FORMATION OF AN ION DEPOT IN THE SKIN BY ELECTROPHORESIS

N. A. Barsukov

Yakutsk State University (Rector – Docent I. G. Popov)
(Presented by Active Member AMN SSSR V. V. Parin)
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It is generally accepted that therapeutic substances introduced by means of a direct current are excreted more slowly than when administered in other ways [1, 3, 4, 5]. We have studied the concentrations of drugs introduced into the skin and underlying tissues, and the time during which they remain in the skin during electrophoresis. The drugs tested were sulfonamides (sulfacetamide, sulfanilamide, and sulfathiazole) and antibiotics (penicillin, streptomycin, chlortetracycline, oxytetracycline, and tetracycline). The investigations were conducted on dogs.

EXPERIMENTAL METHOD

At various intervals after the completion of a session of electrophoresis, pieces of skin and muscles which had been beneath the active electrode were excised and the concentration of the drug was determined. The blood was investigated at the same times.

The sulfonamides were estimated quantitatively in the tissues by a chemical method using a photoelectric colorimeter [2]. Antibiotics were estimated quantitatively by a microbiological method, namely by diffusion in agar using different test organisms: for penicillin, Staphylococcus aureus, strain 209P; for streptomycin, B. mycoides; and for the tetracyclines, a spore-bearing L_2 strain.

At the site of application of the active electrode the hair was shaved, the skin washed with soap and water, dried with a towel, and irrigated with alcohol. As hydrophilic covers we used sterilized paper towels measuring 10 × 12 cm. The sulfonamide solutions for soaking the hydrophilic covers were made up in dilute hydrochloric acid: a weighed amount of crystalline powder of the ordinary (insoluble) forms of the sulfonamides calculated to produce the required concentration (usually 1-2%) was added to distilled water at a temperature of 20-30°; concentrated hydrochloric acid was then added gradually from a pipet with agitation until the preparation had completely dissolved. Solutions of penicillin, streptomycin sulfate, and crystalline chlortetracycline hydrochloride (the latter in powder form) were prepared in distilled water sterilized by boiling. The solutions of oxytetracycline and tetracycline hydrochlorides, issued in powder form for intramuscular injection, were prepared in a 0.1 N solution of hydrochloric acid, for if dissolved in distilled water they form a precipitate after 30-60 min and are unsuitable for electrophoresis.

Ten thicknesses of sterile paper towel were soaked in the solutions of the drugs. Over these were placed 16 to 20 thicknesses of paper towel soaked in 5% glucose solution. The coverings soaked in glucose were smaller than the other towels by about 0.5 cm along each edge. A lead plate was then applied and connected to the anode (during penicillin electrophoresis the active electrode was connected to the cathode). For control purposes, the concentration of the preparation in the tissues after administration by the ordinary methods was also determined.

EXPERIMENTAL RESULTS

Orally administered sulfonamides disappeared from the blood after different times, depending on the type of preparation. For instance, 2 h after administration of a single dose (80-100 mg/kg), sulfanilamide was present in the serum in a concentration of 34-44 μg/ml; sulfacetamide, 32-45 μg/ml; and sulfathiazole, 31-38 μg/ml, i.e., in approximately the same concentration. After 24 h, however, the first two were found in the blood only as traces, while the concentration of sulfathiazole was 10-12 μg/ml.* In these cases, the concentrations of sulfanilamide and sulfacetamide in the skin and muscles after 2 h were 24-32 μg/g, and of sulfathiazole, 14-27 μg/g; after 24 h the first two were present only in traces, while the concentration of sulfathiazole was 5-14 μg/g.

*The figures give the concentration of free sulfonamides only. No difference was found between the relative concentrations of total and free forms of sulfonamides in the tissues when given by mouth and by electrophoresis.

As a rule during electrophoresis no sulfonamide preparations were found in the blood, but higher concentrations were found in the tissues. In particular, when hydrophilic covers soaked in 2% sulfanilamide solution, made up in dilute hydrochloric acid, were used, and the current density was 0.3 mA/cm^2 and the duration of the session 30 min, the concentration in the skin 2 h after the end of the session was 76-114 μg/g; after 24 h, 10-24 μg/g; and after 48 h, traces were present. In these cases the muscles under the active electrode contained 22-38 μg/g sulfanilamide after 2 h, and 4-12 μg/g after 24 h. With the same values of current density and length of sessions, after electrophoresis of 1% sulfathiazole solution in dilute hydrochloric acid, 70-86 μg/g was found in the skin after 2 h, 26-40 μg/g after 24 h, 12-18 μg/g after 48 h, and traces after 72 h.

In the same conditions, the concentration of sulfathiazole in the muscles beneath the active electrode was 16 to 26 μg/g after 2 h, 8-13 μg/g after 24 h, and traces were present after 48 h. The concentration of sulfacetamide in the tissues after electrophoresis was very similar to that of sulfanilamide.

These experimental results showed that the differences in the rate of elimination of the various sulfonamides from the tissues when given by mouth persisted when they were given by electrophoresis: sulfathiazole was eliminated from the tissues more slowly than sulfanilamide or sulfacetamide when given by either method. It should be noted that, if considered individually, these preparations remained longer in the tissues the higher the initial concentration of the substance created in these tissues (irrespective of the method of administration). If, by suitable conditions of electrophoresis, the same concentration of a sulfonamide preparation was created in the skin or muscles beneath the active electrode as after oral administration, its disappearance from the tissues took place at the same rate after both methods of administration. Since the administration of sulfonamides by electrophoresis may produce much higher concentrations than when given by mouth (even in doses which, in our experience, exceeded the optimal therapeutic dose), they remain longer in the tissues after electrophoresis.

The same pattern was observed in the experiments for the comparative study of the different methods of administration of antibiotics. For instance, the concentration of streptomycin in the blood serum 1 hour after intramuscular injection of doses of 10,000-20,000 units/kg was 3-10 units/ml, and in the tissues only 1-3.5 units/g, or else as traces. After 6 h, only traces were present in the blood, and none was found in the tissues (skin and muscles). After administration of streptomycin by electrophoresis, using solutions of 5000 units/ml, not more than 3.5 units/g was found in the skin 1 h after the end of the session, only traces were found after 3 h, and none after 6 h. If a solution of 20,000-30,000 units/ml of streptomycin was used, the concentration in the skin after 1 h was 20-35 units/g, after 6 h, 5-12 units/g, and after 24 h, none was found in every case. In the same series the concentration of streptomycin in the muscles beneath the active electrode was 3-7 units/g after 1 h, and after 6 h only traces or none could be found. The results of the investigation of the tetracyclines (chlortetracycline, oxytetracycline, and tetracycline) were similar. After oral administration of these drugs in a dose of 10,000-20,000 units/kg, their highest concentration was found after 3 h: 4.5-11 units/ml in the blood and 1-2 units/g in the skin and muscles. After 6 h the concentration in the blood was 3-6 units/ml, and only traces were present in the tissues. After electrophoresis of the tetracyclines, using a solution of 5000 units/ml, a current density of 0.3 mA/cm^2, and sessions lasting 30-60 min, 1 h after the end of the session, the concentration in the skin was 2-3 units/g, while none was found in the muscles or in the blood; after 6 h very small amounts or traces only were found in the skin. Using the same current density and duration of sessions, but solutions with a strength of 20,000-30,000 units/ml, the tetracycline concentration in the skin after 1 h remained at 20-30 units/g, and it remained at the level of the therapeutic concentration (not less than 2-3 units/g) for 24 h or more. In these cases the tetracyclines were found in the muscles beneath the active electrode after 1 h in a concentration of 4-6.5 units/g, and a thereapeutic concentration was maintained for 12 h.

The comparison of the rates of elimination of antibiotics from the body tissues after administration by the usual methods and by electrophoresis thus showed that these drugs, too, remained longer in the tissues the higher their initial concentration in these tissues, irrespective of the mode of administration. The difference in the rate of elimination of the various antibiotics is explained by their different properties. However, the rate of elimination of any given antibiotic is the same whether given by ordinary methods (for a resorptive action) or by electrophoresis. Consequently, our results gave no grounds in support of the view that substances administered by electrophoresis remain longer in the body than if administered by other methods.

During the study of the distribution and elimination of penicillin from the body when given by different routes, we observed a phenomenon resembling, at first glance, the formation of a "skin depot." After intramuscular injections of therapeutic doses (2000-5000 units/kg) at the end of 1 h, penicillin was found both in the blood (1-3 units per ml) and in the skin and muscles (0.7-2.4 units/g). After 3-4 h it disappeared both from the blood and from the

tissues. When it was given by electrophoresis, using solutions containing 20,000-30,000 units/ml, a current density of 0.3 mA/cm^2, and sessions lasting 30-60 min, penicillin was found in the tissues beneath the active electrode after 1 h: 15-23 units/g in the skin and 1-2 units/g in the muscles. Whereas, however, in these experiments penicillin did not remain longer than 2-4 h in the muscles after the end of the session, in the skin it remained for 48-72 h or longer. Yet no difference was found in the penicillin concentration in the muscles depending on the method of administration. It is therefore possible that penicillin was retained in the inert layers of the skin, namely in the stratum corneum.

In order to shed light on this phenomenon, we carried out experiments on dogs. The penicillin concentration after electrophoresis was determined separately in the upper and lower layers of the skin at different intervals after the end of the session. In the lower layers of the skin the antibiotic was found for only 3-6 h, while in the upper layers it persisted for 72 h or longer.

We consider from the foregoing facts that the formation of a "skin depot" during electrophoresis of penicillin was due to the retention of the antibiotic in the stratum corneum of the skin, in which metabolism is absent. Penicillin injected intramuscularly does not enter the stratum corneum but, being present in tissues possessing a blood supply, including the lower layers of the skin, it is rapidly eliminated from them. Penicillin introduced into the lower layers of the skin as a result of electrophoresis suffers the same fate. That fraction of the penicillin which enters the stratum corneum during electrophoresis is then gradually broken down and removed along with the desquamated keratin scales.

We have not yet explained the fact that penicillin persists for a long time in the stratum corneum after electrophoresis, while the other antibiotics and sulfonamides do not. From supplementary investigations we consider, however, that the prolonged retention of penicillin is not dependent on the influence of the direct current. For instance, studies of the concentration of penicillin in the tissues beneath simple dressings soaked in penicillin solution, when no current passes through them, showed that penicillin remained for a long time in the stratum corneum of the skin even under these simple dressings.

SUMMARY

Experiments were staged on dogs. As established, sulfanilamide preparations and antibiotics (penicillin, streptomycine, biomycine, teramycine, and tetramycine), administered by electrophoresis method, were not simultaneously eliminated from different tissues.

High concentration of the preparations studied persisted in the skin under an active electrode for longer periods of time, forming a "skin ion depot."

LITERATURE CITED

1. I. A. Abrikosov, Collection: Physiotherapy [in Russian] (Moscow, 1955), p. 33.
2. L. I. Grebenik, Collection: Methods of Experimental Chemotherapy [in Russian] (Moscow, 1959), p. 431.
3. Ya. S. Perlin, Galvano- and Galvano-Ionic Therapy [in Russian] (Moscow, 1958).
4. N. B. Poznanskaya, Byull. éksper. biol., 17, 3, 9 (1944).
5. L. R. Rubin, Physical Methods of Investigation and Treatment in Stomatology [in Russian] (Moscow, 1955), p. 126.
