EFFECT OF CHONDROITIN SULFATE PREPARATIONS ON WOUND HEALING AND STRENGTH OF THE SURGICAL SCAR

M. A. Fialkova, T. Yu. Smirnova, G. I. Ivanova, R. K. Aboyants, V. F. Golubeva, I. Yu. Tsarevskaya, S. Yu. Kuznetsova, V. E. Belai, and G. N. Khlyabich

UDC 616.5-003.93-02:615.361.018.3

Key words: chondroitin sulfate; chonsuride; rumalon.

Sulfated glycosaminoglycsns are known to stimulate regeneration of the skin when injured [3, 5]. This property also is shared by preparations for injection (rumalon, mucartrin, etc.) and by substances for external application (chonsuride) [2]. In the Soviet Union only one preparation based on chondroitin sulfate (ChS) is marketed, namely chonsuride, which is intended for local application in the treatment of indolent wounds, trophic ulcers, etc. [1]. ChS (the sodium salt) is a biologically active animal tissue glycosaminoglycan, and is obtained by extraction from the bovine trachea followed by purification to remove contaminating proteins and nucleic acids. The preparation is readily soluble in water and in isotonic sodium chloride solution, and it contains 95% of total glycosaminoglycans, 3% nitrogen and up to 3% of sulfur. According to the results of analytical ultracentrifugation the mean molecular mass is 11,000 D. The therapeutic form is a 10% aqueous solution of ChS. This paper describes an experimental study of the ability of this preparation to stimulate regeneration of the skin when injured.

EXPERIMENTAL METHOD

There were two series of experiments. In series I, on 50 female Wistar rats weighing 200-250 g, the effect of ChS on the strength of the surgical scar was studied. A linear incision 2.5 cm long was made over the middle of the spine, under hexobarbital anesthesia. The animals were divided after the operation into four groups. Rats of group 1 received ChS intramuscularly in a dose of 0.6 ml once a day for 5 days, whereas the animals of group 2 received 0.3 ml twice a day for 2 weeks. Animals of group 3 received rumalon on the same schedule. The control rats (group 4) were given injections of 0.9% sodium chloride solution. The animals were killed 2 weeks after trauma. The skin at the site of the operation was excised, cut into strips 5-10 mm wide, and the strength of the scar tested by stretching on a model 1122 "Instron" tensile testing machine, with a stretching speed of 20 mm/min. The quality of the scar was characterized by the value of the load at which the tested area of skin ruptured (the relative strength, in N/mm^2, and the relative elongation causing rupture, in percent). In the experiments of series II on 50 female albino rats weighing 180-200 g the effect of external application of ChS was studied on the rate of healing of full-thickness skin wounds [4]. A concentric incision was made under hexobarbital anesthesia and a Teflon ring introduced into the resulting defect, and securely fixed to the surrounding tissue. The area of the wound was 300 mm^2. The test substances were applied to the wound surface inside the ring (30 mg of ChS or chonsuride per course of treatment). Rats of the 1st group received one application of the preparation on the 1st day and the ring was removed on the 5th day of the experiment. Animals of group 2 had chonsuride applied on the 1st and 2nd days, and the Teflon ring was removed on the 3rd day after the operation. The rate of healing of the wounds was judged by the area of the wound surface measured on the 5th, 7th, 9th, 13th, 16th, and 18th days, and also by noting the times of complete healing.

Control Laboratory for Hormones and Organ Preparations. All-Union Research Institute of Blood Substitute and Hormonal Preparation Technology. Laboratory for the Study and Use of Collagens in Medicine. I. M. Sechenov First Moscow Medical Institute. A. N. Kosygin Moscow Textile Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR V. V. Kovanov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 108, No. 9, pp. 350-351, September, 1989. Original article submitted January 20, 1989.

0007-4888/89/0009-1327$12.50

TABLE 1. Effect of ChS on Strength of Skin Scar

Preparation	Daily dose, ml	Total dose, ml	Duration of application, days	Relative strength, N/mm^2	Relative rupture lengthening, %
				$M \pm m$	
ChS	0,6	3,0	5	646,8±57,8	61±5,2
ChS	0,6	8,4	14	774±57,8*	83±6,5**
Rumalon	0,6	8,4	14	705,6±52,9	73±5,8
Control	0,6	8,4	14	607,7±39,2	61±2,5

Legend. $*p < 0.05$, $**p < 0.01$ compared with control.

TABLE 2. Time Course of Healing of Skin Defect

Periods of observation, days	Area of wound, mm^2 (M ± m)					
	single application of preparations			two applications of preparations		
	0.9% sodium chloride solution	chonsuride	ChS	0.9% sodium chloride solutions	chonsuride	ChS
5	300	300	300	212±6,3	188±5,7	183±7,0
7	230±9	184±10	158±8	114±12,6	134±10,1	67±2,0
9	128±6	90±10	96±12	72±8,4	58±4,2	37±1,8
13	61±2	38±2	34±2	64±6,3	42±4,0	22±2,4
16	43±2	10±1	10±1	38±2,4	21±1,1	
18				24±1,3		
Periods of complete wound healing	22,8±0,2	20±0,18	20±0,2	21,1±0,3	18,4±0,21	18,1±0,3

Legend. $*p < 0.05$ compared with control, $**p < 0.05$ compared with treatment by chonsuride.

EXPERIMENTAL RESULTS

Injection of a 10% solution of ChS during the first 5 days after wounding had no effect on the strength of the skin scar (Table 1). A course of 2 weeks of injections increased the strength of the scar by 27% and the relative rupture lengthening by 36% compared with the control. Rumalon in the above dose likewise affected the strength of the surgical scar, but to a lesser degree. For instance, the strength of the scar was increased by 16% and the rupture lengthening by 20% compared with the control (Table 1).

After a single local application of ChS to the wound surface on the 2nd day after the operation a considerable decrease was observed in the edema of the tissue surrounding the wound compared with the control. Hyperemia and exudation in both experimental groups also were much weaker than in the control rats. By the 9th day, separation of the primary scab was observed in the majority of experimental animals, indicating more rapid maturation of granulation tissue in the wounds of these rats. This process in the control rats was observed on the 11th-12th day.

The results of the planimetric investigation showed that the area of the wounds in rats receiving ChS and chonsuride was significantly less than in the control animals (Table 2). The action of ChS, stimulating regeneration, did not differ significantly from the effect of chonsuride. In both cases complete recovery took place on the 20th day (on the 23rd day in the control).

After two applications of ChS to the wound surface, the area decreased significantly faster than after treatment with chonsuride. For instance, by the 8th day the area of the wound in rats receiving ChS was only half of that in the control animals treated with chonsuride. This difference still persisted at subsequent times, although complete healing occurred at about the same time (Table 2).

Thus the sample of ChS tested stimulated regeneration of skin wounds, whether administered parenterally or locally. Its therapeutic effect in both cases was much stronger than that of rumalon and chonsuride.

LITERATURE CITED

1. A. N. Ivankin, S. E. Vasyukov, and V. P. Panov, Khim.-Farm. Zh., No. 3, 192 (1985).
2. B. S. Kasavina, A. I. Rikhter, G. D. Zenkevich, and L. A. Arenberg, Éksp. Khir., No. 5, 10 (1961).
3. M. Matev, D. Pekhlivanov, I. Sheitanov, and L. Neinovskaya, Med.-Biol. Inform. (Sofia), No. 4, 12 (1977).
4. L. I. Slutskii, Biochemistry of Normal and Pathologically Changed Tissue [in Russian], Leningrad (1969).
5. Takehiko Kawano, "Low molecular chondroitin sulfate and method for manufacturing the same," U.S. Patent No. 3405120 (1966).

EFFECT OF CARNOSINE ON HEALING OF LUNG WOUNDS

M. I. Perel'man, Z. Kh. Kornilova, V. S. Paukova, A. K. Boikov, A. A. Priimak, T. V. Bulargina, S. E. Severin, and G. N. Tikhonova

UDC 616.24-001.44-085.324:547.784.2]-036.8-07:616.24-003.9

Key words: lungs; regeneration; stimulation; carnosine.

Carnosine dipeptide (β-alanine, α-histidine) was discovered in 1900 in the composition of "Liebig's extract," obtained from bovine muscle [7]. This preparation is known to accelerate the healing of surgical wounds, bedsores, and erosions of the cervix uteri, and to stimulate adrenocortical function [2, 9, 10]. For many years the biological role of carnosine was unexplained, but then it was found to have the property of increasing the contractility of fatigued muscles [1, 3, 4]. Severin and Yu-Shu-Yui discovered its membranotropic effect [5]. The use of carnosine was shown to prolong considerably the keeping time of mitochondria isolated from the pectoral muscles of pigeons, while maintaining coupling between respiration and phosphorylation. The beneficial effect of carnosine on processes of Ca^{++} transport into vesicles of the sarcoplasmic reticulum and of Na^+ and K^+ transport through the plasma membrane, dependent on ATP hydrolysis. These data are evidence of the beneficial action of carnosine on the structure of membranes which remain capable of effecting active transport of H^+, Ca^{++}, Na^+, and K^+ ions without leakage [6]. The study of the role of the components of carnosine has shown that β-alanine activates nucleic acid and collagen synthesis, and α-histidine is a reserve for histamine synthesis [8, 10]. The antioxidant properties of carnosine and its ability to regulate the development of inflammatory and immune reactions open up new prospects for its therapeutic use in pulmonology.

The aim of this investigation was to study the effect of carnosine on the healing of experimental lung wounds.

EXPERIMENTAL METHOD

Experiments were carried out on 90 male guinea pigs weighing 280-300 g. A penetrating incised wound of the lung served as the experimental model. Under local anesthesia with 0.5% procaine solution a linear skin incision 1.5 cm long was made on the right side, posteriorly, along the sixth intercostal space, and this was followed by division of the subcutaneous cellular tissue, the spinal muscles, and the intercostal muscles. A penetrating incised wound 8-10 mm deep was then inflicted with a special scalpel. To select the optimal dose of carnosine six series of experiments were carried out in which 10, 15, 20, 25, 30, or 45 mg of the preparation was introduced into the lung wound. Each dose was dissolved in 1 ml of physiological saline, and 0.6 ml of the solution was introduced into the lung wound during the operation and 0.4 ml was injected into muscles and skin at three or four points. Physiological saline was injected in the control. The macroscopic picture of the zone of injury to the lung, muscles, and skin was assessed 3 and 7 days after the operation. The absence of a lung tissue defect and

I. M. Sechenov First Moscow Medical Institute. Moscow Research Institute of Traumatology, Ministry of Health of the RSFSR. M. V. Lomonosov Moscow State University. Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 108, No. 9, pp. 352-356, September, 1989. Original article submitted October 15, 1988.