HISTOLOGICAL AND HISTOCHEMICAL CHANGES IN THE MYOCARDIUM IN SEA AND FRESH WATER DROWNING (STUDIED ON CADAVERIC MATERIAL AND TEST ANIMALS)

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Microscopic changes in the myocardium in case of drowning, as compared to changes in the lungs, are scarcely investigated (F. I. Shkaravskii, 1951; M. I. Kasianov, 1954; Y. S. Smusin, 1958; M. L. Kazarnovskaia, 1968; M. Mosinger et al, 1961; L. Ambrosi, H. Dell Erba, 1966 and others). Also, there is a great paucity of literature data concerning histochemical changes in the myocardium in drowning (Y. S. Smusin, 1958; M. L. Kazarnovskaia, 1968). Literature references are available dealing in general with microscopic changes in drowning, without giving an account and differentiating the changes observed depending on the salt composition of the water where drowning took place.

In the present work we undertook the task to study histological and histochemical changes in the myocardium in drowning with a special reference to forensic medical diagnosis, as well as the possibility to differentiate drowning in fresh from drowning in sea water on the basis of such changes.

Material and method

Investigations are made on 169 cadavers of drowned persons, of which 148 — in sea, and 21 — in fresh water, as well as on 192 test animals (guinea pigs, rats and rabbits), drowning in sea and fresh water in different ways (slowly — during free swimming, and quickly — after beforehand fixation)' and in varying water temperatures.

Apart from the histological methods (with hematoxylin after van Gison, Croutchay), staining after Weigert for elastic fibers, staining after Gomori for argyrophilic fibers, histochemical methods were also used to demonstrate glycogen through PAS-reaction after MacManus (at control salivary amylase) and Best, metachromasia, acid mucopolysaccharides with toluidine blue at pH 2, 4, 5, with alcian blue 8_{GS}, as well as to determine fuchsinophilic degeneration after the method of Selye, and changes in DNA and RNA after Feulgen and Brachet.

Results

In sea and fresh water drowning, histomorphologically in the myocardium are observed: acute impairment of circulation, changes in the vascular walls, in the formed elements of blood, dystrophic changes in myofibrils and edema of the interstitium. In fresh water drowning the changes in the formed elements of blood are heavier, and are manifested with alterations in the form (deletion of contours, disruption and hemolysis) and colour (fading).

The acute impairment of circulation is manifested with uneven filling of vessels, dilatation and stasis of capillaries, venules and arterioles, somewhere accompanied by tiny hemorrhages (usually marked in seawater drowning). In many points the endothelium is swollen, partially desquamated, with cells exhibiting elongated or rounded shape; the nuclei are clearly delineated and pyknotic. Here and there, particularly in intramural vessels, projections (horseshoe, pillow-like etc) of the intima are observed; the latter is thickened and loosened owing to impregnation of the vascular wall with a pale-pink material (hydremic plasma). The listed changes are more clearcut in seawater drowning in comparison with drowning in fresh water, and are corroborated by experimental studies on animals. In case of drowning in fresh water, at many points the vessels are partially filled with destroyed erythrocytes or they are virtually empty. Here, pale contours or portions of erythrocytes are detectable in many of the vessels.

Not infrequently, the muscle fibers in seawater drowning show fragmentation (in 53 per cent of the cases under examination); often they are thickened, swollen, homogenized with blurred transverse striation (in 81 per cent of the cases). Also, diffuse tiny granulation is observed in the myofibers. Granules stained with hematoxylin eosin show bright pink tinge.

The muscle fibers acquire the property to stain readily with acid dyes, i. e. the so-called fuchsinophilic dystrophy is observed. The latter is particularly clearcut upon staining after the method of H. Seyle, where the fibers instead of methyl green begin to take in acid fuchsin. Such a fuchsinophilla is noted in isolated areas of the myofibers — in the myofibrils and in isolated zones of the sarcoplasm (beneath the sarcolemma), and rather frequently in the region of intercalary disks. Usually, fuchsinophilic degeneration is observed in the left ventricle, involving whole fibers and individual bundles (Fig. 1) The comparative study shows that in seawater drowning fuchsinophilic degeneration in the muscle fibers of the myocardium is more diffuse and more strongly manifested than in fresh water drowning. Fuchsinophilic degeneration was variably manifested in the different groups of animals: optimally in those drowned in sea water, less pronounced — in slowly drowned animals in fresh water, and very slightly — in rapidly drowned test animals, regardless of the salt composition of the water.

Appearance of PAS-positive substances (persisting upon amylase treatment) and pyroninophilia (pink stain in the reaction of Brachet not disappearing under the effect of ribonuclease) is observed in the sarcoplasm of muscle fibers. Pyroninophilia is recorded in the zones where PAS-positive substances occur.

Usually, the nuclei of the myofibers remain unchanged. In isolated cases they are enlarged and variably stained, but anyway, with no changes whatsoever in DNA.

In the myocardial fibers a reduction and loss of glycogen is established (through PAS reaction after MacManus and carmine after Best). Most frequently, the granular glycogen situated around the nuclei within the anisotropic disks, and in the sarcoplasm beneath the sarcolemma, disappears. Somewhere glycogen disappears in whole muscle fibers. The reduction and loss of glycogen is ununiform, and conditions the motleyed pattern of "glycogen dystrophy". Glycogen disappearance is more diffuse in seawater drowning, in comparison with fresh water.

The glycogen in the myocardium of test animals shows the following changes: well manifested (+++) in animals killed through mechanical injury to the head, reduced (++) in rapid drowning, and virtually fully absent (+) in slowly drowned animals, regardless of the salt composition of water.

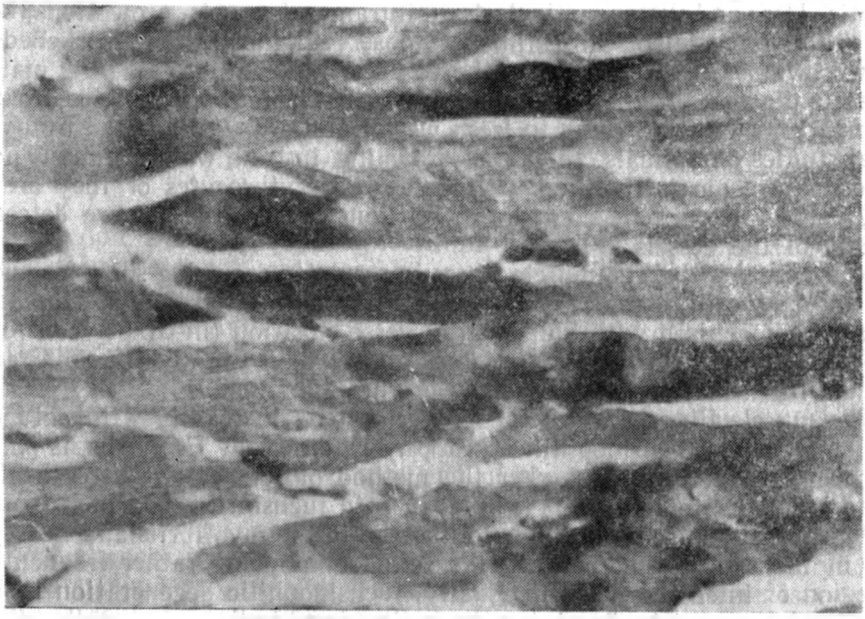

Fig. 1: Fuchsinophilic degeneration of myocardium muscle fibers in seawater drowning.

In all expert examination cases, and in the test animals as well, the glycogen reduction is more pronounced in the muscle fibers of the left ventricle, as compared to the right one.

At the sites of pronounced interstitium edema, and at the points of collagen fibers' loosening substances yielding weak metachromasia (orthochromasia) with toluidine blue and alcian blue staining are accumulated, i. e. acid mucopolysaccharides in insignificant amounts are established. Within the interstitium a roughening and thickening of argyrophilic fibers, as well as heavy impregnation with silver salts (argyrophilia), is observed upon staining after Gomori.

Discussion

In drowning the hypoxia and especially electrolyte changes in the blood exert effect also on the myocardium which is morphologically manifested with histological changes in this particular organ. Some of the latter point to an acute impairment of circulation — sudden blood filling of the vessels, particularly smaller ones, with zones of paretic expansion and stasis of capillaries and arterioles, adjacent to emptied and convoluted vessels, isolated focal hemorrhages, acute plasmatic imbibition of the intima of arteries with

varying caliber and perivascular edema. Others point to necrobiotic changes — uneven staining of the muscle fibers, ochsinophilia and homogenization, disappearance of transverse striation and fragmentation. In practically all cases an unevenly expressed edema of the interstitium is observed. These findings of ours are in agreement with the results reported by Y. S. Smusin and M. L. Kazarnovskaia.

Fuchsinophilic degeneration was more strongly manifested in drowning in sea water, and more clearcut in the muscle fibers of the left ventricle which, in our opinion, is due not so much to electrolyte changes, but rather to the prolonged process of drowning in sea water. In the experiment, it was better pronounced in slowly drowned animals. The data submitted point to the existence of interdependence between hypoxia continuity, occurrence and degree of fuchsinophilic degeneration manifestation.

Our study shows that in drowning, first and foremost the free and more labile granular glycogen is utilized, usually situated perinucleary, whilst glycogen from the sarcoplasm and anisotropic disks is exhausted at a later stage. The degree of "glycogen dystrophy" in the myocardium similarly depends on the duration of hypoxia, respectively on the length of drowning period (a more complete loss in seawater drowning, and in slowly drowned animals).

Changes in the myocardium in drowning resemble to some degree those occurring in death from acute coronary insufficiency — subsequet to functional impairment of the coronary circulation and initial infarction of the myocardium. Unlike infarction changes, they exhibit a diffuse character and involve mainly the muscle fibers subjected to the heaviest functional load.

Conclusions

1. In drowning an acute impairment of circulation is observed in the myocardium, being rather strongly manifested in seawater drowning.

2. The necrobiotic changes are manifested with modification of the tinctorial properties of muscle fibers (fuchsinophilic degeneration, occurence of PAS-positive substances, persisting upon amylase treatment and pyroninophilia). These changes are variably manifested in seawater and fresh water drowning, and depend on the duration of drowning.

3. The reduction and disappearance of granular glycogen is variably manifested in sea and fresh water drowning. The latter difference is mainly conditioned by the continuity of asphyxia.

4. The histological and histochemical changes observed in the myocardium after drowning, although non-specific, contribute to establish forensic medical diagnosis for the particular type of death, and indicate whether or not drowning occurred in sea or fresh water.

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ГИСТОЛОГИЧЕСКИЕ И ГИСТОХИМИЧЕСКИЕ ИЗМЕНЕНИЯ В МИОКАРДЕ ПРИ УТОПЛЕНИИ В МОРСКОЙ И ПРЕСНОЙ ВОДЕ (ПО ДАННЫМ ЭКСПЕРТНОГО ТРУПНОГО МАТЕРИАЛ И ОПЫТНЫХ ЖИВОТНЫХ)

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РЕЗЮМЕ

В работе представлены результаты гистологических и гистохимических исследований миокарда 169 трупов утонувших (148 в морской воде и 21 в пресной) и 192 опытных животных (утопленных в морской и пресной воде разными способами и при различной температуре воды).

Обнаружено: острое нарушение кровообращения; некробиотические изменения, выражающиеся в измении тинкториальных свойств миофибр (фуксинофильная дегенерация, появление ПАС-положительных веществ, неисчезающих при обработке амилазой, и пиронинофилия); уменьшение количества и исчезновение гранулированного гликогена. Эти изменения выражены сильнее у утонувших в морской воде и у животных, которых топили медленно, т. е. они находятся в зависимости от длительности асфиксии.

Гистологические и гистохимические изменения в миокарде у утонувших, хотя и не специфические для этого вида смерти, могут быть использованы в общем комплексе морфологических признаков при судебномедицинском диагнозе утопания и для указания, произошло ли утопание в морской или пресной воде.