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Fisiologia. — *Calculated glucose concentration profile in the intercellular spaces of everted jejunum of rat.* Nota di FRANCESCO ANDRIETTI (*), GIOVANNI ESPOSITO (*), ALIDE FAELLI (*), NEDDA BURLINI (*) e MARISA TOSCO (*), presentata (**) dal Corrisp. V. CAPRARO.

RIASSUNTO. — Sulla base dei dati sperimentali nel digiuno isolato di ratto e utilizzando il modello matematico di Diamond e Bossert è stata calcolata la concentrazione di glucosio negli spazi intercellulari; questa risulta essere di poco più alta di quella presente nello spazio serosale (meno di 2 mM).

The isolated and everted jejunal sac transports some sugars from the mucosal to the serosal side against a concentration gradient. Our aim is to see whether what we have previously named the apparent glucose concentration in the intercellular spaces [2] and what we name here the emergent fluid concentration are much different from the actual glucose concentration along the same spaces.

The basic procedure of the experiment has been reported elsewhere [1], however some additional details will be given here. The everted jejunum of male albino rats (Wistar strain Charles River Italiana, 200–230 g body weight) was cannulated at one end and ligated at the other. The intestine was immersed in 50 ml Krebs-Ringer-bicarbonate solution with 5.5 mM glucose added, gassed with 95 % O_2 and 5 % CO_2: 2 ml of the same solution, with poly ^{14}C-ethyleneglycol (^{14}C-PEG) added as an extracellular marker, was introduced into the cannula. The temperature was kept constant at 28 °C throughout the 30 min of experiment. It has been demonstrated that this temperature is suited to this preparation [2]. Net glucose and Na salt transport per second refer to 1 cm^2 of intercellular surface.

From the cell number of villi per unit length of rat jejunum [3] corrected for goblet cells (30 %) [4] it is possible to calculate the total number of absorptive cells per 100 cm which in turn correspond to 1 g dry weight of intestine [4, 5].

By assuming for the columnar absorptive cell a diameter of $4 \cdot 10^{-4}$ cm and full length of $100 \cdot 10^{-4}$ cm it is easy to obtain the total area of lateral walls of the intercellular space ($2.45 \cdot 10^4$ cm^2) for 1 g dry weight.

The average net sodium salt transport over 8 experiments is (187 ± 34) 10^{-7} μosmoles per cm^2 per sec, whereas that of net glucose and water transport is (19.8 ± 3.7) 10^{-7} μmoles per cm^2 per sec and (6.8 ± 1.5) 10^{-8} cm/sec respectively. The average final sodium salt and glucose concentrations in the serosal side are 274 ± 2 mOsm and 9.1 ± 0.6 mM respectively.

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(**) Nella seduta del 13 maggio 1978.

Net sodium transport does not include the retrodiffusion component towards the mucosal side. On the other hand net glucose transport does not seem to be affected by this component [6]. In order to calculate (see later) the glucose concentration profile in the lateral spaces we have taken

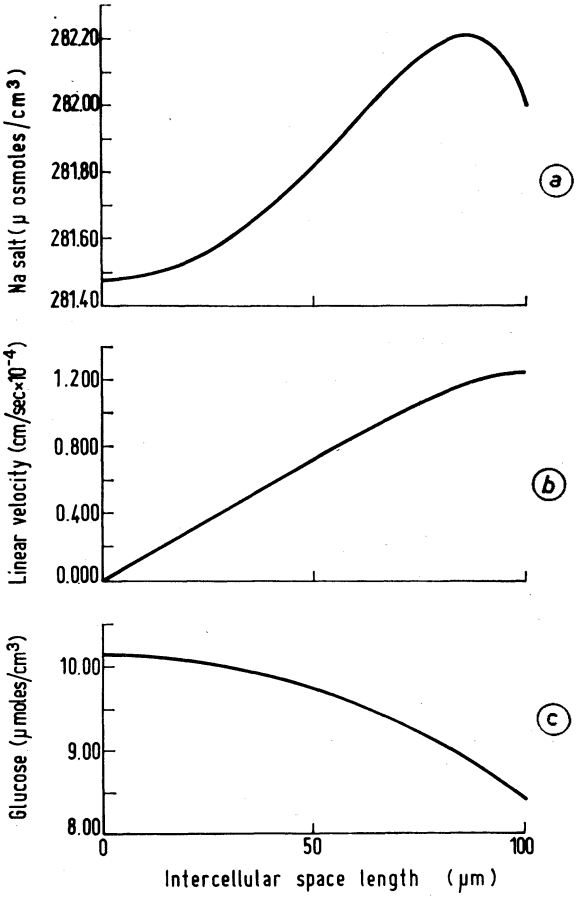

Fig. 1. – (*a*): sodium salt osmolarity profile; (*b*): linear velocity of the fluid; (*c*): glucose molarity profile. Values of experimental parameters: $h = 0.1$ μm, $N_1 = 15.37 \cdot 10^{-7}$ μmoles/cm$^2 \times$sec, $N_2 = 195.30 \cdot 10^{-7}$ μosmoles/cm$^2 \times$sec, $D_1 = 0.5 \cdot 10^{-5}$ cm^2/sec, $D_2 = 10^{-5}$ cm^2/sec, $C_{1_0} = 8.41$ μmoles/cm^3, $C_{2_0} = 282$ μosmoles/cm^3, $C_{cell} = 290.41$ μosmoles/cm^3, $C_{em_1} = 24.43$ μmoles/cm^3, $C_{em_2} = 310.38$ μosmoles/cm^3. The estimated value of P was $0.61 \cdot 10^{-4}$ cm/sec.

into account the results of some single experiments. Figs. 1 and 2 refer to the values of one experiment in which the net glucose transport (N_1, μmoles/cm$^2 \times$sec) and the net Na salt transport (N_2, μosmoles/cm$^2 \times$sec) are closest to the above means (see fig. legends).

Diamond and Bossert [7] have developed a mathematical model to take into account both diffusional and convective components of the transport of one solute along the epithelial intercellular spaces, in order to calculate its concentration profile. We extended their original model to the case of transport of two solutes. In our case, we do not consider the flow system

as a true circular cylindrical channel, but as a space bounded by two parallel rectangular surfaces, with one very long side and the other given by "l" (intercellular space length which is assumed to be 100 μm). The distance between the two surfaces is "h" (intercellular space width; "h" is

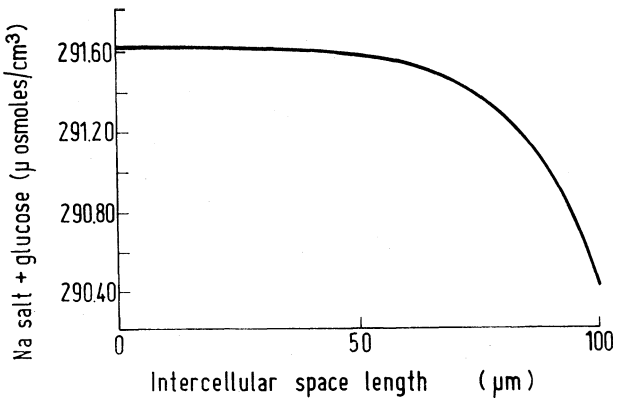

Fig. 2. – Total osmolarity profile.
Same value of parameters as Fig. 1.

assumed to be 0.1 μm). The space is open on the subepithelial side and assumed to be closed at the other end.

Diamond and Bossert's eqn. 1 for two different solutes then becomes

$$2\,N_j/h + D_j\,(d^2 C_j/dx^2) - d/dx\,[C_j\,v] = 0 \tag{1}$$

where $j = 1, 2$ indicates the first (glucose) and the second (sodium salt) solute respectively, "C_j", "D_j" and "N_j" their concentrations, free diffusion coefficients and net transepithelial transports (the latter assumed to be constant on the whole intercellular space surface).

Diamond and Bossert's eqn. 2 becomes

$$dv/dx = (2\,P/h)\,[C_1 + C_2 - C_{cell}] \tag{2}$$

where "v" is the linear velocity of the fluid, "P" the osmotic permeability constant of the intercellular lateral walls and "C_{cell}" the total cellular osmolarity (assumed to be the same as that of the free subepithelial space).

We performed a first integration of eqn. 1 between zero and x taking into account the condition $dC_j/dx = 0$ at the closed end (no solute net flux across the closed end). In this way eqns. 1 and 2 reduce to the system of three first-order differential equations

$$\begin{aligned} dC_1/dx &= (1/D_1)\;[vC_1 - (2\,N_1/h)\,x] \\ dv/dx &= (2\,P/h)\,[C_1 + C_2 - C_{cell}] \\ dC_2/dx &= (1/D_2)\;[vC_2 - (2\,N_2/h)\,x] \end{aligned} \tag{3}$$

with the initial conditions:

(i), $C_j = C_{j_0}$ at the open end, where the values of "C_{j_0}" are the glucose and sodium salt concentrations of the subepithelial space; (ii), the

value of "v", "$v(l)$", at the open end of the space was determined by means of the relation

$$C_{emj} = \frac{2 N_j l}{v(l) h}$$

where "C_{emj}" is the experimental value of glucose or Na salt emergent fluid osmolarity.

Conditions (i) and (ii) allow us to integrate the equations of system 3. The value of the parameter "P" was determined by an iterative procedure in order to obtain $v(0) = 0$ at the closed end of the intercellular space (no water net flux across the closed end).

Integration of the equations of system 3 was numerically performed by the aid of a FORTRAN polyalgorythm for the solution of ordinary differential equations.

In Fig. 1 (*a*, *b*, *c*) the computed results are represented as a function of the space length. The zero value corresponds to the closed end of the intercellular space. Similar results were obtained by using the experimental values of other single experiments.

The value of "P" was found to be $0.61 \cdot 10^{-4}$ cm/sec for the parameter values given in the legend of Fig. 1.

It may seem surprising that the Na salt concentration profile decrease as the distance from the open end increases.

Recently some authors [8] experimentally determined the Na concentration profile in the intercellular space; they found a biphasic pattern of this profile.

The peculiar Na salt concentration profile in our case is presumably due to its diffusion coefficient which is higher than that of glucose. As a matter of fact, the concentration profiles shown by Diamond and Bossert (see also [9]) all present a monotonic decrease.

However, as we are now dealing with two solutes, the result is not absurd, as long as the total osmolarity is still a decreasing function in the direction of the open end (Fig. 2).

As to the behaviour of the glucose concentration profile (Fig. 1, *c*), the glucose concentration in the intercellular space reaches a value which is, at the most, less than 2 mM higher than that present in the serosal space. However, this maximum value must be considered an upper limit due to the assumed absence of water net flow across the tight junctions (closed end); should junctions be open to water flow, the linear velocity at the beginning of the intercellular space would be slightly positive and the glucose concentration presumably lower.

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