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EFFECT OF PLASMA FLOW ON REGENERATION OF SKIN WOUNDS AND ON REACTIVITY OF THE BODY

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The introduction of the "plasma scalpel," whose action is based on the use of the flow of energy of ionized gases, into surgical practice for dividing biological tissues and treating wounds [1], necessitates a deeper study of this subject.

The aim of this investigation was to study the character of action of the plasma flow on biological structures, on individual systems, and on the body as a whole.

EXPERIMENTAL METHOD

The effect of the flow of a plasma jet on wound healing was studied in three series of experiments (six rabbits in each series). In series I and II, under aseptic conditions, wounds were inflicted on the animals' back by removal of flaps of skin and fascia measuring 4 × 4 cm. In series III, only the hair was removed from the animals in the corresponding region. Daily, starting from the 1st day, the wound region in the animals of series II and the intact skin of rabbits in series III of the experiments were "irradiated" with a plasma flow. The temperature of the plasma flow at the site of contact with the animal's tissues was 37-38°C. The wounds healed by the open method. Assessment of the character of wound healing was based on times of regeneration, planimetry of the wound, and cytology of the wound exudate. Reactivity of the body was judged on the basis of the study of peripheral blood and bone marrow morphology, determination of proteins, glucose, and lipids in the blood plasma, and investigation of lipid peroxidation (LPO) by determining accumulation of malonic dialdehyde (MDA) in the blood.

EXPERIMENTAL RESULTS

Immediately after wounding of the rabbit a vasomotor response developed at the site of the defect and exudation of the wound took place. After the end of 3 days, in the experiments of series I a marked inflammatory reaction developed, accompanied by hyperemia of the edges and floor of the wound defect, with features of hydration. From the 3rd through the 7th day after wounding, healing took place in all the animals in this series of experiments, beneath a scab. Later, the scab on one animal was rejected and the wound granulated openly until epithelization was complete. Granulation processes and epithelization took place in the other animals beneath the scab.

In the experiments of series II, on the 2nd day after application of the plasma flow, features of exudation in the wound region were mild in degree, and later a delicate, thin scab formed. The wounds contracted well and healed with an accurate scar, almost indistinguishable in appearance from the surrounding skin. The area of the wound in this series of experiments decreased by 47% until the 9th day, whereas in the animals of series I it decreased by only 22%. The mean rate of regeneration in series I, without treatment by plasma flow, was 2.9% in 24 h, compared with 6.1% after treatment with plasma.

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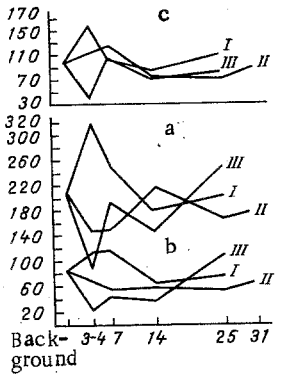

Fig. 1

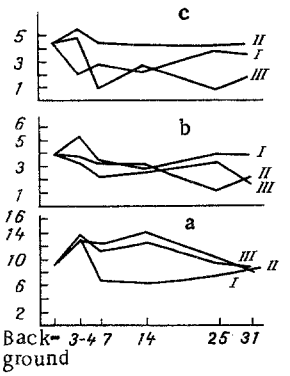

Fig. 2

Fig. 1. Time course of number of nucleated bone marrow cells during treatment of skin wounds in rabbits with plasma flow. Abscissa, time of observation (in days); ordinate, number of nucleated cells ($\times 10^9$/liter). a) Myelokaryocytes; b) erythroid bone marrow cells; c) neutrophils. I) Wounded, not irradiated groups; II) wounded and irradiated groups; III) irradiated, not wounded group.

Fig. 2. Time course of changes in glucose and MDA concentration and leukocyte count during treatment of skin wounds in rabbits by plasma flow. a) Leukocyte count ($\times 10^9$/liter); b) MDA concentration (in nmole/ml); c) glucose concentration (in mmoles/liter). Remainder of legend as to Fig. 1.

The opposite picture was observed after 9 days. Wound healing began to be activated in the animals in the experiments of series I and the rate of contraction of the wound surface in the experiments of series I by the 31st day was 59%, compared with 44% in the experiments of series II, although the average rates of healing did not differ significantly, namely 5.7 and 6.8%, respectively.

Under the influence of the plasma flow on the skin of intact animals in the experiments of series III no marked skin reaction was observed.

Cytological study of the wound exudate in the experiments of series I revealed considerable bacterial contamination, with the appearance of large quantities of debris. Most of the cells of the wound exudate were neutrophils, with a few mononuclears.

Some degree of activation of the monocytic-macrophagel reaction began only after the first 10 days.

In the group of animals of series II, under the influence of the plasma flow many monocytes were released into the wound during the first 10 days, and marked phagocytosis took place. An increase in the number of fibroblasts was observed in the wound.

During the study of the peripheral blood of the animals in the experiments of series III with no wound, leukocytosis was observed after application of the plasma flow, and during adaptation of the animals it gradually diminished, and returned close to the initial values. Fluctuations in the leukocyte formula were within normal physiological limits. The red blood picture differed from the original in the presence of some degree of anemia with a tendency toward hyperchromia, returning to normal only at the end of the course of plasma treatment. In the bone marrow of the animals in this series of experiments myelokaryocytopenia was present, mainly due to a decrease in the number of erythroid cells, which was particularly marked during the first 3 days (from 209,000 to 89,000 in 1 μl of puncture material). After the end of the sessions of irradiation with plasma flow the number of nucleated cells was restored, and after 2-3 weeks it actually exceeded the initial value (Fig. 1a).

In the animals in the experiments of series I with a skin wound a marked leukocytic response was observed on the 3rd day after the operation (Fig. 2a). The leukocytosis was due mainly to polymorphs and monocytes.

Under the influence of the plasma flow on wounds in the experiments of series II, after transient leukocytosis the total leukocyte count returned to normal but changes still re-

mained in the leukocyte formula: relative lymphocytopenia, polymorphonuclear neutrophilia, and monocytosis.

A synchronized study of the bone marrow cells in rabbits in the experiments of series I showed that peripheral blood leukocytosis coincided with periods of myelokaryocytosis of the bone narrow, and it came about as a result of an increase in the number of all cells of the myelogram (Fig. 1, a-c).

After treatment of the wound surface with the plasma flow the number of cells of the erythroid series fell somewhat (Fig. 1b), but changes in the neutrophils of the bone marrow, just as in the peripheral blood, were characterized by a reaction to trauma. The phase of neutrophilia both in the bone marrow and in the peripheral blood coincided with a phase of inflammatory reaction of the wound. A biochemical investigation showed that in response to trauma the animals in the epxeriments of series I developed hypoglycemia, which returned to normal on the 25th day after wounding (Fig. 2c). As a result of exposure to the plasma flow animals of series II developed hyperglycemia, which changed into normoglycemia. Unlike the experiments of series I, in which hypoproteinemia alternated with hyperproteinemia, in series II no significant changes took place in the plasma protein concentration, but after 11 sessions of irradiation a tendency toward hyperproteinemia developed.

The plasma lipid concentrations showed no significant changes and no difference between them was found in any of the groups of experiments.

The MDA concentration, reflecting the state of LPO (Fig. 2b), rose in the animals of series I until the 3rd day after the operation, after which it fell toward the end of the first 10 days, and rose again on the 25th day, which coincided with infection of the wound. Under the influence of the plasma flow reduced activity of lipid peroxidation was observed in the animals of series II (Fig. 2b).

After infliction of skin wounds a neutrophilic leukocytosis thus appears, due to participation of the bone marrow in this reaction; breakdown of proteins, glycogen, and lipids is increased, and their oxidation disturbed. This situation is one of the nonspecific complex of changes, known as the "stress reaction." These parameters later returned to normal.

Exposure to the plasma flow leads to active growth of granulations during the first 10 days of irradiation, quiescence of the perifocal inflammatory process, the escape of many monocytic cells in a state of phagocytic activity into the wound, the appearance of islets of epithelial cells, and some stimulation of hematopoietic function in the body. Metabolic activity was enhanced, against the background of depression of free-radical oxidation. It was concluded that these results justify attempts to introduce the "plasma method" of treatment into clinical practice for the treatment of indolent wounds.

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