# USE OF ELECTROPHORESIS FOR INTRODUCING CHEMICALS INTO THE DEVELOPING CHICK EMBRYO

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The authors suggest a method of introducing various chemical ions into a hen's egg without disturbing the integrity of the shell by means of the AGN-2 galvanizing apparatus.

Considerable attention is now being paid to methods of controlling embryonic development in hens. Some important criteria characterizing the formation of the embryo are grown and developed.

Vital processes can be stimulated in various ways. The introduction of biologically active substances into the egg is of considerable interest. The injection method has been used for many years by experimenters studying the effects of various substances on development of the chick embryo [1]. However, puncture of the egg frequently leads to death of the embryo, especially if the test substances are injected in the late stages. In addition, even if sterile precautions are observed, pathogenic microorganisms will sometimes enter the egg. The injection method is also accompanied by trauma, which is bound to influence embryogenesis.

Another method sometimes used to introduce substances is immersing the eggs in the corresponding solutions [4]. This method requires a large quantity of the substance and, in addition, water also penetrates through pores in the shell.

The galvanic current is frequently used in medical practice in order to introduce therapeutic substances into the body through the skin. This method is called ionophoresis or electrophoresis.

The objective of the present investigation was to determine whether certain ions can be introduced selectively through the pores of the shell into an egg by electrophoresis and to study how this process is affected by the polarity of the ion.

The current for electrophoresis was generated by an AGN-2 galvanizing apparatus [2]. Rectangular lead plates measuring 20-25 cm were used as the electrodes. They were connected by flexible leads to the source of current. Pads made of hydrophilic material (flannel or gauze), folded into 15-20 layers so that the thickness was not less than 1 cm, were placed beneath the electrodes. The size of the pads was chosen so that they projected 1-2 cm beyond the edges of the electrode.

To obtain uniform distribution of the current and to secure the eggs more firmly, circular holes 35 mm in diameter were cut out of a sheet of plywood measuring $200 \times 300\ mm^2$ and 4 mm in thickness; the distance between the holes was 3 cm. The wet pad was placed beneath the sheet of plywood, and then underneath it,the electrode. A certain number of eggs (15) was placed vertically or horizontally in the holes in the sheet of plywood so that the corresponding part of the egg was in firm contact with the wetpad. The upper surface of the eggs was covered by a second sheet of plywood with holes, after which the pad andelectrode from the other poles were applied. Both pads were first soaked with warm water and wrung out so that they were equally damp. Depending on the polarity of the ions to be introduced, one of the pads was

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soaked with the chemical substance and the electrode connected to the corresponding pole; by the rule that like charges repel, positively charged ions are introduced from the anode and negative ions from the cathode.

When substances are to be introduced by means of electrophoresis, it is recommended that the dose be calculated in coulombs. Uusually 1 C is given per $cm^2$ of pad area (i.e., 1 mA for 16 min 40 sec). The quantity of substance introduced equivalent to 1 C can be calculated. To do this, the atomic weight of the substance (in g) is divided by 96,500 (Faraday's number), and the appropriate correction for valency is introduced [3].

The following tests were carried out to confirm that ions of chemical substances pass through the shell under the influence of the galvanic current.

The pad placed under the electrode was connected to the cathode and was soaked in Lugol's solution, while the other pad was soaked in tap water. The electrodes were applied alternately. The egg was opened after 15-20 min, and by means of the starch test, a blue color was found on the inner surface of the shell on the side of the active electrode, indicating that iodine had penetrated through the shell under the action of the current. Parallel with the experiment, similar pads were applied to the egg in a control test, but not connected to the source of current. In this case the starch test gave a negative result (no blue color).

Other tests were carried out to study the introduction of radioactive iodine by means of the galvanic current. The pad under the electrode, connected to the cathode, was soaked in physiological saline containing 1 $\mu$Ci radioactive iodine, while the other pad was soaked in plain physiological saline. The current was passed for 15-20 min. The electrode pads of the control eggs were also soaked in physiological saline containing radioactive iodine, but they were not connected to the apparatus. After the end of the test both the experimental and the control eggs were carefully rinsed under a strong jet of water (15 min) in order to remove traces of radioactive iodine, after which they were carefully wiped dry and applied to the $\gamma$-counter of a type B-2 instrument. The experimental eggs gave 140 pulses/min, whereas in the control tests the number of pulses did not exceed the background level for the instrument.

These experiments demonstrate that ions pass through the shell under the action of a galvanic current.

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