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STUDY OF PERMEABILITY OF FISH SKIN

M. A. PARSADANYAN [*], M. S. MIKAELYAN [**]

Chair of Biophysics, YSU, Armenia

Skin permeability of Sevan trout has been studied during 30 *min* in different solutions: in distilled water, in 70% ethanol, in solution of 0.125 *M* KCl. The permeability was measured by methylene blue. It was shown that the skin permeability for this dye changes depending in what solution these shreds of Sevan trout were kept and from which side the shred was tightened. It was also shown that distilled water, solution of 0.125 *M* KCl and ethanol cause structural changes in the skin layers of the Sevan trout, in particular the epidermis.

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Introduction. Still in XX century the problem of out-intestinal absorption (dissolved organic compounds in tank) was significant, i.e. it is an assumption about fish. Fish skin is consisted of three main layers: epidermis, derma and subcutaneous tissue. Epidermis is relatively thin, composed of several layers of cells, mainly of flask-shaped and gland cells, releasing a secretion – mucus. Epidermis is connected to derma weakly. Epidermis of fish skin is composed of living cells through whole volume [1].

J. Homkovich [2] maintained that the isolated skin of carp and tench is able to pass from out to in a solution with 1% concentration of glucose, fructose and pentose by single side.

D. Corde, J. Borbe [3–5] studied the permeability of dace skin (*Leuciscus rutilus*) for ethanol and established that the compound penetrates through the skin in big amounts, the higher is its concentration in the solution and the higher is the temperature, the less is the amount of dissolved oxygen in water.

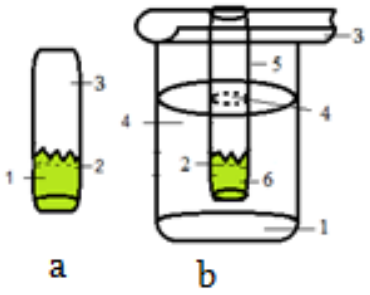
N.S. Stroganov, A.P. Lashmanov [6] assumed that the experiments, carried out on the whole organism do not give an opportunity to completely judge about properties of fish skin in relation to its permeability for different compounds.

Study of fish skin permeability is also important, because recently the problem of water pollution of fishery destination becomes global [7]. Recently connected to environment pollution by technogenesis products, heavy metals, placed specially among polluting materials of freshwater ecosystems, are under a big attention for ecological monitoring [8, 9].

[*] E-mail: marine.parsadanyan@ysu.am
[**] E-mail: m.mikaelyan@ysu.am

Hydrobionts – sea and freshwater organisms, continuously living in water surrounding, are able to accumulate polluting compounds in organs and tissues from environment and can serve as bioindicator for toxicant spread in water. Accumulation of dangerous materials in fish skin creates a threat of their effect on human organism through fish products, used in food [10].

Materials and Methods. Sevan trout isolated shreds were used in experiments. Isolated shreds of Sevan trout were used to study the organic dye permeability, in this case – methylene blue (MB) [11]. Eight parts of the skin were examined. The shreds of the skin, located alternately by fish length lower side line, were cut by scissors. Skin shreds are tightened on expanded ends of glassy cylinder by threads or elastic bands (Fig. 1).



a – glassy cylinder with skin shred:
1 – skin;
2 – threads;
3 – cylinder;
b – glassy cylinder with skin shred in solution:
1 – weighing bottle;
2 – threads;
3 – rubber for cylinder anchoring;
4 – solutions in cylinder and weighing bottle,
5 – cylinder,
6 – skin).

Fig. 1. Scheme of the experiment on fish skin permeability study.

Shreds of fish skin were fixed on cylinders directed inward by epithelium as well as outward by epithelium. Rubber tubes were put on the glassy cylinders. Weighing bottles were filled by 0.65% solution of NaCl, 10 *mL*. In glassy cylinders with skin shreds the solution of 0.65% NaCl was added to be sure in their tightness. On the basis of the solution of 0.65% NaCl the solution of MB was prepared.

Then the solution of NaCl was poured out and the dye solution was poured in by 2 *mL*. The prepared glassy cylinders with skin shreds and dye solution were loaded in the weighing bottle with the solution of 0.65% NaCl. One should be attentive, because levels of the solutions of NaCl in weighing bottle and the dye solution in glassy cylinder should coincide. Skin shreds in weighing bottles were incubated for 3 weeks at 22°C. For this aim, we place these shreds in previously prepared thermostat. Along the incubation time the experiments were carried out at wavelength 665 *nm* [12].

As a control solution the solution of 0.01% MB was used [13]. Permeability of skin shreds previously placed in the following solutions during 30 *min*: in distilled water, in ethanol 70% and in solution of 0.125 *M* KCl was measured.

MB concentration was calculated by comparison method [14]. Comparing the values of optic densities of standard A_{st} and studying A_x solutions, the average value of unknown concentration C_x of determining compound was found.

According to the main rule of light absorption:

$$A_x = \varepsilon_\lambda \cdot C_x \cdot l\,, \qquad A_{st} = \varepsilon_\lambda \cdot C_{st} \cdot l\,.$$

Dividing the first expression to the second one and taking into account that the optic density is measured in the same conditions, i.e., at the same wavelength and cuvette, we will receive:

$$\frac{A_x}{A_{st}} = \frac{\varepsilon_\lambda \cdot C_x \cdot l}{\varepsilon_\lambda \cdot C_{st} \cdot l}\,, \qquad C_x = \frac{C_{st} \cdot A_x}{A_{st}}\,.$$

In experiments the error does not exceed 5–10%.

Results and Discussion. Experiments have shown that the skin permeability changes depending on the solution, where the skin shreds of Sevan trout were kept for 30 *min*: the shreds were kept in distilled water, in 70% ethanol and in solution of 0.125 *M* KCl. The shreds of Sevan trout that were not kept in solutions served as a control sample.

The permeability of Sevan trout skin was judged by determination of MB concentration in physiological solution – fixing the skin shreds on cylinders with direction inward and outward of epithelium.

Studies showed (Fig. 2) that at the keeping of the shreds in the solution of 0.125 *M* KCl, with epithelium direction inward the concentration of MB (skin permeability) decreases by 10.8 times as compared to the control, while in the case of epithelium direction outward the concentration of MB (skin permeability) decreases by 11.6 times, as compared to the control. At the keeping of the shreds in distilled water with epithelium direction inward the concentration of MB decreases by 9.3 times as compared to the control, but at the epithelium direction outward the concentration of MB decreases by 6.0 times as compared to the control. At the keeping the skin shreds in 70% ethanol with epithelium direction inward the concentration of MB decreases by 16.4 times, as compared to the control; for epithelium direction outward the concentration of MB decreases by 14.6 times as compared to the control.

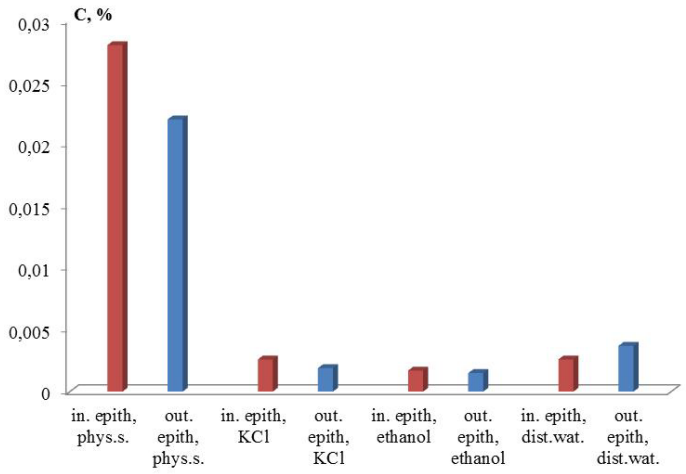


Fig. 2. MB concentration in physiological solution, while keeping skin shreds in distilled water, 70% ethanol and solution of 0.125 *M* KCl.

Analyzing the data one can conclude that the separated skin of Sevan trout is permeable and in the case of the direction of the epithelium inward and outward the dye permeability through the Sevan trout skin can be arranged as follows:

$$\text{Control} \rightarrow \text{distilled water} \rightarrow 0.125\ M\ \text{KCl} \rightarrow \text{ethanol}.$$

It was established that the skin shreds of Sevan trout, kept in various solutions, possess different degree of permeability.

Conclusion. Generalizing the results one can assume that Sevan trout skin is permeable for MB, but this permeability changes depending on both the solution it was kept in and direction of shred fixation. Comparing the abovementioned data one can say that distilled water, solution of 0.125 *M* KCl and ethanol result in structural changes of skin layers of Sevan trout, particularly of epidermis. It should be mentioned that the main channels of penetration of various compounds through the fish skin can be considered intercellular spaces [15] and, most apparently, the aforementioned solutions can lead to changes of intercellular spaces of the skin, which, in turn, results in permeability alteration of Sevan trout skin.

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Մ. Ա. ՓԱՐՍԱԴԱՆՅԱՆ, Մ. Ս. ՄԻՔԱԵԼՅԱՆ

ՁԿՆԵՐԻ ՄԱՇԿԻ ԹԱՓԱՆՑԵԼԻՈՒԹՅԱՆ ՀԵՏԱԶՈՏՈՒԹՅՈՒՆ

Հետազոտվել է Սևանի իշխանի մաշկի թափանցելիությունը 30 *րոպեի* ընթացքում, տարբեր լուծույթներում՝ թորած ջրում, 70%-անոց էթիլ սպիրտի և KCl-ի 0,125 *Մ* լուծույթներում: Թափանցելիությունը որոշվել է մեթիլենային կապույտի օգնությամբ: Ցույց է տրվել, որ մաշկի թափանցելիությունն այս ներկանյութի նկատմամբ փոխվում է՝ կախված նրանից, թե Սևանի իշխանի մաշկի կտորները ինչ լուծույթում են պահվել և որ կողմից են ամրացվել: Նաև ցույց է տրվել, որ թորած ջուրը, 0,125 *Մ* KCl-ի լուծույթը և էթիլ սպիրտը հանգեցնում են Սևանի իշխանի մաշկի շերտերի կառուցվածքային փոփոխություններին, մասնավորապես, էպիդերմիսի փոփոխություններին:

М. А. ПАРСАДАНЯН, М. С. МИКАЕЛЯН

ИССЛЕДОВАНИЕ ПРОНИЦАЕМОСТИ КОЖИ РЫБ

Исследована проницаемость кожи севанской форели в течение 30 *мин* в различных растворах: в дистиллированной воде, 70%-м растворе этилового спирта и в 0,125 *М* растворе хлористого калия. Проницаемость измеряли с помощью метиленового синего. Показано, что проницаемость кожи для этого красителя изменяется в зависимости от вида раствора, в котором были помещены лоскутки кожи севанской форели, и от ее стороны, на которой она была закреплена. Показано также, что и дистиллированная вода, и 0,125 *М* KCl, и спирт приводят к структурным изменениям слоев кожи севанской форели, в частности эпидермиса.