

PANCREATOPROTECTION OF MALATE-CONTAINING INFUSION SOLUTIONS IN EXPERIMENTAL HEMORRHAGIC SHOCK

UDC 616.001.6–005.1–001.36:616.37–002
Received 2.05.012



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The aim of the investigation was to study in experiment the effect of Sterofundin isotonic on the development of post-hemorrhagic damage of the pancreas.

Materials and Methods. The experiments were carried out on 36 male Wistar rats weighing 230–250 g. Hemorrhagic shock was stimulated by acute massive hemorrhage to the extent of 2.5 mL/100 g at rate of 2 mL/min. The blood loss an hour later was followed by the replacement of hypovolemia within 60 min by 200% of the lost volume: in control group — by Ringer's solution, in test group — by Sterofundin isotonic. Then there was performed reinfusion in the volume of 70% of blood loss. On day 1 and 3 after hemorrhagic shock we assessed the laboratory findings and morphological changes of the pancreas.

Results. There was found pancreatoprotective action of isotonic Sterofundin, a malate-containing blood substitute in infusion therapy of experimental hemorrhagic shock.

Key words: hemorrhagic shock; malate; Sterofundin isotonic; pancreas.

Since constantly high level of mortality associated with the development of posthemorrhagic polyorgan failure the active search of critical care measures contributing the improvement of adaptiveness of inner organs to ischemia and following reperfusion [1–4]. The pancreas is one of target organs for this injury mechanism as it is always in severe shock being off from the circulation due to its centralization. The subsequent development of acute pancreatitis, especially its destructive forms, significantly increases the risk of death [5]. It determines the development of new directions in therapy [6]. In the arsenal of modern blood replacement in recent years, the products containing substrate antihypoxants appeared, but their effect on hemorrhagic damage to the pancreas is unstudied. Fumarate is added as a substrate antihypoxants in blood substitutes (Mafusol), succinate (Reamberin and Remaxol) and malate (Sterofundin isotonic and Sterofundin D-5).

Sterofundin isotonic is norme osmolar balanced polyion solution that can be safely administered by bolus in critical conditions involving hypovolemia and it determined the selection of the study drug.

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the effect of Sterofundin isotonic on the development of post-hemorrhagic damage of the pancreas.

Materials and Methods. Experiments were conducted on 36 male rats of Wistar line, weighing 230–250 g. The research was performed according to the ethical principles established by the European Convention for the protection of vertebrata used for experimental and other scientific purposes (adopted in Strasbourg, Mar, 18, 1986, and confirmed in Strasbourg Jun, 15, 2006). Hemorrhagic shock was simulated under light Nembutal (25 mg/kg) anesthesia by acute massive hemorrhage (AMH) of 2.5 ml/100 g at a rate of 2 ml/min (30% of blood volume) from the tail artery where catheter was installed for the direct measurement of arterial pressure (AP). After the bloodletting AP average was in the range of 40–50 mm Hg. Without any pharmacological support.

1 h after the AMH there was the replenishment of hypovolemia with the selected agent in the volume of 200% of blood loss: in the control series (n=18) — with Ringer's solution, in the experimental series (n=18) — Sterofundin isotonic (B/Braun, Germany). The time of drug injection was 60 min. Within the following 60 minutes the blood

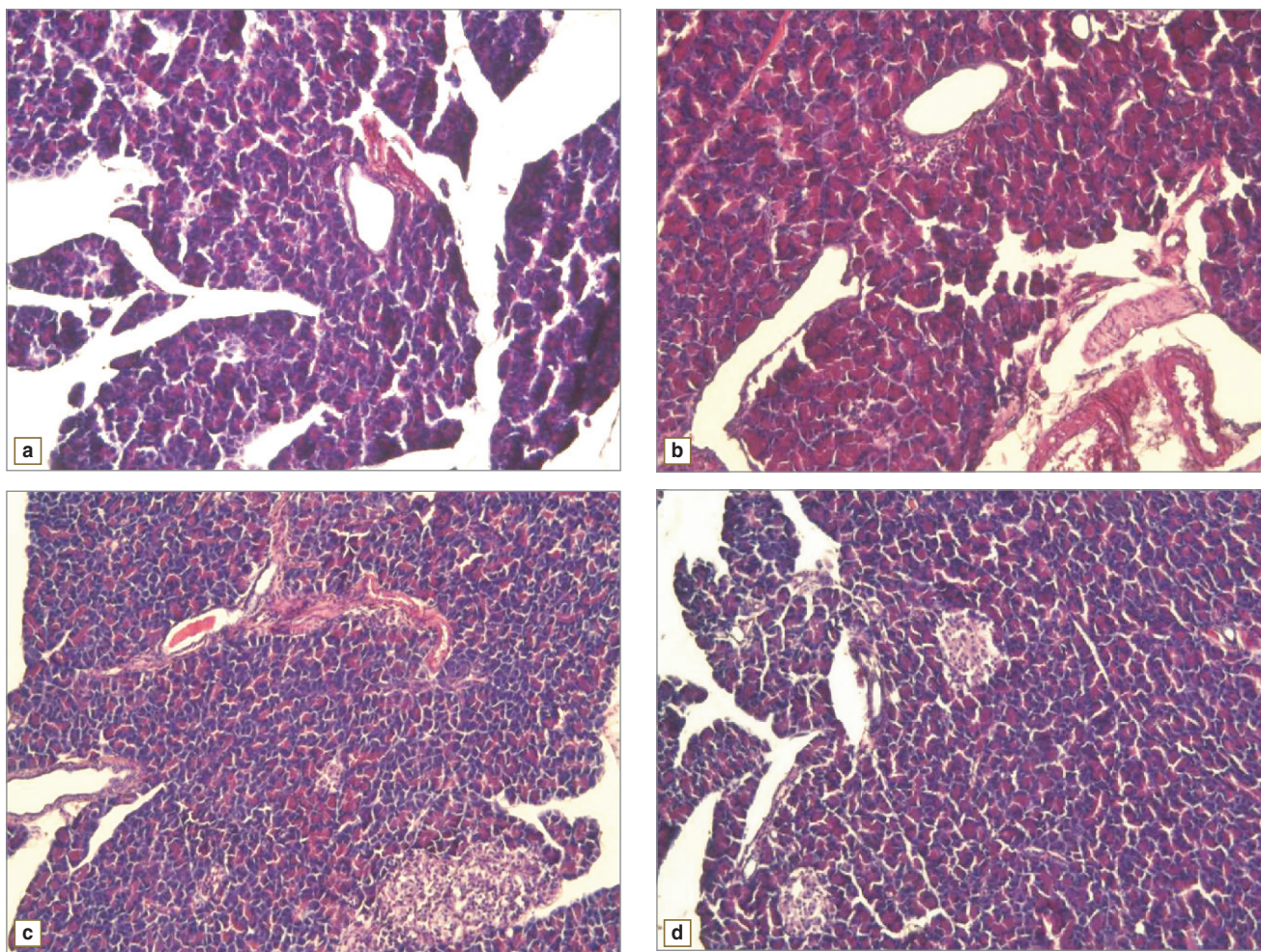
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was reinfused in the volume of 70% of the blood loss. The animals were removed from the experiment in 24 and 72 h after the blood loss. Before the light optical microscopy of tissue histologic specimen were fixed in 10% neutral formalin 72–96 hours, dehydrated in spirits of ascending concentration and embedded into paraffin. 7 μm thick sections were produced on a Leica SM 2000R (Germany) microtome and stained with hematoxylin and eosin. The view of histologic specimen was performed with the microscope Leica DMLS (Germany). Photomicrographs were obtained by video-based computer with a Pentium III CCD camera. Assessment of studied biochemical parameters (glucose, lactate, LDH, amylase) was carried out with biochemical analyzer Konelab 60i (Finland) using Thermo scientific reagents (Finland). Statistical processing of the results was performed using the programs Microsoft Excel and Statistica 6.0 following the criteria of nonparametric statistics, using the criterion of inter-group comparison, Kruskal–Wallis ANOVA.

Results. Morphological studies of the pancreas conducted a day after the removal of rats from hemorrhagic shock in animals treated with Ringer's solution, showed the presence of edema of the stroma, pericellular edema

in the islets of Langerhans, the expansion of the ducts of exocrine glands (See Fig., a). Using malate-containing Sterofundin isotonic solution as a single agent of infusion therapy significantly reduced the edema of the stroma of the pancreas in the absence of pericellular edema in the islets of Langerhans and perivascular edema (See Fig., b). Consequently, blood malate-containing substitutes contributed prevention of early reperfusion injuries of the pancreas. On the third day of post-hemorrhagic period in animals of the control series the morphological changes of the pancreas increased progressively (See Fig., c). Defined stroma and capsule edema, perivascular edema, pericellular edema in the islets of Langerhans, the expansion of the ducts of exocrine glands were detected. Most vessels contain only plasmatic components indicating the microcirculatory disorders in the pancreas.

In the test series the increase in stromal edema of the pancreas was observed, perivascular edema, pericellular edema was observed in the islets of Langerhans (See Fig., d). The blood stagnation was noticed in the vascular bed. The increase in the severity of the morphological changes in the test and to in the control series are primarily due to the late reperfusion changes. But this



Structure of pancreas: a — Ringer solution, day 1; b — Ringer solution, day 3; c — Sterofundin isotonic solution, day 1; d — Sterofundin isotonic solution, day 3. Stained with hematoxylin and eosin; a, c, d — ×100; b — ×40

The change of biochemical parameters in the blood serum of rats after acute massive blood loss (M±SD)

Observation period	Glucose	Lactate	Glucose/lactate	LDH	Amylase	Ca ²⁺
Ringer solution						
Day 1	8.47±1.53	3.92±0.52	2.18±0.32	752.1±128.8	1378.1±262.5	1.10±0.05
Day 3	7.83±1.01	3.10±0.38	2.48±0.20	792.2±156.8	885.0±240.5*	0.56±0.10
Sterofundin isotonic						
Day 1	7.34±1.89	3.39±0.48	2.16±0.40	484.7±95.4##	804.0±149.7##	1.22±0.02
Day 3	6.35±0.73*#	2.31±0.35*##	2.82±0.28*#	580.2±104.0#	508.5±151.1***	0.82±0.08

Note: statistically significant difference of values with the first 24 h. * — p<0.05, ** — p<0.01; # — p<0.05, ## — p<0.01 — with the control group.

does not exclude the influence of the pancreas of other organs of the gastrointestinal tract involved in multiple organ dysfunction, including bowel and liver. [7]

The analysis of the results of basic laboratory indicators characterizing ischemic and subsequent reperfusion injury of the pancreas [8, 9] (see the Table), showed that in the early posthemorrhagic period, all animals showed marked changes in carbohydrate metabolism, hyperglycemia, hyperlactatemia, decrease of index “glucose/lactate”. They regressed significantly more rapidly in animals of experimental series, and that confirms antihypoxic activity of malate, being contained Sterofundin isotonic. The rate of decline of hyperlactatemia was higher than those of other metabolic parameters. Presented in animals of the control series hyperglycemia on day 3 was the reflection of not only post aggressive insulin resistance, but also of the endocrine dysfunction of the pancreas, confirmed by the morphological picture of the islets of Langerhans.

High indexes of enzymemia in animals of the control group within posthemorrhagic period show the degree of necrotic damage of internal organs cells. Hyperamylasemia along with hypocalcaemia confirmed the special vulnerability of the pancreas to ischemic injury, and lactate dehydrogenase indexes persisting for three days were greater than 750 units, and in addition it proved the high probability of the development of destructive forms of acute pancreatitis. Postischemic hyperenzymemia in animals of the experimental group was significantly lower due to cytoprotective and membrane protective properties of exogenously administered malate by supporting energy production in the citric acid cycle and malate-aspartate shunt. The known association of metabolism of malate and succinate does not preclude correction of energy shortage by increasing the bioavailability of succinate cells [10–12]. Reducing the severity of cytolytic syndrome confirms the importance of the substrate support mechanisms malate-dependent mechanisms of organ-protection with Sterofundin isotonic.

Reducing the severity of morphological and laboratory manifestations of acute pancreatitis using Sterofundin isotonic we associate with recovery oxygen-dependent bioenergetic processes in the cell along with the delayed complex protective action of malate on adaptation mechanisms after AMH [13–14].

Conclusion. Malate-containing blood substitute Sterofundin isotonic, used in the early stages of the experimental treatment of acute massive blood loss, and has a preventive effect on the development of pancreatic injuries in the early and delayed posthemorrhagic periods.

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