 10^{-6}$ ml cm^{-2} sec^{-1} at^{-1} ± SE for n readings on at least 3 membranes (n=7).

Table 2. Volume flows (J_v) through egg-case membranes separating distilled water and molal solutions of solutes

Solute	Molecular weight	J_v(ml cm^{-2} sec^{-1})	σ_s
Acetamide	CH_3CONH_2 59	$7.0 \pm 0.2 \times 10^{-6}$	0.25
Formamide	$HCONH_2$ 45	$8.6 \pm 0.2 \times 10^{-7}$	0.03
Formic acid	$HCOOH$ 46	$-1.67 \pm 0.7 \times 10^{-6}$	−0.06
Methanol	CH_3OH 32	$3.4 \pm 0.5 \times 10^{-6}$	0.11
Methylamine	CH_3NH_2 31	$6.6 \pm 0.2 \times 10^{-6}$	0.23
Oxalic acid	$(COOH)_2$ 90	$-1.3 \pm 0.05 \times 10^{-5}$	−0.46
Thiourea	$CS(NH_2)_2$ 76	$-2.04 \pm 0.1 \times 10^{-6}$	−0.07
Urea	$CO(NH_2)_2$ 60	$1.25 \pm 0.7 \times 10^{-6}$	0.04

Values for σ_s taken from mean values of J_v ± SE for 4 reading each of 2 membranes.

$\bar{V}_w$ (18 ml $mole^{-1}$), it may be seen that the ratio $L_p : \bar{V}_w \omega_T$ is greater than unity, in this case 12.2. The difference between these coefficients measured by bulk flow and isotope methods probably implies that the membrane is porous[8]. In fact, assuming laminar flow through channels, the relationship[9]

$$\frac{L_p}{\bar{V}_w \omega_T} = \frac{r^2 RT}{8 \eta \bar{V}_w D}$$

where r is the channel radius, η is the viscosity of water and D is the self-diffusion coefficient of ^3H_2O in water, holds and the pore radius is 13.6 Å. The egg-case would appear to have a very porous membrane that will allow a large number of solutes through. The solute coefficients P_{Na} and P_{urea} show, as expected, that urea (mol. wt 60) diffuses more slowly than the sodium ions (mol. wt 23). $PM^{1/2}$ (where M is the mol. wt) are similar confirming the relationship $PM^{1/2}$ = constant[10] for this membrane.

What is confusing is that $\sigma_{NaCl} \gg \sigma_{urea}$ when calculated from the J_v values. This implies that Na ions which diffuse faster than urea through the membrane are better able to generate an osmotic flow. To investigate further the properties of molecules capable of producing an osmotic flow, molal solutions of selected solutes were opposed to distilled water in the experimental apparatus and J_v determined. The results are shown in table 2. The ability to generate an osmotic flow in this experiment lies not on molecular size or weight, but, it would appear, on the presence of a CH_2-group. Very low or negative σ_s were returned for compounds lacking these groups. These results imply a solute/solvent interaction more than a solute/membrane interaction.

Discussion. The egg-case of the dogfish would appear to be highly permeable to water, ions and urea. In the case of water, L_p is 13 times greater than that quoted for the frog egg[11] and may be contrasted with the highly impermeable trout egg (3×10^{-10} cm sec^{-1} at^{-1})[12]. Why NaCl will almost generate its full osmotic potential ($\sigma_s = 0.87$), where urea, a similar size molecule with a slower diffusion rate can not ($\sigma_s = 0.04$) is not clear, but it may be related to a charged membrane system. This membrane presents potential for the investigation of transport phenomenon through a charged protein complex and is under further study.

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