British Journal of Medicine & Medical Research
13(10): 1-6, 2016, Article no.BJMMR.23197
ISSN: 2231-0614, NLM ID: 101570965

SCIENCEDOMAIN *international*
www.sciencedomain.org

Structure of the Small Intestinal Mucosa after Acute Hemorrhagic Shock and Reperfusion of the Ischemic Limbs

Rakhmatova Mukaddas Kholtaevna[1*], Tarinova Margarita Vladimirovna[1], Nugmanova Umida Takhirovna[1] and Nishanova Aziza Abdurashidovna[2]

[1]Department of Histology and Medical Biology, Tashkent State Institute of Dentistry, Tashkent, Uzbekistan.
[2]Department of Normal Physiology, Tashkent Medical Academy, Tashkent, Uzbekistan.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJMMR/2016/23197
Editor(s):
(1) Ashish Anand, Department of Orthopaedic Surgery, GV Montgomery Veteran Affairs Medical Center, Jackson, MS, USA.
Reviewers:
(1) Somchai Amornyotin, Mahidol University, Bangkok, Thailand.
(2) Ketan Vagholkar, D.Y. Patil University, India.
Complete Peer review History: http://sciencedomain.org/review-history/13295

Short Research Article

Received 19th November 2015
Accepted 7th December 2015
Published 14th February 2016

ABSTRACT

Aims: The purpose of the report is to study Structure of the Small Intestinal Mucosa after Acute Hemorrhagic Shock and Reperfusion of the Ischemic Limbs.
Place and Duration of Study: The Department of Histology and Medical Biology, Tashkent State Institute of Dentistry, between March 2013 and March 2015.
Methodology and Study Design: Acute hemorrhagic shock was developed by using Wigger modified method. 76 Shinshilla rabbits having 2.6 + 0.2 kg of body weight fasted for 15 hours before they were undergone acute hemorrhagic shock. In one hour the clamps were removed and simultaneously with the blood flow restoration the ischemic limbs were injected with physiologic solution (II), Rheopolyglukin (III; 20 mg/kg), Succinasol (IV; 1.2 mg/kg). The functional system of digestion and absorbtion integrated with the immune system of the small intestional mucosa which normally regulates homeostasis of the inner medium of an organism was defected one hour after reperfusion of the ischemic limbs of rabbits.
Results: The basement membrane beneath enterocytes had usual thickness, stroma of villi contained large number of lymphocytes, functionally active eosinophils and mast cells, the

**Corresponding author: E-mail: science@eline.uz;*

plasmatic cells were large in size and contained rough endoplasmic network with dilated profiles. Blood vessels had normal structure.

Conclusion: Structural and functional lesions of the supraepithelial layer of mucus in small intestinal mucosa at the acute hemorrhagic shock and reperfusion have led to direct interactions of microorganisms and microvilli of enterocytes, their translocation into the absorbing cells and stroma of villi, phagocytosis and depletion by macrophages, activation of immunocytes and other cells of the connective tissue. The results indicated the Succinasol had prevented effectively the structural and functional lesions of the small intestinal mucosa and, as a consequence, the interactions of intestinal microorganisms with glycocalix and microvilli and their translocation into the inner medium.

Keywords: Small intestine; antigen; hemorrhagic shock; ischemic limb.

1. INTRODUCTION

It is known that in the small intestinal mucosa which is localized at the edge of outer and inner media of an organism, contains a lot of microorganisms and their antigens, in-takes nutrients of non-predictive volume and property with microorganisms and their antigens there was developed perfect and adaptive functional system providing homeostasis. Owing to it, the processes of digestion and absorption of nutrients, prevention of microorganisms and their antigens interactions and passage in are optimal at the normal physiologic conditions [1,2,3,4].

However, in some somatic diseases, shock, traumatic surgical interference, the barrier-protective and homeostatic functions of the small intestinal mucosa, integrations of its immune and digestion-absorption functions may be impaired [1,4,5,6,7]. Having considered all the above mentioned, this investigation was undertaken to study in dynamics the structure of small intestinal mucosa after hemorrhagic shock and reperfusion of the ischemic limb and injection of antihypoxant Succinasol.

2. MATERIALS AND METHODS

Acute hemorrhagic shock was developed by using Wigger modified method [8]. 76 Shinshilla rabbits having 2.6 + 0.2 kg of body weight fasted for 15 hours before they were undergone acute hemorrhagic shock. Course of the experiment: the animals were fixed to a special device and for local anesthesia 0.5% solution of Novocain was injected in the inguinal area. The right femoral artery after surgical isolation was cannulated with a system of silicone tubes filled with physiologic solution. Fractional blood letting (2.4 + 0.1% to body weight of an animal) was made portion by portion every 15 minutes by scheme: 0.4; 0.3; 0.2; 0.1 parts of its summed up volume. Hemorrhagic shock was considered to be developed from the period when the arterial pressure fell to 40 mm. of Mercuric column. Then a clamp was adjusted below the cannulated area of the femoral artery. In the first group of animals without shock effect clamps on the artery were adjusted too. Influence of hemorrhagic shock and reperfusion on the ischemic limb was studied one hour after the arterial pressure became 40 mm. of mercuric column. In one hour the clamps were removed and simultaneously with the blood flow restoration the ischemic limbs were injected with physiologic solution (II), Rheopolyglukin (III; 20 mg/kg), Succinasol (IV; 1.2 mg/kg). Animals of the first and second groups served as controls. Morphology of the jejunum (5 cm below the Treytsligament) and ileum (5 cm before the end of the ileum) were studied 1 and 24 hours after the restoration of blood flow and administration of the solutions.

The specimens were obtained after the animals were given narcosis and incision on the anterior wall of peritoneum along the abdominal white line was made. The specimens were fixed in 12% neutral formalin or in 2.5% buffered Glutaraldehyde (20 min) and post fixed in 1% OsO4 solution (1.5 hours). After the appropriate processing the specimens were imbedded in paraffin or araldite. Cuts for light microscopy were stained with hemotoxilin and eosin, and for the electron microscopy cuts were contrasted in Uranyl acetate and Lead citrate. The cuts were examined under MBI-15 and JEM-100S.

3. RESULTS

Mesentery and intestine were more pale in the experimental animals than that in controls (I group) one hour after the hemorrhagic shock and blood flow restoration. The mesenteric vessels running from its root towards the intestine were collapsed. Small intestine was slightly swollen and contained gas and chymus with mucus in some parts. Visible lesions in villi and crypts of jejunum and ileum were not revealed under a light microscope. But a large number of goblet-like and Panneth cells did not almost contain the secretory material, the supra epithelial layer of mucus showed uneven thickness, capillaries in villi stroma were spastic. Under electron microscope the supra epithelial layer was found to be polymorphic, had fibrillar structure and contained, besides extruded cells, microorganisms. The hydro-electrolyte layer, glycocalix and microvilli of the brush-bordered enterocytes in villi did not show visible alterations. Cytoplasm of the absorbing cells was heterogenic: in some cells it was dense in other ones it was light. Microvilli in the former cells had typical structure, contained a lot of endocytotic vesicles between their bases. The Golgi complex located in the supranuclear cytoplasm was hypertrophied, mitochondria were elongated and contained matrix of moderate density with moderate number of crysts. In light enterocytes microvilli were partly or fully vesicular, the quantity of ribosomes and polysomes was less, matrix of mitochondria was light and crysts were reduced. Between enterocytes having such ultrastructure or at their bases above the basement membrane there were found, as a rule, lymphocytes and rarely other leucocytes. If rabbits of the first group contained solitary lymphocytes between epithelial cells, one hour after shock and reperfusion of ischemic limbs their number between enterocytes increased and neutrophils, eosinophils, macrophages and rarely mast cells were found too. In some areas the interepithelial spaces had conic-shaped dilatations, being the result of edema. In lumens of some crypts solitary microorganisms were noted. The Panneth cells located at their bottoms contained small number of secretory granules or were in the phase of secretion. Tunica propria contained increased number of macrophages with polymorphic lysosomes, monocytes and lymphocytes, mast and eosinophilic cells were degranulated. Nerve endings among cells of the connective tissue and near capillaries were clarified, did not contain secretory granules; had solitary mitochodria had clarified matrix and reduced number of crysts. Majority of capillaries were spasmic and lined by low cubic shaped endothelium containing numerous endocytotic vesicles. In some areas of villi stroma capillaries were dialated unevenly and contained leukocytes in lumens.

24 hours after modelling acute hemorrhagic shock and reperfusion of the ischemic limb the small intestinal epithelium was heterogenic. If some areas had normal structures, the other ones had clearly seen signs of structural lesions in mucosa. On the surface of villi epithelium the supraepithelial layer was either absent or significantly thinner Solitary or groups of microorganisms in the intestine interacted with destructively altered microvilli or were revealed in the supranuclear cytoplasm of enterocytes. Microvilli on the surface of some enterocytes in the villi were contracted or at the state of vesiculation. Such cells were often extruded into the intestinal lumen at any part of villi surface.

Stroma of villi contained relatively more macrophages with polymorphic lysosomes, monocytes and lymphocytes. Mast and eosinophilic cells were often interacting and contained small quantity of secretory granules. Profiles of granular endoplasmic reticulum were dilated in plasmatic cells. Nerve endings did not contain secretory granules, single mitochondria were light and contained solitary crypts. Structure of capillary net in the villi stroma was heterogenic: if some of them were lined with flattened endothelium, the others with that of cubic-shaped one.

Rheopolyglukin being injected to animals with modulated acute hemorrhagic shock, right after the clamp was removed from the femoral artery did not improved essentially the structure of small intestinal mucosa described above. Twenty four hours after rheopolyglukin was administered to the experimental animals the structure of mucosa was almost restored to normal state. Infiltration of the epithelial layer with leukocytes preserved, the secretion process was enhanced in Panneth and goblet-like cells. The supra epithelial layer became thinned or was absent in some places. If it was preserved there were found microorganisms and fragments of extruded cells. Microvilli having altered or preserved structures interacted rarely with microorganisms. Microorganisms were revealed within lysosomes in the cytoplasm of enterocytes but they were very rarely detected. Epithelium was infiltrated mainly with lymphocytes, but mast and eosinophilic cells were found rarely. The basement membrane beneath enterocytes of villi was thickened. Quantity of the connective tissue cells in villi stroma became more numerous. Mast and eosinophilic cells were functionally active and contained various number of secretory granules. The nerve endings in main mass did not contain secretory granules. Blood capillaries located beneath epithelium were lined with flattened endothelium and contained moderate number of endocytotic vesicles along their both surfaces.

One hour after intravenous injection of Succinasol the structure of the small intestinal mucosa almost did not show signs of injury. On the surface of villi the mucus layer, glycocalix, microvilli and cytoplasmic structures did not show visible lesions. Solitary brush-bordered enterocytes were only light and contained less organelles, polysomes and ribosomes. Goblet-like cells was found to be at various functional states: some of them were degranulated, others contained moderate or significant volume of secretion. In lumens of some crypts there were revealed solitary macrophages, Pannett cells were functionally active and contained small number of secretory granules in the supra nuclear zone.

Number of lymphocytes located between the villi enterocytes was significantly less then in the animals of other groups; other types of leukocytes were almost not detected. The basement membrane beneath enterocytes was almost unchanged. Blood capillaries in villi stroma like that of in group I, were not altered and were lined with flattened endothelium. Moderate quantity of endocytotic vesicles was seen along their plasmalemma. Plasmatic, eosinophilic and mast cells showed moderate activity, macrophages were of large size and contained polymorphic lysosomes. Nerve endings without secretory granules were revealed near capillaries and cells in villi stroma.

24 hours after Succinasol injection to animals with acute hemorrhagic shock and reperfusion there were not noted detectible lesions in the structure of small intestinal mucosa. Mucus layer on the surface of villi enterocytes, glycocalix, microvilli and cytoplasmic structures were not altered. Goblet-like cells like that of in normal animals were heterogenic. The Panneth cells were of moderate activity, lumens of crypts did not contain microorganisms. The epithelial layer was still infiltrated with lymphocytes and rarely with other types of leukocytes. The basement membrane beneath enterocytes had usual thickness, stroma of villi contained large number of lymphocytes, functionally active eosinophils and mast cells, the plasmatic cells were large in size and contained rough endoplasmic network with dilated profiles. Nerve endings contained solitary light and dark secretory granules. Blood vessels had normal structure.

4. DISCUSSION

According to investigations of I.M.Baybekov et al. [1], Yu. M. Galperin and P. I. Lazarev [3], A. Yu. ldashev et al. [4], I. A. Morozov [9], I. A. Morozov et al. [10], K. A. Zufarov and A. Yu. Yuldashev [11], Mazo V. K. et al. [12] a small intestine at normal physiologic conditions on various levels – in the intestinal lumen, supraepithelial layer of mucus, hydro-electrolyte layer, glycocalix and plasmalemma of microvilli – provides optimal digestion of biopolymers and absorption of monomers in spite of the volume and the property of meals and microorganisms contained in it. Such highly perfect processes are possible owing to close integration of structure and function at various levels, high concentration of pancreatic and enteral enzymes and secretory immune globulin A (sIgA). Besides, the exclusive property of the paramembranous layer (glycocalix of the brush-bordered enterocytes and hydro-electrolytic layer) is in its sterility at physiologic conditions being provided by barrier-protective ability of the supraepithelial layer of mucus (high concentration of pancreatic and enteral enzymes and sIgA). Sterility of crypt lumens is provided owing to constant secretion of lysozyme by Panneth cells localized at their bottoms Owing to it, the processes of enterocytes proliferation and differentiation in crypts, their migration to the surface of villi and extrusion into the intestinal lumen in proper time take place relatively autonomic, as well.

High rate of enterocytes renewal in the crypt-villi system needs in constant blood supply containing nutrients and energy. Oxygen is necessary for the optimal utilization and providing intensive processes taking place in the small intestinal mucosa. If oxygen and nutrients supply is disturbed, for instance, at the acute hemorrhagic shock and reperfusion of ischemic limb there was noted disintegration in functioning of digestion-absorption and immune processes in small intestinal mucosa. According to our findings shock and reperfusion affected the supraepithelial layer of mucus and other barriers integrating with it even in the absence of factors having direct influence on the small intestinal mucosa. That is why, direct interactions of intestinal microorganisms with glycocalix and plasmalemma of enterocyte microvilli, their transition into cytoplasm and stroma of small intestinal villi were revealed. By the opinion of T. S. Popova et al. [13], in cases of trauma or shock the common mechanisms were operating and resulted in the development of syndrome of intestinal insufficiency characterized by lesions of all functions of the organ. At the first stage there took place dysbalance in sympathetic and parasympathetic innervation (degranulation of nerve endings), motility, dyscoordination of digestion, absorption and secretion processes. Lesions in microcirculation in small intestinal mucosa at acute hemorrhagic shock and reperfusion, lack of nutrients and energy had led to acceleration of villi enterocytes wearing out, enhancement of secretion processes in goblet-like, mast and eosinophilic cells. In consequence with these and other processes, as well as translocation of as epithelial layer with leukocytes were mobilized.

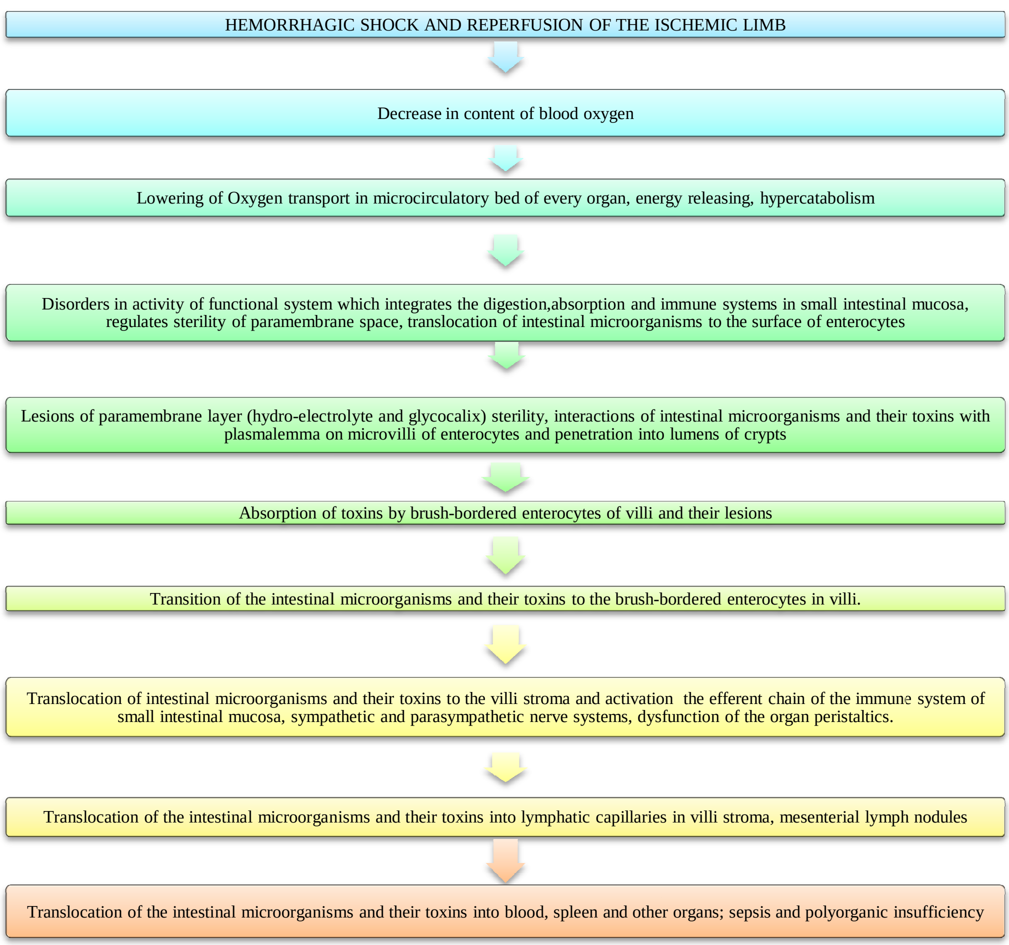
Translocation of microbes and their toxins via the affected structures of the intestinal mucosa might be one of causes leading to development of polyorganic insufficiency and sepsis syndrome.

Taking into account that insufficiency in oxygen, nutrients for digestion and absorption and energy might be considered as the basic factors resulting in development of lesions in the structure and function of small intestinal mucosa, the native antihypoxant Succinasol containing succinic acid had been administered for correction. It helped, in certain degree, not only to increase the circulating blood volume, but joining in Krebs cycle [8] also to become the source for energy releasing in intestine and other organs. By our findings and that of other investigators [14], Succinasol was revealed to prevent development of ischemic lesions of organs. One hour after acute hemorrhagic shock development and reperfusion of the ischemic limb Succinasol, besides prevention of structural and functional lesions in the mucosa, has regulated interactions of microorganisms with glycocalix and enterocyte plasmalemma, their translocation into cells and villi stroma.

5. CONCLUSION

1. Structural and functional lesions of the supraepithelial layer of mucus in the small intestinal mucosa at the acute hemorrhagic shock and reperfusion have led to direct

Pathogenesis of sepsis and polyorganic insufficiency development at acute hemorrhagic shock and reperfusion of the ischemic limb



interactions of microorganisms with microvilli of enterocytes, their translocation into the absorbing cells and stroma of villi, phagocytosis and depletion by macrophages, activation of immunocytes and other cells of the connective tissue.

2. Succinasol has prevented effectively structural and functional lesions of the small intestinal mucosa and, as a consequence, the interactions of intestinal microorganisms with glycocalix and microvilli and their translocation into the inner medium.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Baybekov IM, Khodjibaev AM, Kasimova Kh. Structural bases of indications and consequences of vagotomy. Tashkent, M. Abu Ali Ibn Sino. 1992;272.
2. Valker VA. Role of microflora in the development of protective functions of intestine. J. Pediatrics. 2005;1:25-31.
3. GalperinYu M, Lazarev PI. Digestion and Homeostasis, M. Science. 1986;304.
4. Yuldashev A. Yu, Rakhmanov RR, Nishanova AA. Mechanisms for regulation of homeostasis in protein absorption from a small intestine into blood. Medical Journal of Uzbekistan. 2004;5:79-87.
5. Akhmedova Kh Yu, Gulyamova NG, Akhmedova MD. Conception on clinic immunology and immune correction at Salmonollosis. Tashkent; 2008.
6. Petukhov AB, Lisikov Yu. A, Ishkova V. Yu. Digestive function of the parietal layer of mucus at the digestive organs pathology. M. Nutrition. 2004;6:21-24.
7. Khavkin AI. Microflora of the digestive tract M. Fund of Social Pediatrics. 2006;480.
8. Levin GS. Bioenergic processes in blood loss and shock. Tashkent, M. Abu Ali Ibn Sino. 1991;231.
9. Morozov IA. Structure and function of the mucus layer of mucosa. M. Medicine. 1998;228.
10. Morozov IA, Lisikov Yu. A, Pitran VV, Khbilya SI. Absorption and secretion in a small intestine (submicroscopic aspects). M. Medicine. 1988;224.
11. Zufarov KA, Yuldasheva Yu. Small Intestine. Handbook on Histology. St. P. 2002;115–140.
12. Mazo VK, Gmoshinsky IB, Sablina VV, Zorin SN. Protein antigens permeability via a small intestinal wall in human beings and experimental animals. M. Nutrition. 2008; 2:10-22.
13. Popova TS, Shestopalov AE, Tamasashvili T. Sh, Leyderman IN. Support with nutrients of patients being at crucial states.
14. Shneyvays VB, Drullea Ya, Dregeris Ya. Ya., Levin GS. Influence of Nitrogen containing derivatives of 1,4-Naftichinon on processes of peroxydation in mitochondria. Problems of Medical Chemistry. 1992; 6:37-41.

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/13295